**Crop Vulnerability Statement**

**Leafy Vegetable Crop Germplasm Committee**

**2021**

**Summary of key points**

This report deals with the germplasm status of four major genera: *Lactuca* - lettuce and related species; *Apium* - celery and related species; *Spinacia* - spinach; and *Cichorium* – chicory and endive. The report is restricted to the status of public sector collections of the National Plant Germplasm System (NPGS). These collections comprise cultivars, landraces and wild species, which are tracked in the Germplasm Resources Information Network (GRIN-Global) database once they have been assigned an accession number (either a PI number or a site number). Leafy vegetables play an important role in the U.S. agriculture and human diet/nutrition. Potential sources of genetic variability are always important to allow breeders to develop genetic solutions to new or existing diseases, insects, or pathogens that continue to appear or evolve, challenges of climate change and abiotic stresses, and changes in production practices, consumer preferences, and marketing approaches for growers, shippers, researchers, the industry, and consumers. Some beneficial traits, especially disease and insect resistance, have been identified and successfully transferred into commercial cultivars in breeding programs. The increase in worldwide popularity of crisphead and romaine lettuce cultivars with a relatively narrow genetic base does threaten the diversity of the cultivar base. The spinach cultivar base is currently very vulnerable due to dependence on single genes for resistance to rapidly evolving races of downy mildew. The genetic base for celery and chicory/endive also appears narrow. Maintenance of diverse germplasm collections is essential for widening genetic bases of leafy vegetable cultivars and reducing crop vulnerability. There are few native species of these leafy vegetables in the U. S., except *Lactuca* species; therefore, the potential for *in situ* conservation in the country is limited. Constraints in staffing, facilities, and financial resources hamper the management, regeneration, and evaluation of the expanding crop collections. There are limited genotypic data in the GRIN-Global for leafy vegetable collections. Priority should be given to the acquisition or exchange of wild species to fill the gaps in the collections. In addition to disease and insect resistance, future germplasm evaluation should also include environmental stresses and nutritional value.

**1. Introduction to the crop**

**1.1 Biological features and ecogeographical distribution**

Lettuce (*Lactuca sativa* L.) is a self-pollinated annual plant. It forms a deep taproot with dense lateral roots near the soil surface for water and nutrient absorption. Most cultivars produce nearly sessile leaves spirally arranged in a dense rosette on a shortened stem. There is considerable diversity in leaf color, shape, surface texture, margin appearance, thickness and pliability among the different types and forms of lettuce. Leaf margins may be entire, lobed, incised, indented, or undulating. Leaf surface can be smooth, savoy, or crinkled. The color of lettuce leaves ranges from yellow to dark green and may be dull to glossy; pigmentation from anthocyanin may cover all or part of the leaves, or in a spotting pattern or just along the margin. Stem elongation signals the end of the vegetative growth and the beginning of reproductive stage. A single stem is usually formed to bear the inflorescence, which is a dense corymbose panicle composed of many capitula, each consisting of many florets. The number of florets usually ranges from 12 to 20 but can be as few as seven and as many as 35 (Feráková, 1977). Each floret produces a single-seeded achene, which is ribbed and topped with a pappus. Seed color may be white, yellow, brown, gray, and black. Newly harvested seeds usually have a short period of dormancy, and most cultivars exhibit varying levels of thermodormancy. As a cool-season crop, lettuce is extensively grown on all the continents, particularly in temperate and subtropical regions. The existence of the primitive forms in the Middle East provides strong support for the idea that lettuce probably originated in the eastern Mediterranean basin.

Spinach (*Spinacia oleracea* L.) is a leafy cool season vegetable that produces a rosette during its vegetative phase of growth. Leaves can be ovate, rounded, triangular, or lobed with surface ranging from flat to savoy (crinkled). The crop has a shallow root system and requires good levels of soil moisture and NPK nutrients. In the reproductive phase of growth, the stem elongates and develops a seedstalk with narrow, pointed leaves. Most spinach plants are dioecious, bearing male and female flowers on different plants. In addition, a minority of plants could be monoecious with varying proportions of male and female flowers on the same plant. Seed types could be spiny or round (smooth), although the latter is the standard seed type in the U. S. and Europe. Wild spinach seeds are aggregated in hard spiny units of about five seeds. Spinach is cultivated on all continents, especially in Asia, Europe, and the U. S. Spinach originated in Central Asia, most probably Persia (Iran) (Ryder, 1979). Spinach has always been known as a green leafy vegetable, but a variety with red color on the surface of leaves has recently been released (Mou, 2019).

Celery (*Apium graveolens* L.) is mostly grown for its enlarged, tender, edible petioles. These petioles have sheathing bases and prominently ridged abaxial surface consisting of vascular strands of mostly parenchyma and collenchyma, whereas the adaxial surface is smooth. Small flowers are arranged in large compound umbels. The fruit is a schizocarp that splits when mature into two single-seeded mericarps, remaining attached by a filament (carpophore). Celery is normally biennial, although a few strains and related wild species are annual. The plants will bolt when exposed to cold temperatures, making them unmarketable. Celery is a popular crop that is widely cultivated throughout the world. Although the Mediterranean basin is considered to be the center of the origin for *Apium*, its wide distribution, particularly in the Mediterranean basin, Australia, New Zealand, South Africa and South America, questions this assumption (Quiros, 1993). Argentina and Chile are the countries richest inspecies of the genus. At least six species have been reported as endemic to South America (Quiros, 1993).

Chicory (*Cichorium intybus* L.) and endive (*Cichorium endivia* L.) strongly resemble each other morphologically. Both species have a tap root that is larger in chicory and subdivided in parallel branches and may penetrate deep in the soil to over 1 m in endive. Chicory is a biennial or, in the wild, a perennial, while endive is an annual. They form a rosette of leaves before developing a flowering stalk. The inflorescence has a cluster of 15-25 single hermaphrodite flowers, born on a receptacle and protected by an involucre. Chicory is characterized by a strong sporophytic self-incompatibility system, but endive is self-fertile. Chicory and endive are two traditional European crops, although they are grown in relatively small amounts in North America. Chicory originated in the southern Balkan Peninsula and northern Middle East, while the area of origin for endive is the Middle East (Lucchin et al., 2008).

**1.2 Genetic base of crop production**

Lettuce belongs to the largest dicotyledonous family in the plant kingdom, Asteraceae (Compositae). Of about 100 species of *Lactuca*, only four can be crossed to each other by conventional hybridization methods and thus form the most important breeding group. They include *L. sativa* L., *L*. *serriola* L., *L*. *saligna* L., and *L*. *virosa* L. Crosses between *L. sativa* and *L. virosa* may need to be facilitated by using bridge crosses or embryo rescue. They are all self-fertilized diploids with 2n = 2x = 18 chromosomes. The genome of lettuce has been released (version 8) (Reyes-Chin-Wo et al., 2017). There is a great diversity of shape, size, and color among lettuce cultivars, which are classified into types mainly based on leaf shape, size, and texture, head formation, and stem type. The six generally recognized types are crisphead, butterhead, romaine, leaf, stem, and Latin (Mou, 2008).

Spinach is a diploid with the chromosome number 2n = 2x = 12. Spinach is a member of the Amaranthaceae (was Chenopodiaceae) family that also contains amaranths, sugar beet, chard, and quinoa as well as some common weeds such as lambsquarter. It has two diploid relatives, *Spinacia tetranda* and *S. turkestanica*, which can be crossed with spinach. Currently, hybrid spinach makes up 85 to 90% of production areas in the U. S. and contributes to the yield increase worldwide ([Morelock and Correll, 2008](#_ENREF_59)). The spinach genome has also been sequenced (Dohm et al., 2013; Xu et al., 2017).

