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Wild relatives of fruit trees in Syria: Genetic resources threatened by conflict

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Abstract: Wild relatives of fruit trees (WRFT) are highly valued for food and tradable products by rural communities, especially in low-income countries and as such are a vital resource for tree improvement. During periods of conflict, a lack of support and protection by national authorities may make WRFT vulnerable. In Syria, WRFT are at risk of extinction due to the ongoing crisis, which has limited efforts to conserve and propagate these unique genetic resources. We collected information about the current status and key threats to WRFT in northwest Syria from 50 agricultural experts using structured interviews. Our results show that many sites have experienced erosion, overgrazing and drought. To initiate *in situ* and *ex situ* conservation, the locations of WRFT exposed to deterioration were identified and mapped using GPS, and seeds from five genotypes per species were collected from each of ten species to be used for the establishment of an *in situ* and *ex situ* WRFT collection.

Keywords: Conservation of wild fruit plants, In situ conservation, Plant Genetic Resources, Genetic Erosion, Crop wild relatives

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Introduction

Ensuring future global food security is one of the biggest challenges facing humans today. One in nine people worldwide already suffers from poor access to food resources (FAO, IFAD and WFP, 2015), and with predicted human population growth, pressure on food resources is likely to increase dramatically (Godfray *et al*, 2010; UN DESA, 2015). New high-yielding varieties of crops that can withstand adverse climatic conditions and new pests and diseases are urgently needed (McCouch *et al*, 2013). All cultivated crops derive from historic human domestication of wild genotypes, which have been gradually selected to produce commercial varieties (Engels and Thormann, 2020). Crop wild relatives are the wild original sources of these modern cultivars and remain the primary resource of genetic diversity for breeding new, climate-change tolerant, and higher-yielding cultivars (Maxted and Kell, 2009; Vincent *et al*, 2019). As such, it is vital to ensure wild relatives are identified, protected and propagated to help secure the future of the species we depend on.

Globally, fruit trees are of enormous economic importance. In many places (Erciŝli *et al*, 2009; Otieno, 2017), the wild relatives of fruit trees (WRFT) remain essential sources of food and medicines (Symphorien *et al*, 2016) but also provide feed for domestic animals and food additives. They also play a crucial

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role in maintaining ecological diversity (Musayev and Huseynova, 2016). Notably, some of these wild relatives are known to possess traits that could greatly benefit contemporary agriculture through the breeding of new, improved cultivars. For example, wild genotypes of species in the genus Prunus are characterized by adaptation to extreme climatic conditions (Uğur and Gündeşli, 2020). Moreover, wild fruit tree genotypes are an essential source of resilient rootstock, as grafting is one of the key methods for making new fruit varieties available to growers (Zhebentyayeva *et al*, 2019). Several wild edible fruits are used by rural and tribal populations and significantly contribute to their livelihood. The use of non-cultivated foods, of which wild fruits form a part, as a dietary supplement or as a coping mechanism in times of food shortage, provides an important safety net for the rural poor, especially in low-income countries (Ercişli and Sagbas, 2017).

The term underutilized and neglected species refers to a category of wild and cultivated plant species characterized by a low level of investment in research and development, and a recognized but untapped livelihood potential (Eyzaguirre *et al*, 1998). Widespread genetic erosion of these species has been recorded worldwide: for example, in Nepal, more than half of the traditional landraces have become absent from the farmer's fields (Paudel *et al*, 2016). Generally, despite several initiatives in recent years aimed at the conservation of such genetic resources, these wild relatives are still at risk of erosion or extinction (Pilling *et al*, 2020). They have been almost entirely neglected in *ex situ* genebanks (Castañeda-Álvarez *et al*, 2016), although some efforts to change this have begun (Dempewolf *et al*, 2013). In addition, current *in situ* reserves often do not meet the required management standards to maintain the wild relatives and their genetic diversity for long-term use (Iriondo *et al*, 2012; Weibull and Phillips, 2020).

Syria lies within a region of substantial topographic and climatic diversity in the Fertile Crescent, which is the centre of origin and domestication of many globally important crops. Syria is rich in agrobiodiversity (food, forage crops and fruit trees) like wheat, barley, lentils, chickpeas, vetch, olives, almonds, pear, plums, medic, clover, as well as other ornamental, medicinal and aromatic plants. Such plants are the main stocks and heritage for farming in Syria and are widely used elsewhere in the world. According to the *Fourth National Report on Biodiversity in Syria* (MSEA, 2009), the reduction in plant genetic resources was caused by urbanization, climate change, decreasing rainfall and increasing temperatures in dry seasons, which the region has experienced in the last seven years. An increasing incidence of wildfires also poses a particular threat.

During the conflict period from 2012 to 2019, Syria lost 20.4% of its tree cover (Gaafar, 2021), much of it in Idlib and Lattakia governorates. The depletion of wild tree populations has been associated with several factors related to the conflict, including intense reliance on trees for heating and shelter, frequent fires in wild forests, charcoal production, illegal logging, agricultural expansion, and the weakness of state institutions in protecting natural resources and in managing environmental development. The losses of wild tree species include the cherry *Prunus mahaleb*, oak trees, wild pines and hundreds of other plant (tree and crop) species are also threatened.

Unfortunately, there is no data available about the current situation of the WRFT, the only available data were published by FAO (1996), which clearly mentioned the deterioration of plant genetic resources in Syria started before the crisis. It can be assumed that the deterioration of WRFT has increased following the ongoing crisis in Syria resulting from the absence of any control for the protection of these genotypes against the deterioration. Moreover, there was clear evidence of plant genetic resources decline in northwest (NW) Syria based on the finding of previous research about crop landraces in the same locations in NW Syria (AlDarvish *et al*, 2022) The primary causes were the spread of cultivation to new areas, the need to grow economic crops, particularly cereals, forages and food legumes, and accidental forest fires (FAO, 1996; AlDarvish *et al*, 2022). Hence the WRFT were already facing extinction before the Syrian crisis began. Since then, the absence of research authorities responsible for the conservation and propagation of genetic resources has severely exacerbated the problem. Currently, no conservation activity is occurring as the conflict is preventing any access to the field by Syrian botanists, in particular in mountain areas where the threat from mines and bombs is severe (Valderrabano *et al*, 2018).

As a first step towards addressing the lack of activity on these vital global resources, this study aimed to: i) collect information about the current status and critical threats to ten species of WRFT in NW Syria, ii) characterize and map the locations of WRFT exposed to deterioration, and iii) collect seed from multiple genotypes of each species to be used for the establishment of an *in situ* and *ex situ* WRFT collection.

Materials and methods

Data collection and analysis

The National Strategy for Conservation and Management of Plant Genetic Resources for Food and Agriculture 2015–2035 (FAO, 2015) directs national actions for WRFT in Syria, and our methods were designed to address these goals using guidelines recommended by Valderrabano *et al* (2018). The study was conducted between September 2021 and February 2022 in Idleb Governorate (Ehsem, Mhambal, Jisr-Ash-Shugur, Badama, Darkosh, Janudiyeh, Harim, Dana, Kafr Takharim, Qourqeena, Armanaz subdistricts). This region is located within Syria's second and third agroecological zones, incorporating mountains and plains. Agroecological zone 2 covers 2,473,000 ha (13.4% of the country) with an annual rainfall of 250 to 350mm and no less than 250mm across two-thirds of the monitored years. Agroecological zone 3 comprises 1,306,000 ha (7.1% of the country) with an annual rainfall of 250 to 350mm and no less than 250mm over half of the monitored years (FAO, 2003). These agroecological zones are key factors affecting the spread of the WRFT, which is key information when it comes to the planning for subsequent studies on their propagation and preservation. It is worth noting that this region is currently exposed to climatic changes, especially drought, based on the amount of seasonal rain mentioned in the description of this region. To identify the location of WRFT exposed to deterioration after the conflict, the researchers conducted focus group discussions with community representatives in each subdistrict to explain the aim of the study, its methods and objectives (one in each of 11 subdistricts under study during the period from 25 September 2021 to 25 October 2021). These representatives nominated 50 key informant candidates, such as agricultural engineers, senior farmers and agricultural researchers, which had considerable knowledge of the agriculture sector in their region and were targeted for data collection via interviews and a multiple-choice questionnaire (Supplemental data). The questionnaire was designed to gather local knowledge on the location, usage, reasons for decline and appropriate conservation strategies for WRFT.