Celery, from Apiaceae (Umbelliferae), 2n = 2x = 22, comprises three distinct cultivated forms or taxonomic varieties (Quiros, 1993). *A. graveolens* var. *dulce,* common name celery, features succulent, solid petioles. *A. graveolens* var. *rapaceum*, common name celeriac or root celery, develops enlarged hypocotyl and root tissue, resulting in a globe-like structure. *A. graveolens* var. *secalinum*, common name smallage or leaf celery, has slender, leafy, and often hollow petioles. About 14 species of *Apium* have been described, of which only *A. graveolens* is the cultivated one. The genome sequence of celery was reported recently (Li et al., 2020).

Chicory and endive, both n = 9, are in the Asteraceae (Compositae) family, just like lettuce. Chicory has four horticultural types: 1) a green vegetable for salad/cooking; 2) Radicchio, a heading type with red, green, or mixed colors and spherical or elongated head; 3) Witloof, an apical bud (chicon), which is produced on roots in sand or soil or in the dark (forced), is composed of white or yellow tightly folded leaves with a pointed upper end; and 4) root chicory, roots of which are roasted, extracted, or cooked. Endive consists of two types: 1) Broad-leaved: called escarole or scarole, forms a semi-open head. The outer green leaves are quite bitter, while the inner creamy white to yellow leaves are not as bitter. 2) Narrow-leaved: called endive or frisée, its leaves are much more frilled and its heads are looser, larger, and bitterer than the escarole type (Ryder, 1999). The genome of Radicchio has been sequenced (Galla et al., 2016).

**1.3 Primary products and their value (farmgate)**

Lettuce is a major fresh vegetable and its leaves are commonly found in salad mixtures and sandwiches. In some eastern countries like China and Egypt, stems instead of leaves of lettuce are consumed, either cooked, raw, pickled, dried, or as a sauce. Lettuce products in the U.S. can be categorized into three broad areas: whole heads, bulk harvest (for salad processing, food service, or value-added products), and “baby leaf” or “spring mix.” A significant portion of the crop, about 30%, is shredded or chopped for sale in a container to the restaurant trade or in consumer salad packs. Lettuce is economically the most important crop of the group. It is the most consumed and most valuable fresh market vegetable in the U. S. with 4.1 million tonnes produced on 262,600 acres valued at $3.5 Billion in 2019 (NASS, 2020). In 2019, 51% (by weight) of the lettuce produced in the U.S. was head lettuce, followed by romaine (34%) and leaf (15%) lettuces (NASS, 2020).

Spinach is normally harvested before the bolting stage to avoid the development of the seed stalk, and to reduce yellowing, breakage, and other leaf deterioration. Most spinach is harvested mechanically. Ready to eat, bagged spinach is cut at the crown just above the root, elevated into bins or totes on a trailer, and transported to a packing facility where it is cooled, washed, sorted, and bagged. Still some spinach crops are harvested manually, especially if the whole plant with leaves attached to the root is desirable for fresh markets. In such case, roots are pulled up from the ground and cut below the crown with a sickle and the harvested plants are tied into bunches of 8 to 12 plants. About 20% of fresh market spinach is packed in the field as bunched produce, while the other 80% is bulk harvested for bagged products. Processing spinach is cut above the growing point so that only leaves are harvested, allowing the plant to continue growing for a second and perhaps a third crop. Sometimes the first harvest of a field is used for fresh market, and subsequent harvests are for processing since the crops harvested later are less uniform. About 488,007 tonnes (9,606,000 cwt) of spinach are produced on approximately 26,628 hectares (65,800 acres) in the U.S. annually for both fresh and processed markets with a crop value of about $527 million in 2019 (NASS, 2020).

Celery petioles are consumed mostly raw in salads or as “sticks”, and are often cooked in soups and stews. Celeriac or root celery has a strong flavor and is commonly cooked in stews and soups or grated on salads. Smallage or leaf celery is mostly stir fried in Chinese foods, and are used for their leaves as a condiment garnish and for medicinal purposes. Annual forms of smallage are also grown for seeds, which are used as a condiment and for beverages. The U. S. celery production totaled 799,425 tonnes on 28,100 acres worth $475 million in 2019 (NASS, 2020).

Chicory and endive are mostly produced as a salad or cooked vegetable. Root chicory is also used for coffee substitute, cooked food, or extraction of inulin, a food additive that functions as a filling agent and fat replacement and to prolong shelf life. The escarole and endive production in the U.S. was 47,399 tonnes from 5,170 acres valued at $28 million, as determined in the last survey by NASS in 2001.

**1.4 Domestic and international crop production**

**1.4.1 U.S. (regional geography)**

Lettuce production in the US is concentrated in California (ca. 75-80%) and Arizona (ca. 20-25%) (USDA-[ERS,](#_ENREF_21) 2011), where production is rotated in different regions to ensure a year-round supply of lettuce products. Summer lettuce production (April through October) occurs in three coastal districts of California: Salinas-Watsonville, Santa Maria, and Oxnard, which produce ca. 66% of the total production. Huron to Bakersfield in the west side of the San Joaquin Valley of California, serves as a fall (October to November) and spring (March to April) transition period. Winter production occurs in the low deserts of California (Coachella, Imperial and Palo Verde Valleys) and Arizona (Yuma Valley area). Rotation of production among these distinct areas (coastal districts, San Joaquin Valley, low desert) insures that successive crops are grown in the range of temperatures to which lettuce is adapted. These areas and production dates encompass extensive variation for photoperiod, soil characteristics, and prevailing diseases and pests. However, lettuce production in the San Joaquin Valley has been greatly reduced in recent years due to water shortages. Drip irrigation has been widely adopted in lettuce production to conserve water resources. Lettuce is also produced in Florida from late fall to spring at the Everglades Agricultural Area ( EAA) and other states where lettuce is mainly grown in the frost-free periods from late spring to early fall.

Roughly 66% of the spinach in the U. S. is produced in California, and 23% are produced in Arizona. The remaining 11% production is from other states like Texas, Florida, Oklahoma, Arkansas. Colorado, Maryland, and New Jersey (NASS, 2020). About 87% of the production (by weight) is for fresh market spinach but it accounts for 97% of the value (NASS, 2020). Spinach for processing is grown mostly in California (56%) and Texas (34%). Approximately 90% of the processing crops is for frozen products, while the remaining 10% is for canned spinach.

California produces two-thirds of the celery in the U. S., and the rest are mainly grown in Florida, Texas, and Michigan. Production of chicory and endive in the U. S. is mostly from Florida, California, New York, New Jersey, and Ohio.

**1.4.2 International**

The major lettuce producing countries are China with 57.0% of the total production (by weight), U.S. (13.5%), India (4.5%), Spain (3.4%), Italy (2.8%), Japan (2.1%), Iran (1.9%), Mexico (1.8%), and Turkey (1.8%) that together produced 89% of the world production that totaled 27.3 million tonnes in 2018 (FAOSTAT, 2018). The United States has the largest production of lettuce as a salad crop.

World spinach production was at more than 26 million tonnes in 2018, of which 90.7% was produced in China followed by the U.S. with 1.5% ([FAOSTAT,](#_ENREF_22) 2018). The development of baby leaf and spring mix in bagged salad makes it more convenient to consume this vegetable, and it has contributed to the rapid increase in demand and production of spinach in recent years. Other countries with large acreages of spinach include France, Indonesia, Iran, Italy, Japan, Kenya, Pakistan, and Turkey.