Due to the outbreak of COVID-19 in NW Syria during data collection, 38 of the targeted informants elected to participate via online interviews, while the remaining 12 agreed to in-person interviews. It's worth mentioning that the quality of online interviews was the same as in-person data collection.

Data collection started on 25 September 2021 and was completed by 25 October 2021. The data collected from the questionnaires were transferred to MS Excel (Microsoft Office 2020) prior to analysis. Data cleaning was undertaken to ensure the accuracy and integrity of the dataset. This involved identifying and rectifying any inconsistencies, outliers or missing values that could potentially skew the results. Variability and dispersion within the dataset were calculated with Excel formulas, including but not limited to measures such as standard deviation, to extract valuable insights.

Seed collection and storage

The main criterion to decide whether the collected seeds were WRFT or not, was their natural spread sites. More specifically, the collected WRFT were collected from wild and forest locations. For species with similar phenotypes and for which phenotype is not sufficiently reliable to distinguish between genotypes, for example, *Ficus carica*, *Olea europaea* and *Vitis vinifera*, the seeds were collected from wild mountainous sites far away from agricultural areas, and according to the interviewees and field team observations. The collected WRFT existed in these wild sites for a long time, and their morphological growth differs from the cultivated species and landraces of the same species (especially the shape of leaves, fruits and trees).

WRFT fully ripe fruits were collected during the harvesting season and the fruit flesh (pulp) was removed. The extracted seeds were then spread out on paper sheets and dried in the open air for 30 days, with the seeds flipped daily to make sure they were completely dry. Then they were disinfected using thiram fungicide, placed in a paper bag labelled with the species name, location, date and storage data, and stored in airtight plastic barrels with dry silica gel for future studies associated with the propagation and establishment of local WRFT *in situ* conservation sites and *ex situ* collections. This conservation method was selected due to the ongoing war in Syria, where electricity is not available for preserving seeds under refrigerated conditions, based on the recommendation of the Millennium Seed Bank staff provided to the research staff through their visit to the Millennium Seed Bank in July 2019 as an alternative method for seed conservation. Moreover, Ashok *et al* (2017) showed that this method of conservation could help to conserve seeds for several years as recommended by Yoshinaga (2010).

For each of the collected accessions, 100 seeds were weighed for each genotype, and the value was multiplied by 10 and expressed as g/1,000 seeds; average values per species, with standard deviation, were calculated.

Results

Focus group discussions conducted with community representatives and experts in the agricultural sector in the targeted subdistricts showed that several WRFT locations had been subject to neglect after the Syrian crisis and had experienced significant deterioration (Figure 1).

Data from the 50 interviews showed that WRFT were found in the following four types of locations within the study area: forests, verges along agricultural roads, rocky and neglected parts of farms, and working farms. The percentage of responses among the 50 participants for the distribution of WRFT in study locations is illustrated in Figure 2.

The interviews revealed that the following ten WRFT species are experiencing erosion: *Prunus mahaleb*, *Prunus orientalis*, *Vitis vinifera*, *Rhus coriaria*, *Olea europaea*, *Crataegus azarolus*, *Ficus carica*, *Pistacia atlantica*, *Prunus spinosa*, and *Pyrus syriaca*.

A range of reasons was reported for WRFT deterioration (Figure 3). The primary causes were overgrazing, frequent droughts and climate change. The predominant conflict-related factors reported were neglect by local authorities, lack of awareness of the importance and value of WRFT and deforestation resulting from the collection of wood for fuel and conversion to arable lands. Several respondents mentioned the lack of interest by the younger generation in making use of local wild fruit landraces, overuse (massive unmanaged fruit gathering), desertification and expansion of building into for-

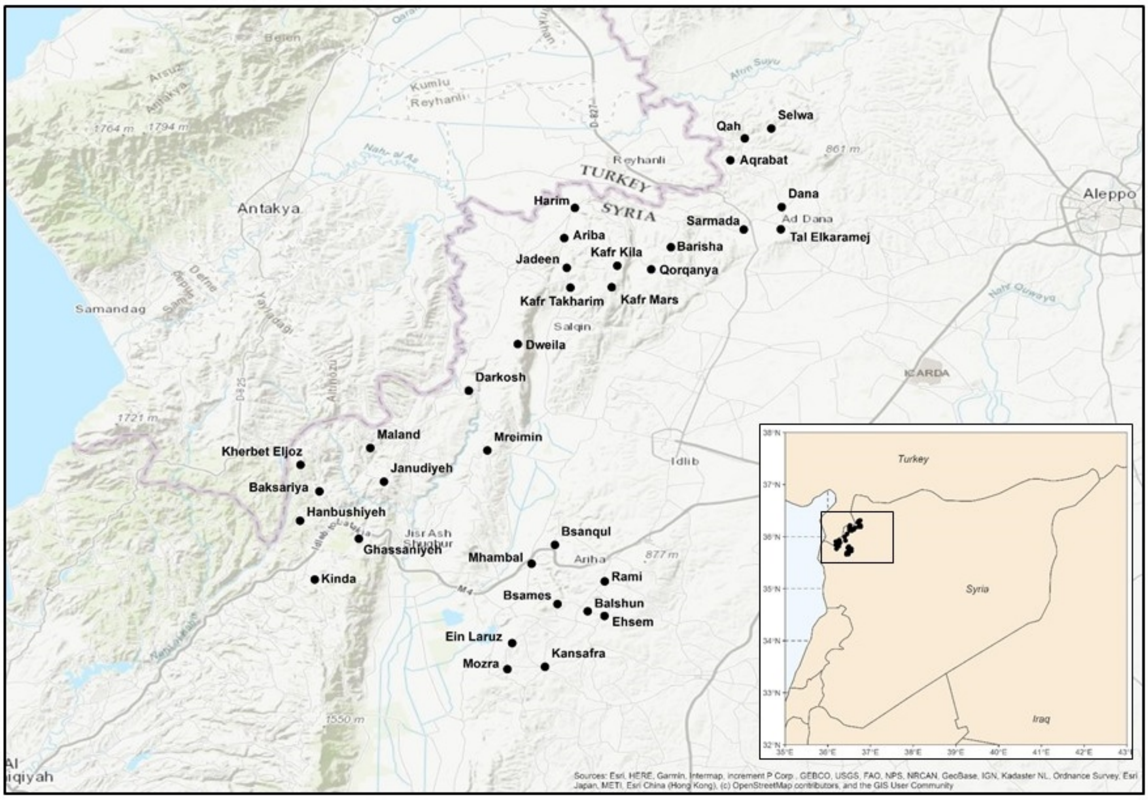

Figure 1. Study area for identification of wild relatives of fruit trees (WRFT) exposed to deterioration. Source: Humanitarian Data Exchange