Celery is widely grown in Europe and the Americas. Root celery is grown mostly in Northern and Eastern Europe. Smallage is consumed mainly in Asia and the Mediterranean countries (Quiros, 1993).

Now chicoryis mainly grown all over continental Europe, in South Western Asia, and on limited areas in North America, South Africa, and Australia. In addition to continental Europe, endive is cultivated in Central and Southern America and all along the Mediterranean coast of the African continent. It is also grown in an area in Eastern Asia that includes South Eastern China, Korea, and the eastern part of Inner Mongolia (Lucchin, et al., 2010).

**2. Urgency and extent of crop vulnerabilities and threats to food security**

**2.1 Genetic uniformity in the “standing crops” and varietal life spans**

A. Lettuce

The size and redundancy of the known collections in this country and elsewhere make it extremely unlikely that the crop species will be subject to a drastic change in its genetic diversity. The diversity itself is at a relatively high level. Lettuce occurs in six major forms: crisphead (iceberg and Batavia), butterhead, romaine, leaf, stem, and Latin types. The importance of these types varies from country to country; crisphead and romaine are the most important in the U.S., butterhead is the principal type in northern Europe, romaine in southern Europe, etc. There is therefore a built-in maintenance of certain variability on a geographic scale. On the other hand, the increase in worldwide popularity of crisphead and romaine lettuce cultivars with a relatively narrow genetic base does threaten the diversity of the cultivar base, and therefore, the proper maintenance of collections assumes greater importance.

In the public sector, only the University of California, University of Florida, and the USDA/ARS in Salinas, CA have active breeding and genetic research programs in the U.S. Lettuce is subject to many diseases, in part due to extremely intensive monocultural production systems in the primary growing regions of CA and AZ. A major vulnerability is the worldwide dependence on single genes for resistance to downy mildew (*Bremia lactucae*), which have been rapidly overcome following their deployment. That results in relatively short life spans of commercial cultivars. The pathogens causing diseases such as corky root, Verticillium wilt and Fusarium wilt exist as multiple races capable of overcoming the currently used resistance genes. While it is difficult to predict when new strains of a pathogen will become widespread, it is generally believed that monocultures of crops possessing single gene resistance will select for more virulent strains.

B. Spinach

There are a few spinach breeding programs in the U. S. In the public sector, the University of Arkansas, the USDA/ARS in Salinas, CA, the Texas A&M University at Weslaco, the University of California-Davis, and the Clemson University at Charleston, South Carolina have active breeding and genetic research programs. The program at Salinas was initiated by Beiquan Mou in 2001 and represents the first new spinach breeding project since Teddy Morelock established his program in Arkansas in 1974. Dr. Ainong Shi was hired as the new spinach breeder at University of Arkansas in 2013 after the death of Dr. Morelock in 2009. Carlos Avila leads the spinach breeding program in Texas since 2014. The program at Davis, headed by Charlie Brummer and Allen van Deynze, debuted in 2016. Sandra Branham just started a new spinach improvement program at Charleston in 2020 for the East coast production. As with lettuce, spinach is affected by many diseases and the production system is currently very vulnerable due to dependence on single genes for resistance to rapidly evolving races of downy mildew (blue mold, *Peronospora* *farinosa*). Thirteen new races appeared in the 19 years from 1996 to 2015 (Correll and Koike, 2012; Feng et al., 2014; Ribera et al., 2020), so current cultivars have to be replaced by new resistant ones frequently. Also, since spinach is essentially grown year-round, environmental hazards, particularly excess moisture, sudden freezes and high temperature periods, may be severe. The present germplasm base remains vulnerable.

C. Celery

The genetic base of celery is quite narrow. Most U.S. cultivars derive from 2 or 3 European introductions. The use of a wider germplasm base to breed for disease resistance is just starting. For example, celeriac is being used as a source of *Fusarium* resistance in Lynn Epstein’s program. In 2011, Dr. Epstein replaced retired Carlos Quiros as a manager and primary researcher for the University of California-Davis celery breeding program and has continued to study Fusarium yellows disease (Epstein et al., 2017).

D. Chicory and endive

As relatively minor crops, cultivars of these species may rest on a narrow genetic base, despite substantial morphological diversity. The known collections, composed primarily of European cultivars, are relatively small both in the U. S. and elsewhere in the world. *Cichorium* is native to the Middle East, and the acquisition of wild species, landraces and obsolete cultivars should be a high priority. Currently, there is no public and private breeding programs for chicory and endive in the U.S.

**2.2 Threats of genetic erosion in situ**

Modern agriculture increasingly becomes monoculture and standardized to achieve high efficiency. Commercial lettuce cultivars look very similar, and a crisphead cultivar has to be the ‘Salinas’ type to find large markets. Fresh-market spinach produced in the U. S. are all of the “Western” type with round, flat leaves. Agriculture, housing, and other development reduce the habitat areas for wild relatives of leafy vegetables. Global warming and climate change also accelerate the disappearance of these cool-season wild species.

**2.3 Current and emerging biotic, abiotic, production, dietary, and accessibility threats and needs**

**2.3.1 Biotic (diseases, pests)**

Leafy vegetables suffer from many diseases and insects. The importance of a specific disease or insect varies between production regions. Some common diseases and pests of leafy vegetables are discussed here.

Major fungal diseases for lettuce include downy mildew caused by the obligate parasite, the oomycete *Bremia lactucae* Regel; lettuce drop caused by two related fungal species of *Sclerotinia*, *S. minor* Jagger or *S. sclerotiorum* (Lib.) de Bary; Verticillium wilt (*Verticillium dahliae* Kleb.); and Fusarium wilt by *Fusarium oxysporum* f. sp. *Lactucae*. Lettuce mosaic, caused by lettuce mosaic potyvirus (LMV), is the most important viral disease of lettuce in the world. Big vein is caused by *Mirafiori lettuce big-vein virus* (MLBVV) and vectored by a soilborne fungus *Olpidium virulentus*. The tospoviruses, *Tomato spotted wilt virus* (TSWV) and *Impatiens necrotic spot virus* (INSV) are increasingly a problem. The pathogen most commonly isolated from diseased roots of corky root is the bacterium *Sphingomonas suberifaciens*. The bacteria *Xanthomonas campestris* pv. *vitians* that causes bacterial leaf spot has become a consistent problem of fall harvested lettuce in California and causes up to 100% losses in Florida. Powdery mildew (*Golovinomyces cichoracearum sensu stricto*, more frequently observed in warm, dry production areas in recent seasons), Pythium wilt (*Pythium uncinulatum*), and dieback (*Tomato bushy stunt virus* and *Lettuce necrotic stunt* *virus*) can also cause serious losses in certain years and locations. The lettuce aphid, *Nasonovia ribisnigri* Nr:0 and Nr:1 biotypes, is the most important aphid pest in Great Britain and Holland and has become a serious problem in the U.S. in recent years. Infestation of leafminer (mainly *Liriomyza trifolii, L. huidobrensis*, and *L. langei*) insects causes stand reductions at the seedling stage of lettuce and results in reduced crop quality and crop contamination at harvest. Thrips (western flower thrips (*Frankliniella occidentalis*)) are problematic not only because of the damage they inflict and the diseases they vector, especially INSV, but also because of standards set by export customers regarding the presence of these pests. Soil-borne pests remain an issue for lettuce seedlings, particularly springtails (*Protaphorura fimata*), leading to poor crop stand.

Downy mildew (DM) caused by *Peronospora farinosa* f. sp*. spinaciae* has been a problem in spinach for decades. Other major diseases of spinach include white rust (*Albugo occidentalis*), Fusarium wilt (*Fusarium oxysporum* f. sp. *spinaciae*), Cladosporium leafspot (Cladosporium *variabile*), damping-off diseases (Pythium and Rhizoctonia), and a host of viral diseases ([Correll et al., 1994](#_ENREF_17)). There are several emerging diseases that pose new threats to spinach production, such as Stemphylium leafspot (*Stemphylium botryosum* f. sp. *Spinacia*), *Impatiens necrotic spot virus* ([Liu et al., 2009](#_ENREF_51)), and *Beet necrotic yellow vein virus* ([Mou et al., 2012](#_ENREF_62)). Important insect pests of spinach include leafminers (*Liriomyza* spp.), green peach aphid (*Myzus persicae*), loopers worms (*Trichoplusia ni*), beet armyworms (*Spodoptera exigua*), whiteflies (*Bemisia argentifolii*), thrips (*Frankliniella occidentalis*), and mites (*Rhizoglyphus* spp.) ([Koike et al., 2011](#_ENREF_48)).

Important diseases for celery include Fusarium yellows caused by *Fusarium oxysporum* f. sp. *Apii*, late blight by *Septoria apiicola*, early blight by *Cercospora apii*, bacterial blight of celery (*Pseudomonas syringae pv. Apii* and *Pseudomonas cichorii*)*,* Apium virus Y,and Western celery mosaic virus. Anthracnose (*Colletotrichum acutatum*) appeared in celery in the eastern states and caused very devastating damage in Michigan in 2010 and 2011.  It had been reported in Australia decades ago causing very severe damage as well. Leaf curl caused by  *Colletotrichum fioriniae* was first reported on celery in 2018 in New York state, and is also seen as an emerging disease in nearby states such as Ontario (Canada), Pennsylvania, and Michigan (Sharma et al., 2019 and references therein).Aster Yellows virus while controllable is an issue in Michigan and managed only through control of its vector (Leaf Hopper). Common celery insects are leafminers, beet armyworms, cutworms, as well as symphylan (not a true insect). Aphids of numerous species are also an issue in celery. Lygus bugs are a prominent issue requiring control on celery as their stings cause devastating distortion. Thrips can cause damage but are more cosmetic. In some environments and in particular the northern states like Michigan, root knot nematode can be particularly damaging and can actually enhance the impact of root diseases like Fusarium yellows.

Chicory and endive are affected by many of the same diseases as lettuce: fungal diseases sclerotina drop, downy mildew, damping off, anthracnose (*Michrodochium panattoniana* Berl.), and gray mold (*Botrytis cinerea* Pers.); bacterial diseases soft rot (*Erwinia carotovora* (Jones) Holland) and leaf spot (*Pseudomonas cichorii* (Swingle) Stapp.); and viral diseases Bidens mottle and lettuce mosaic. In addition, a number of diseases have been specifically reported for chicory: violet root rot (*Helicobasidium brebissonii*), black rot (*Phytophtora erythroseptica*), chicory rust (*Puccinia cichorii*), and Verticillium wilt (*Verticillium albo-atrum*). Many aphids and Lepidoptera insects attack chicory and endive plants. Maggots of two dipterous flies, *Ophiomyia* spp. and *Napomyza* spp., are common pests of witloof chicory (Lucchin et al., 2010).

Production of organic products is making genetic resources more critical.  Tolerances are becoming more important due to the expanding demand for organic crops.  Organic production of each of the leafy crops is creating an environment where disease and insect populations are more prolific and potentially more endemic.  This is particularly true since these organic products are prominently produced adjacent to conventional production.  The public demand for organics has been increasing even during a depressed economy suggesting that there is staying power for the organic movement in the United States.  For example, at least 40% of the spinach acreage in California is for organic production. In addition, some alternative control methods (such as fumigation with methyl bromide) for root diseases like Verticillium and Fusarium as well as nematodes and symphylans and spring tails are no longer at the growers’ disposal creating an environment more critical for genetic resources.   

**2.3.2 Abiotic (environmental extremes, climate change)**

As cool season crops, leafy vegetable production is vulnerable to abiotic stresses brought on by climate change. Production of leafy vegetables at high temperature ranges results in yield and quality losses through thermo-inhibition of germination, leaf damages, pre-mature bolting, and internal quality defects such as tipburn. However, brief exposure to frost during the winter season may reduce quality of lettuce. Frost damage often occurs in the winter lettuce production areas of Imperial Valley, California and Yuma, Arizona. Freezing temperatures cause blisters and peeling of lettuce leaves, leading to decay and rot. Damaged leaves also provide entrance for plant pathogens.

Global climate change may affect agriculture more through water availability than temperature. All lettuce and spinach production in California and Arizona are irrigated, but water availability for growers is expected to decrease. Water is a precious resource and has become increasingly scarce due to population growth, environmental needs, and frequent drought. Climate change has resulted in reduced precipitation, less snow pack, and earlier snowmelt, leading to periods of water shortages in California. Court orders limit the pumping of Northern California water to farms in San Joaquin Valley, severely restricting the leafy vegetable production. Coastal Monterey County faces water shortage and seawater intrusion and has planned the construction of several desalination plants for residential uses. Warmer weather accelerates the rates of plant transpiration and water evaporation from soil, and every 10 oF (5.5 oC) increase in temperature translates into a 50% increase in the amount of water required by a crop ([Anonymous, 2010](#_ENREF_1)). Development of germplasm with improved water utilization efficiency/capabilities and generally improved adaptability for better performance under increasingly stressful environmental conditions is needed.

Salinity is a major constraint to crop production in all major lettuce and spinach districts of California. The seawater intrusion has continued to move farther inland into groundwater aquifers beyond city limits of Castroville and Salinas because of continuing overdraft conditions for municipal and agricultural uses in the coastal regions. In the Central Valley, salts accumulate in farmland due to irrigation water from the Sacramento-San Joaquin Delta contaminated with brackish water from the San Francisco Bay, a shallow saline water table, and a lack of adequate drainage outlet. In the Imperial Valley, salts in irrigation water from Colorado River must be carefully managed to prevent yield losses. The water quality problem is exacerbated by the climate change. Global warming leads to higher sea levels, which intensify saltwater intrusion in coastal California. Raising temperatures also promote water transpiration from plants and evaporation from soil, leaving more salts behind in soil.

* + 1. **Production/demand (inability to meet market and population growth demands)**

Lettuce in California and Arizona is produced on raised beds that are typically ca. 10 to 25 cm (4 to 10 in.) high and have either a ‘narrow’ or ‘wide’ width. Narrow beds are 102 to 107 cm (40 to 42 in.) wide, center-to-center, with two parallel seed lines that are 28 cm (11 in.) apart. Growers in Arizona may, for leaf and romaine types, plant a third seed line down the middle of narrow beds (107 cm, 42 in.). Wide beds are 203 to 213 cm (80 to 84 in.) wide with five or six seed lines. These changes in bed width and numbers of seed lines per bed, enable growers to increase the number of plants per hectare by 25% or 50% by eliminating an unneeded furrow with minimal increase in cost ([Smith, 2012](#_ENREF_92); [Whitaker et al., 1974](#_ENREF_103)). These changes have resulted in yield increases.

Bunching and processing spinach is now sown on 102 cm (40 in.) beds with 6 to 9 seed rows or on 203 cm (80 in.) beds with 12 to 21 seed lines and 0.8 to 2.3 million seeds per acre. In general, clipped baby and teenage spinach is planted only on 80 in. beds with 21 to 48 seed lines and 2.7 to 4.0 million seeds per acre ([Koike et al., 2011](#_ENREF_48)).