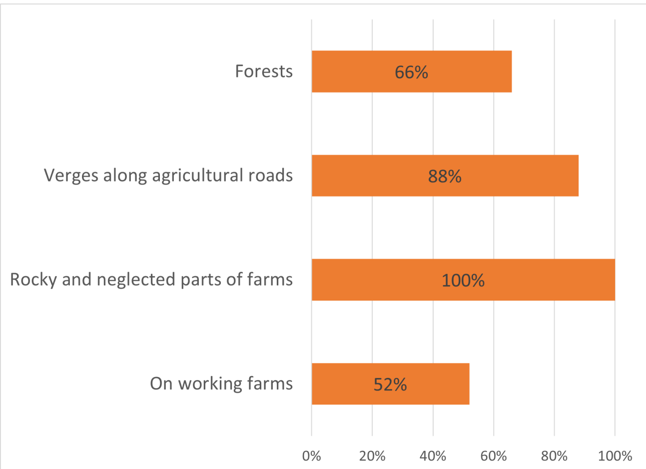

Figure 2. Proportion of respondents reporting types of locations in which wild relatives of fruit trees (WRFT) are found.

est locations as contributing factors. Floods were not considered a threat by any of the respondents. Based on the assessment result with the interviewees, the reasons for erosions of WRFT are equal within all areas of the study.

With respect to the impact of the ongoing conflict in NW Syria on WRFT, all participants stated that the Syrian armed conflict had intensified the deterioration due to the lack of government control leading to overgrazing and excessive cutting of trees in these locations.

When questioned about conservation actions that were considered to be the most important for the protection of WRFT against the current deterioration, participants responses showed the following answers: 36% suggested exchanging seeds of WRFT with other farmers, 26% supported the establishment of local collections for multiplication of these species, 22% considered public awareness campaigns and 14% favoured seed collection and conservation of these species in local genebanks. Only 2% considered awareness campaigns to be the most important action for the protection and preservation of these species.

Regarding the availability of seeds or cuttings of the WRFT species, 56% of the participants stated that seeds or cuttings of these species were not available, whereas 44% stated that they have access to seeds and cuttings. This response suggests that some WRFT are currently at high risk of decline and may even suffer extinction over time.

Collection of WRFT genotypes

The results of the interviews and questionnaire allowed the identification of locations where WRFT had been exposed to deterioration and needed conservation. Based on available resources, the researchers collected mature seeds of each WRFT species from these locations as a first research and conservation step.

For each of the priority WRFT species identified, five genotypes were collected from areas facing particularly severe deterioration There were slight variations in seed