Commercial celery is planted as transplants in double rows on 91- to 100-cm (36- to 40-inch) beds with plant spacing of 22.5 cm (9 inches) and row spacing of 36 cm (14 inches). Transplants grown in greenhouses and nurseries are used because the seed is very small and difficult to germinate (Daugovish et al., 2008).

Leafy vegetables are losing acreage to housing and industrial development as well as competition from other crops such as strawberry, and land cost in the major production areas of coastal California is very high. Growers are increasing plant densities to increase yield on limited area in order to meet the demand of a growing population. High seeding rates result in dense crop canopies, leading to prolonged periods of leaf wetness and high humidity that favor development of various foliar diseases. These biotic and abiotic stresses including heat, frost, drought, and salinity limit the ability of the leafy greens industry to meet future demand.

* + 1. **Dietary (inability to meet key nutritional requirements)**

Lettuce is the most consumed fresh vegetable in the U.S. In 2019, annual consumption of all types of lettuce was 25 pounds per person, of which 12.7 pounds (51%) were head lettuce and 12.3 pounds were romaine and leaf lettuces (USDA-ERS, 2020). However, the most-popular crisphead type is a poor source of vitamins and minerals. Spinach is called a “super food” due to its high content of beta-carotene (provitamin A), lutein, folate, vitamin C, Ca, Fe, P, and K. But spinach also has a high content of oxalic acid that gives an astringent taste, reduces the bioavailability of minerals, and may cause kidney stones in susceptible people. Therefore, spinach consumption in the U.S. is only about 1/10 of lettuce, of which about 2/3 are fresh market spinach and 1/3 are processed spinach.

Celery is low in calories and is reported to possess many medicinal properties such as antioxidant, hypolipidemic, hypoglycemic, and anti-platelet aggregation activities. However, it also belongs to a small group of foods that can cause a severe allergic reaction that can lead to fatal anaphylactic shock (Lukschal et al., 2016). Per capita consumption of celery was 5.3 pounds in 2019 (USDA-ERS, 2020).

Compliance with the dietary recommendations might also pose a challenge to agriculture. If Americans were to fully meet the 2005 U.S. Dietary Guidelines, the area harvested for fruit in the U.S. would need to increase from the current 3.5 million acres to 7.6 million acres while vegetable acreage would need to be more than doubled from 6.5 million acres to 15.3 million acres (Buzby et al., 2006).

* + 1. **Accessibility (inability to gain access to needed plant genetic resources because of phytosanitary/quarantine issues, inadequate budgets, management capacities or legal and bureaucratic restrictions)**

With the adoption of the Convention on Biodiversity (CBD) in 1993 by the United Nations Environment Program, access to genetic resources worldwide began to become more restricted. Article 15 of the CBD provided that countries have national sovereignty over their genetic resources and access to these resources should be on the basis of prior informed consent and mutually agreed terms. Concerns that this bilateral system of access and benefit sharing would reduce the flow of plant genetic resources for food and agriculture led to the development of the FAO International Treaty (IT) on Plant Genetic Resources for Food and Agriculture, which came into force in 2004 The IT allows for the exchange of many plant genetic resources without excessive transaction costs, but it does not cover all crops, including the leafy vegetables under the purview of this CGC. It is also unclear how the IT relates to access to genetic resources obtained from *in situ* conditions. As a result of these developments in international access, the opportunities to acquire new samples of genetic resources from other countries are limited. For example, it is difficult to collect or exchange crop germplasm in China directly, unless the plant materials are involved in a collaborative evaluation or research with a Chinese institute. The center of origin/diversity for lettuce is the Eastern Mediterranean and Balkan regions. Some countries in this region have granted access to genetic resources on terms acceptable to the NPGS, while others have not. In many countries, there is a lack of financial resources to protect, collect, or maintain plant germplasm resources. Germplasm collected from foreign countries may take a year or longer to go through the quarantine process.

**3. Status of plant genetic resources in the NPGS available for reducing genetic vulnerabilities**

**3.1 Germplasm collections**

**3.1.1 Holdings**

Leafy vegetable germplasm collections in the U. S. and their storage conditions are listed in Appendix (Table 1). Principal lettuce germplasm activities are in Pullman, WA; Salinas, CA; Davis, CA; and Fort Collins, CO (Table 1). Pullman is the official NPGS station for *Lactuca*, conserving accessions of *L. sativa* and several related species ([www.ars-grin.gov/cgi-bin/npgs/html/site.pl?W6](http://www.ars-grin.gov/cgi-bin/npgs/html/site.pl?W6)). The National Center for Genetic Resources Preservation (NCGRP, formerly NSSL) at Fort Collins houses a collection of cultivars and a back-up collection of accessions held at Pullman and Salinas. The collection at Salinas is the largest in the U.S., containing cultivars, PI lines, genetic stocks and breeding lines, and is both a working collection and repository. Pullman and Salinas sites both distribute seeds upon request. The collections at Davis are similar to the Salinas collection but smaller. The status of the lettuce collections in Salinas and Davis was documented in "Genetic Resources of Lettuce and *Lactuca* Species in California", by a task force assembled by the Genetic Resources Conservation Program of the University of California, Davis (1993).

The official NPGS spinach collection is at Ames, IA. There is a collection of cultivars and breeding lines at Salinas, CA and Fayetteville, AR. The NCGRP at Fort Collins houses only a back-up collection of NPGS accessions.

The principal celery collections are at Geneva, NY; Davis, CA; and Fort Collins, CO. Geneva maintains the official NPGS collection; Fort Collins has cultivars of celery and celeriac. The Davis collection is both a working collection and repository. A working collection that was formerly maintained in Belle Glade, FL, is now being maintained in the private sector.

The official NPGS chicory/endive collection of 285 accessions includes over 200 cultivars (primarily of European origin) and is conserved at Ames, IA; other collections are at Salinas, CA. The Salinas collection is primarily for preservation. The NLGRP houses only a backup collection of the NPGS *Cichorium* accessions.

* + 1. **Genetic coverage and gaps**

A. Lettuce

Periodic collection trips have provided input into *Lactuca* holdings but more are needed, particularly with regards to wild *Lactuca*. There are important gaps in the geographic coverage: notably from Egypt, Iraq, Syria, the Balkans, Turkey, and China. Stem type lettuces are important in Egypt and China. These lettuces are historically old and should be evaluated for possible contribution to breeding, most specifically in earliness of market maturity and of seed stalk elongation. Within *L. sativa*, landraces and primitive forms should be collected. Traits for which collections are providing sources for improvement are: *Sclerotinia* resistance, *Botrytis* resistance, additional sources of resistance to lettuce mosaic, yellowing virus diseases, Verticilliumwilt, bacterial leaf spot, leafminer, lettuce aphid, and broad bean wilt, as well as genes for improved quality and nutritional value. Of the four species that make up the present breeding pool, *Lactuca sativa* is well represented in the collections, particularly in the CGN collection in the Netherlands. *L. serriola,* *L. saligna,* and *L. virosa* are poorly represented. In the future, genetic transformation and other technologies are likely to facilitate the use of a wider gene pool, or more distant relatives. In preparation, species that are not sexually compatible with these four should be collected, primarily from Africa, the Middle East and the Far East.