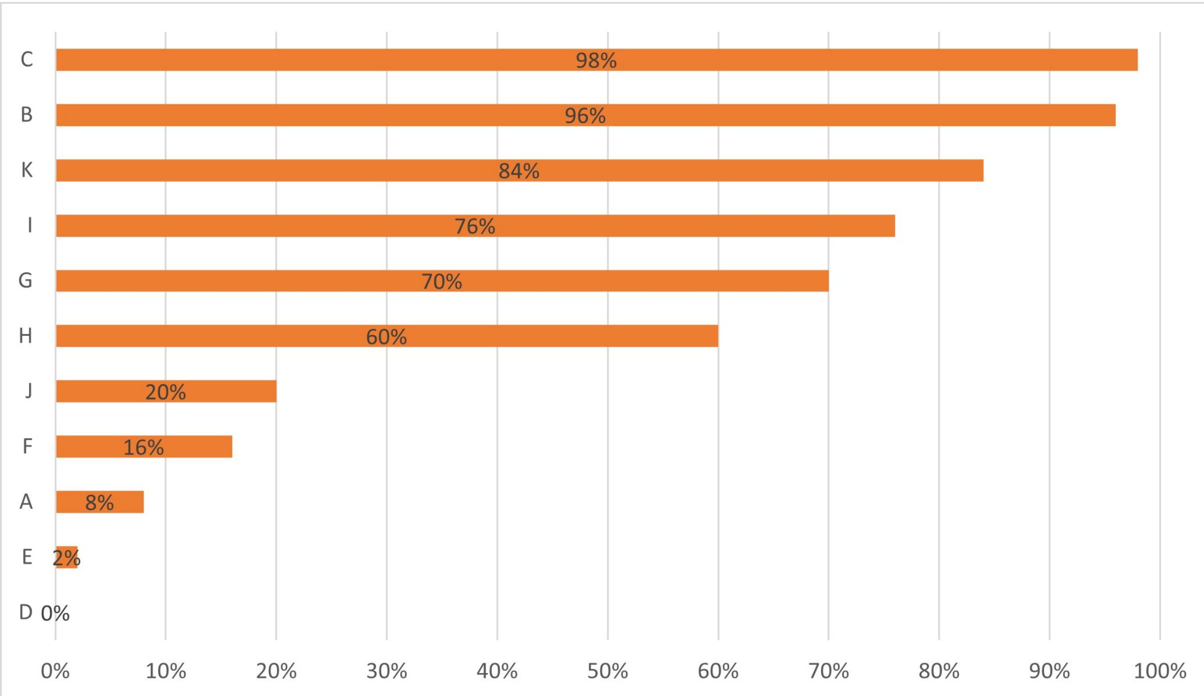

Figure 3. Reasons given, and percentage of respondents reporting, for deterioration of wild relatives of fruit trees (WRFT). From top to bottom – C: Overgrazing; B: Frequent droughts and climatic changes; K: Consequences of Syrian crisis (lack of law enforcement); I: Lack of knowledge about the importance and value of these species; G: Deforestation, including collection of wood for fuel; H: Neglecting by local authorities; J: Interest of the younger generation for local wild fruit landraces is very low; F: Overuse (massive unmanaged gathering); A: Spread of cultivation to new areas and need to grow economic crops; E: Desertification and expanding of buildings towards forest locations and arable lands; D: Floods.

weight among the genotypes within species, and the species averages are given in Table 1.

As seed weight can influence the speed and percentage of germination and can be particularly important in challenging environments (Upadhaya *et al*, 2007), and since maternal effects should be accounted for in any subsequent comparative testing of plants derived from the collection, seed weights for each genotype were taken. The primary purpose of presenting the weight of seeds was to know the standard deviation for the seed weight within different genotypes collected from the same species. This can be investigated in future studies (genetic fingerprinting for these genotypes) to distinguish between phenotype and genotype within the same species and to identify if these variations in seed weight result from environmental factors or are related to genetic features

Table 1. Average seed weights (g/1,000 seeds) for each wild relative of fruit trees (WRFT) species collected.

WRFT Species	Average seed weight (g/1,000 seed)	Standard deviation
Ficus carica	14.82	0.38
Olea europaea	720.00	144.05
Prunus spinosa	309.00	20.74
Vitis vinifera	100.00	7.91
Prunus orientalis	506.00	20.74
Pistacia atlantica	141.00	20.43
Rhus coriaria	92.00	9.08
Prunus mahaleb	180.00	16.96
Crataegus azarolus	400.00	15.81
Pyrus syriaca	165.20	2.83

Discussion

Future food security for humankind is likely to rely heavily on the sustainable availability of a wide range of plant genetic resources for food, agriculture and nutrition, amongst other needs. WRFT are highly valued genetic resources for food production and to support the livelihoods of rural populations, especially in low-income countries. WRFT in Syria are threatened by extinction due to conflict, which has led to increased direct pressure on the tree populations themselves, and to an absence of research authorities working in the field of conservation and propagation of genetic resources. Consequently, there is a high risk that this vital resource may be lost to future research, and its fruits to future generations.

Our results parallel those of previous studies, which have identified that plant genetic resources in Syria are particularly exposed to deterioration arising from the ongoing conflict (Gaafar, 2021; AlDarvish *et al*, 2022). However, similar threats to wild plant genetic diversity have also been reported worldwide (Iriondo *et al*, 2012; Castañeda-Álvarez *et al*, 2016; Paudel *et al*, 2016).