B. Spinach

In this country, limited collections are located at the University of Arkansas, Fayetteville, AR, at the NCGRP in Fort Collins, CO, at the NC-7 Station in Ames, IA, and at Salinas, CA. The collection at Ames emphasizes landraces in a region stretching from Yugoslavia to India. Northern European and Russian landraces are not well represented. There are very few cultivated materials from Japan, Korea, and Southeast Asia. The two wild species of *Spinacia* (*S. tetrandra and S. turkestanica*)are now represented, but should be acquired from more areas. These deficiencies should be addressed through exploration and exchange with other gene banks in order to better represent genetic diversity.

C. Celery

Collections of *Apium graveolens* are adequate, particularly at Davis, CA, but less so at Geneva, NY. The greatest need is for collection of related wild species, at least 20 of which are known only through herbarium specimens. The primary area for collection is South America, particularly from Chile and Argentina. Collecting should also be done in South Africa, Australia and New Zealand. Disease resistance (*Septoria* and *Fusarium*) and insect resistance (leafminers) are the greatest specific needs.

D. Chicory and endive

All existing collections in the U.S. (Ames, Salinas, and NCGRP) consist primarily of modern European cultivars. About 85% of the collection at Ames is made up of cultivars of *C. endivia* and *C. intybus*. The base of the *Cichorium* collection should be broadened by acquiring wild and landrace accessions from the Eastern Mediterranean.

**3.1.3 Acquisitions**

Three plant explorations in the 1980s and 1990s resulted in the collection of over 250 cultivated and wild lettuce samples for the NPGS:

|  |  |  |  |
| --- | --- | --- | --- |
| **Year** | **Country** | **U.S. Cooperators** | **Number of accessions** |
| 1982 | Turkey, Greece | Whitaker and Provvidenti | 245 |
| 1990 | former Soviet Union | Seiler | 25 |
| 1996 | Bulgaria | Hannan, Kaiser | 12 |

Recently, the Plant Exchange Office, in close consultation with Ryan Hayes in an effort to find sources of resistance to *Verticillium* wilt, arranged and supported the following seven plant explorations for wild *Lactuca* species by in-country scientists:

|  |  |  |  |
| --- | --- | --- | --- |
| **Year** | **Country** | **Cooperator** | **Number of accessions** |
| 2009 | Georgia | Mosulishvili | 19 |
| 2009 | Armenia | Tamanyan, Fayvush | 44 |
| 2010 | Russia | Litvinskaya | 19 |
| 2012 | Kyrgyzstan | Lazkov | 22 |
| 2012 | Uzbekistan | Khassanov | 24 |
| 2012 | Georgia | Mosulishvili | 19 |
| 2014 | Azerbaijan | Asgarov | 66 |

A total of 213 accessions of wild Lactuca species were added to the NPGS as a result of these explorations (*Lactuca altaica*, *L. georgica*, *L. orientalis*, *L. quercina* subsp. *quercina*, *L. quercina* subsp. *wilhemsiana*, *L. saligna*, *L. serriola*, *L. tatarica*, *L. undulata*, and *L. virosa*).

The most recent acquisitions of spinach germplasm came from plant explorations planned by David Brenner and funded by the Plant Exchange Office. In 2000, collection of wild relatives of spinach by in country scientists (Durikov and Mamedov) in Turkmenistan resulted in nine accessions. In 2001, collection of wild and cultivated spinach by in country scientists (Akhalkatsi and Mosulishvili) in the Republic of Georgia produced 14 accessions. In 2008, a project was initiated to collect wild species in the United States that are closely related to *Spinacia*. The collected accessions were used in taxonomic revisions (Fuentes-Bazan et al., 2012) which have overturned the previous nomenclature. Most of the close relatives are now in the genus *Blitum*, but *Suckleya* germplasm was also acquired. A set of nine spinach downy mildew disease differentials was acquired from Lindsey du Toit of Washington State University in 2010.

The most recent celery collecting trip was by Orton in 1980. The most recent acquisitions for Cichorium were five “retired” open-pollinated endive varieties donated by Nunhems Netherlands B.V. in 2018, and one expired PVP from Rijk Zwaan Zaadteelt en Zaahandel B.V., Netherlands in 2019.

* + 1. **Maintenance**

Seed storage facilities range from adequate to excellent. The lettuce collections in Salinas and at Fort Collins are stored at –18oC. Celery at Geneva is stored at -20° C with distribution lots stored at 4° C. Original seed lots for the NPGS lettuce collection in Pullman and spinach, chicory, and endive collections in Ames are stored at -18° C and distribution seed lots are stored at 4° C and –18oC (Pullman). Other storages are at 4-5oC at various levels of relative humidity control. The level of redundancy is good for lettuce and spinach, and fair for chicory, endive, and celery. This is outlined in Table I. The accessions are regularly tested for germination % and regenerated if the percentage is low.

* + 1. **Regeneration**

Curators generally take basic characterization data and images as part of the regeneration/maintenance process to “document” an accession. These general characterizations are used to verify taxonomy and also to verify that they are preserving the genetic diversity of an accession to the best of their abilities.

At Pullman, some lettuce accessions have contaminated seeds and low germination. Now cages are used in the field regeneration of cultivated lettuce to prevent cross pollination by insects and seed mixing, while wild species are regenerated in greenhouse because their seeds shatter easily. The off types plants in named cultivars are being rogued out during regeneration, while some PIs were collected as mixtures and are kept as is. Plants are regularly tested for LMV by ELISA.

Beginning in 1994, spinach accessions were sent from Ames to Salinas as a cooperative effort by the USDA and Sakata Seeds to regenerate the collection in isolation chambers. This was successful enough that yearly seed regenerations were not needed for some years. But, increased demand from well-funded research projects has depleted our inventories and availability has fallen so that only 76 % of the 424 accessions are available for distribution. Regenerations are advancing in Salinas at the rate of 15 per year, reduced from 30 per year because of increased demand by spinach research projects for the same greenhouse facilities. Construction of additional facilities for spinach regeneration is recommended. The new greenhouses, like the existing greenhouses, should have positive pressure to exclude drifting pollen of wind pollinated spinach. The wild-species accessions have yielded poorly in the cooperative seed regeneration system; and are now regenerated by the Curator at the NPGS spinach collection in Ames, IA. The principle difficulty is seed dormancy, which is partly overcome by laboriously excising the seeds and waiting up to three years for unsynchronized germination. Seventeen of the eighteen wild accessions are available for distribution (94%) after regeneration at the rate of two regenerations per year.

Chicory and endive accessions are regenerated in Ames in isolation cages using honey bees and alfalfa leaf cutting bees to facilitate pollinations. Eighty-two percent of the collection has had a germination test within the last five years (2014 -2019) to monitor viability of the distribution seed lots, and 91% of the collection is backed up at NLGRP.

* + 1. **Distributions and outreach**

Seeds of leafy vegetable collections, especially lettuce and spinach, are steadily requested and distributed worldwide for crop breeding, scientific research, and educational uses. Seeds are ordered either directly from collection curators or through the GRIN website. In 2019, 93 Cichorium packets in 5 requests were distributed domestically and internationally. In 2018, 224 samples of celery were distributed in 13 orders. In 2019: 2,622 lettuce samples in 148 orders and 3,430 spinach packets in 19 orders were distributed.

* 1. **Associated information**

**3.2.1 Genebank and/or crop-specific web site(s)**

Information regarding the leafy vegetables collections and individual accessions can be obtained by searching the GRIN website (<https://npgsweb.ars-grin.gov/gringlobal/search.aspx>). American Society for Horticultural Science (ASHS) publishes Lists of Vegetable Cultivar Descriptions for North America (Wehner and Mou, 2013) that can be searched online (<https://cucurbit.info/vegetable-cultivar-descriptions-for-north-america/> ) for about 560 lettuce, more than 140 spinach, 70 celery, and 50 chicory/endive cultivars grown in North America. University of California at Davis maintains a Lettuce Cultivar Database (<http://compositdb.ucdavis.edu/database/lettcv2/display/>), which archives genetic, passport, and performance data available for over 4,500 lettuce cultivars.