Prior to this study, the most recent available information on the current status of WRFT dates from 11 years ago (FAO, 1996) before the start of the conflict. At that time in the study area, the key locations for WRFT were rich in wild genotypes because they were protected by government authorities such as the Ministry of Agriculture and the Directorates of Environmental Protection. As reported by our interviewees, the absence of state protection has resulted in increased deterioration of WRFT populations in these locations, as exploitation such as grazing or cutting of fuelwood for heating is now uncontrolled (Gaafar, 2021). This neglect and lack of protection for WRFT by state institutions has and continues to exacerbate ongoing pressure from climate change and is resulting in severe genetic erosion of these species. As government bodies have not been present in the study area for over a decade, the condition of WRFT has continued to deteriorate, as reported by Valderrabano *et al* (2018). Moreover, all interviewees stated that the weakness of current state institutions, which demonstrate little interest in wild fruit trees, has led to increased genetic erosion and deterioration of these genotypes in NW Syria.

WRFT are highly valued for use directly as food resources and as commodities that contribute to livelihoods, especially in rural populations in low-income countries. They are also an essential source of genetic diversity for breeding new, higher-yielding, climate change-tolerant varieties. Despite their high value for food and agriculture (Maxted and Kell, 2009; Musayev and Huseynova, 2016; Symphorien *et al*, 2016; Ercişli and Sagbas, 2017; Vincent *et al*, 2019; Zhebentyayeva *et al*, 2019; Engels and Thormann, 2020; Uğur and Gündeşli, 2020), WRFT have been almost entirely neglected in both *ex situ* and *in situ* conservation programmes. As they are likely to be locally adapted and genetically distinctive, WRFT can provide vital novel variation for domesticated fruit species to help meet the challenge of increasingly stressful environments and climatic changes. For example, they have been used by plant breeders to develop more efficient nutrient uptake and utilization, and to integrate genes for adaptation to stressful environments such as water stress, salinity and high temperatures (Ayenan *et al*, 2019; Mishra *et al*, 2022). As for WRFT, Spiegel-Roy (1986) stated that wild relatives of fruit trees are one of the essential resources for fruit tree breeding, especially for disease tolerance, rootstocks and genetic engineering.

To secure the future of these vital resources, conservation protection efforts are urgently needed for the degraded areas they occupy. The priority species we have identified need to be included in plantation programmes to secure the genetic diversity that will be needed for the improvement of commercially cultivated crops. Countries of the Near East and the Mediterranean basin have perhaps the longest tradition in fruit cultivation for historical, environmental and evolutionary reasons (Barone and Caruso, 1998; Wolf *et al*, 2000; Khoury *et al*, 2017; Migicovsky and Myles, 2017; Bissessur *et al*, 2019).

This study represents the first step in highlighting the extremely threatened status of these genotypes. Our initiative to obtain accurately mapped locations for these genetic resources can underpin future research efforts towards *ex situ* and *in situ* conservation programmes. Furthermore, the seed we have collected will form the start of a local genebank collection for future studies of genetic variation and propagation, which can help to ensure resilient and sustainable agricultural development for future generations in Syria and worldwide.

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Supplemental Data

Interview questionnaire

Author contributions

Munzer Aldarvish coordinated the research and contributed to the research design and manuscript; Anas Al Kaddour, Akram Bourgol, and Yasser Ramazan contributed to the research design and manuscript and undertook the data analyses, Yousef Hallak contributed to the research design and carried out the field data collection; Stephen Cavers and Joan Cottrell provided academic guidance and support throughout the research process and contributed to the manuscript.

Conflict of interest statement

The authors of this manuscript have no conflicts of interest to declare. All co-authors have seen and agree with the contents of the manuscript, and there is no financial interest to report. We certify that the submission is original work and is not under review at any other publication.

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