**3.2.2 Passport information**

Passport information is generally available for leafy vegetable accessions in GRIN. It may include collection date and location or donor and donation date, date PI number was assigned, taxonomic classifications, ancestor, plant type, growth habit, etc. GRIN has a feature that allows bibliographic citations of research papers to be loaded and toggles easily to lists of accessions included in the publication. So it is easier to bridge between literature and germplasm than was previously possible. Nineteen key citations have been loaded this way for spinach. Passport descriptions are less useful for some lettuce accessions that are mixtures.

**3.2.3 Genotypic characterization data**

There are limited genotypic data in the GRIN for leafy vegetable collections. For example, a set of SNP markers are available for lettuce (Kwon et al., 2013). This area has a lot opportunities afforded by the new sequencing technologies and the need to characterize the collections at the DNA level. The whole genome of ‘Salinas’ has been sequenced and assembled into scaffolds (Reynes-Chin-Wo et al., 2017). The *Spinacia* genome was recently sequenced by Dohm et al. (2013) and Xu et al. (2017).

**3.2.4 Phenotypic evaluation data**

A large amount of phenotypic evaluation data exist in GRIN. There are many descriptors for each crop. Lettuce descriptors currently include disease (Verticillium wilt Race 1 and 2), morphology (seed weight and color, bolting, stem, leaf and flower color, head and leaf characters), phenology (days to bolting, plant and seed maturity), and digital images. Spinach descriptors can be divided into chemical (leaf mineral concentrations), morphology (leaf and base color, leaf surface, shape, and erectness), growth (leaf tallness), flower/fruit (monoecious frequency), phenology (bolting), production (seed weight), and digital pictures. Chicory has descriptors for morphology (flower diameter and color, leaf and rib color, leaf characteristics, and plant height), phenology (bolting and flower dates), production (seed weight), and images. Celery descriptors include growth (crown and plant width, plant height, vigor, uniformity, and growth habit), morphology (petiole straightness, compactness, smoothness, and color, leafiness, leaf color, and number, diameter, length, type and split of petiole), and production (keeping quality: pithiness). All available accessions in the *Cichorium* collection were planted in an observation plot in 2006 and characterized for 11 morphological and 2 phonological descriptors which were approved by the Leafy Vegetable CGC.

* 1. **Plant genetic resource research associated with the NPGS**

**3.3.1 Goals and emphases**

The primary need in all crops is to proceed with evaluation of useful or potentially useful characters. In all crops, certain traits have been evaluated reasonably well, others not at all. For the leafy vegetable species, unlike for tomato, wheat and other major crops, there are relatively few researchers among whom to distribute evaluation assignments. Support is necessary to enable the few research groups to maximize their coverage of traits for evaluation. The lettuce group is the largest, with 10 to 12 people able to carry out evaluation assignments. Celery is researched by very few people and evaluation of more than a few of the higher priority traits would be difficult. There is very little research on chicory and endive in this country, although their consumption is increasing. In all crops, evaluation for disease resistance is the main evaluation activity and at the same time continues to be the area of greatest need. Disease evaluation data sets held privately by researchers should be donated to be publicly available in GRIN. Insect resistance, basic physiological studies (bolting), and characterization at DNA level are also important needs.

**3.3.2 Significant accomplishments**

Through germplasm collection and exchange efforts, leafy vegetable accessions have steadily increased in quantity and diversity. The lettuce collection size changed from 1,690 accessions in 2002 to 2,670 accessions in 2020, a 58% increase. The celery collection had 211 accessions in 2002, and 248 accessions in 2019 (18% increase). The collections have been evaluated for many important traits, including disease/insect resistance and human nutrition.

Leafy vegetable germplasm has been utilized as sources of valuable traits (Hayes et al., 2007; Lebeda et al., 2006; Morelock and Correll, 2008; Mou, 2005). This includes identification of resistance to diseases, for example, downy mildew in lettuce and spinach, corky root of lettuce, Verticillium wilt in lettuce, lettuce big vein, and lettuce mosaic virus. Some examples of insect resistance traits are for lettuce aphid and leafminers in lettuce, spinach, and celery. Nutritional traits include carotenoid concentrations in lettuce and spinach and oxalic acid content in spinach.

Beneficial traits, especially disease and insect resistance, have been successfully transferred into commercial cultivars in breeding programs. Depending upon the crop and the situation, both public and private institutions have been involved in germplasm enhancement. Present activity in lettuce includes all stages of enhancement including official release of germplasm and cultivars. Cultivar release is done in both public and private sectors. In celery, California researchers have emphasized production of breeding lines that are stable for specific traits. Proper enhancement in spinach requires increased emphasis on public breeding. The needs for chicory and endive are as yet undetermined but should be assessed.

* 1. **Curatorial, managerial and research capacities and tools**

**3.4.1 Staffing**

Germplasm management is a labor-intensive process that includes seed preservation and regeneration, characterization, taxonomic verification, viability testing, plant/seed health assays, seed distribution, and record keeping. A curator often maintains germplasm collections of many genera. As the sizes of the collections increase every year, the workloads also rise accordingly but resources to accomplish the work do not. The lack of sufficient manpower often results in much longer intervals for viability testing and seed regeneration than desired. For example, the lettuce collection at Pullman is tested for germination every 5 to 10 years. This has led to the distribution of some accessions with low seed germination percentage. Curators may not have enough time, labor, or resources to conduct adequate evaluation, characterization, or collection of germplasm.

The Vegetable Project at the North Central Regional Plant Introduction Station (NCRPIS) is comprised of one full-time Iowa State University Research Scientist III (Curator), one part-time Iowa State University Agricultural Assistant III, and one half-time Iowa State University Agricultural Specialist I (position shared with NCRPIS Farm Management team) which is responsible for not only the NCRPIS *Cichorium* collection, but also the NPGS *Cucumis*, *Cucurbita pepo*, *Daucus*, *Ocimum*, and *Pastinaca*collections (almost 8,000 accessions).  They are assisted by three to four full-time-equivalent Iowa State University student employees as well as supported by the station’s administrative, farm management, plant pathology, information technology, seed storage, order processing, and seed germination personnel.

**3.4.2 Facilities and equipment**

Seed regenerations are often delayed or slowed because there is not enough greenhouse space, field capacity, or isolation cages. Regenerations for *Cichorium* and *Apium* collections have been postponed for a few years because of insufficient facilities and resources, leaving many accessions not available for distribution and/or not backed up at the NCGRP, Ft. Collins, Colorado. Spinach seed regenerations have to compete for isolators with the spinach breeding and other research needs in Salinas, California.

The Ames location is rapidly running out of space in both the 4° C and -20° C seed storage rooms. A request has been made for support for a 2,500 sq. ft. -20° C storage building. A -20° C cold room could essentially double the longevity of viability of many of the taxa maintained at the NCRPIS. Greenhouse space is inadequate, and the facilities are outdated. Continued efforts are made to upgrade as funding becomes available such as the purchase of LED lighting fixtures to improve plant productivity and reduce costs. A few new germplasm regeneration cage frames and screens are purchased annually to replace old and damaged equipment that cannot be repaired on-site. The construction of colder seed storage facilities -18-20oC, at the active sites is helpful for maintaining seed viability and deserves applause.

**3.5 Fiscal and operational resources**

Present activity in public programs is supported largely by in-house research funds. In California, additional support for lettuce, spinach, and celery researches is obtained from commodity group research funds. Portions of these funds are used for germplasm activities. It is legitimate to use these funds, as the germplasm activity supports breeding research. However, the level of germplasm preservation and characterization will always be limited by the need to make decisions about how to divide research support into research and germplasm needs. Therefore, there is a continuing need for unrestricted germplasm support through federal funding.

The operations of the NCRPIS are supported by the USDA-ARS Plant Introduction Research Unit’s CRIS Project, by Hatch Multistate Project NC-007, and from in-kind support from Iowa State University, its host institution.  This funding support provides for the Station’s farm and facilities operations and the scientific, technical, and administrative personnel responsible for curation, maintenance and distribution of more than 1,700 plant taxa, including 1,563 accessions of *Daucus* and associated information.  These funds also support phytosanitary and pollination efforts, and software development focused on improved genebank information workflows.

**4. Other genetic resource capacities (germplasm collections, in situ reserves, specialized genetic/genomic stocks, associated information, research and managerial capacities and tools, and industry/technical specialists/organizations)**

Besides the U.S., there are several germplasm collections of lettuce, both cultivars and wild species,around the world. The major ones (with more than 500 accessions) include Centre for Genetic Resources, Wageningen, the Netherlands (CGN, <http://www.wageningenur.nl/en/Expertise-Services/Statutory-research-tasks/Centre-for-Genetic-Resources-the-Netherlands-1.htm>); Institute of Plant Genetics and Crop Plant Research, Gatersleben, Germany (IPK, [www.ipk-gatersleben.de](http://www.ipk-gatersleben.de)); N.I. Vavilov Institute of Plant Industry, St. Petersburg, Russia (VIR, [www.vir.nw.ru](http://www.vir.nw.ru)); GEVES, Brion, France; INRA, Versailles, France; Research Institute of Crop Production, Olomouc, Czech Republic; Horticulture Research International, Wellesbourne, United Kingdom (HRI); Institute of Agrobotany, Tápiószele, Hungary; Institute of Crop Germplasm Resources, Beijing, China. Passport and evaluation information about the accessions can be searched on-line at the websites provided and seeds may be requested from the institutions. The CGN, IPK, and VIR also have relatively large spinach collections.

The CGN maintains a searchable database for lettuce, spinach, and chicory/endive, the International Leafy Vegetables Database (<https://ecpgr.cgn.wur.nl/LVintro/>) which currently contains information of more than 11,697 accessions from 30 institutes and genebanks in 23 countries. This represents all major *Lactuca* collections in the world except China. According to the database, the major collections of chicory/endive (with more than 100 accessions) include IPK (692 accessions), GEVES (the French Variety and Seed Study and Control Group, 464 accessions), and VIR (149 accessions).

According to Genesys PGR (<https://www.genesys-pgr.org/welcome> ), the largest non-U.S. holdings of *Apium graveolens* are IPK (250 accessions), VIR (155 accessions) and the Center for Plant Diversity Institute (NODIK) in Hungary (91 accessions).

With the exception of *Lactuca* species, there are few native wild relatives of these leafy vegetables in the U.S.; therefore, there is not much opportunity to establish *in situ* reserves to protect them. Eleven *Lactuca* taxa have been recorded in the U. S., including species that originated and are distributed only in North America, as well as species that are synanthropic and cosmopolitan (*L. serriola, L. saligna, and L. virosa*), although *L. serriola* (prickly lettuce) is a fairly common weed across the country (Lebeda et al., 2004). In a survey, 80% of 231 seed samples collected in North America were identified as *L. serriola* (Lebeda, et al., 2011). Seven species in North America can be considered autochthonous, of which *L. canadensis, L. graminifolia,* and *L. biennis* are the most common (Lebeda et al., 2004).

**5. Prospects and future developments**

A. Lettuce - Evaluation and acquisition should remain top priority. There have been seven collection trips from 2009 through 2014; these should be continued, perhaps in conjunction with the collection of other species. Effort should continue to clean up the contamination in some accessions and increase the frequency of viability testing. Evaluation of disease and insect resistance, bolting activity, and reaction to environmental stresses are of paramount importance. Future evaluation should also include nutritional value. With the application of new sequencing technologies, there should be more molecular characterization of the collections. Significant advances in documentation of existing materials have been made with extensive field and greenhouse plantings. New data, including disease resistance evaluation results, should be routinely entered into the GRIN system. Data from NPGS funded evaluations should be required to be in GRIN-Global friendly format. All the information should be integrated so as to be useful for breeders. Enhancement should continue in several programs and the quality of the storage facilities should be maintained.

B. Celery - Priority should be given to collection of wild *Apium* species. There are 20 known wild species, which are not well represented in collections. Geneva, NY has only one accession for one species and Davis, CA has only one or two for three other species. Other species are known only through herbarium specimens. The degree of danger of extinction in natural habitats is not known and should be determined. The potential usefulness of the wild species is extrapolated from the finding of several resistances to diseases and insects in available materials. Disease/insect resistance data should be collected and entered in GRIN. A second priority is to determine cross compatibility between cultivated celery and related species.

C. Spinach - Collection of wild species from Iran, and landraces from northern Europe, Russia and Eastern Asia is also important. Since disease resistance is the main breeding concern, the highest priority should be given to acquiring disease resistance data on the NPGS collection for inclusion in GRIN to make information available to the user community. Insect resistance, bolting response, and adaptation to extreme environmental conditions are also important to the continued cultivation of this crop in the U.S.

D. Endive and chicory - Collection of wild species and landraces from the eastern Mediterranean is of high importance. Additional collections of European cultivars and breeding lines may be needed to fill in the gaps in these holdings. The evaluation and verification of existing cultivar collections would increase their potential utility to users as would development of a standardized descriptor list that include biotic/abiotic stresses. Existing samples of cultivars may need to be selected for trueness-to-type. Some accessions appear to be mixtures. It would help culling off-types if cultivar descriptions are available to compare accessions to. Such descriptions may be hard to find, especially for some older European varieties. There is increasing interest in radicchio and cultivars with unusual leaf characteristics for use as specialty salad greens.

**6. References**

Anonymous. 2010. How real is global warming? Pioneer Growing Point Magazine: 12-13.

Buzby JC, H. F. Wells, G. Vocke. 2006. Possible implications for U.S. agriculture from adoption of select dietary guidelines. Economic Research Report No. 31, Economic Research Service, U.S. Dept. of Agriculture, November 2006.

Correll, J., S. Koike. 2012. Race Pfs: 14 – Another new race of the spinach downy mildew pathogen. Monterey County Crop Notes July/August: 16-17.

Correll, J.C., T.E. Morelock, M.C. Black, S.T.Koike, L.P. Brandenberger, and F.J. Dainello. 1994. Economically important diseases of spinach. Plant Dis. 78:653-660.

Daugovish, O, Smith, R, Cahn, M, Koike, S, Smith, H, Aguiar, J, Quiros, C, Cantwell, M, Takele, E (2008) Celery Production in California. UC Vegetable Research & Information Center. Vegetable Production Series. <http://anrcatalog.ucdavis.edu/pdf/7220.pdf>. Accessed July 31, 2019.

Dohm, J.C., A.E. Minoche, D. Holtgräwe, S. Capella-Gutiérrez, F. Zakrzewski, H. Tafer, O. Rupp, T. Rosleff Sörensen, R. Stracke, R. Reinhardt, A. Goesmann, T. Kraft, B. Schulz, P.F. Stadler, T. Schmidt, T. Gabaldon, H. Lehrach, B. Weisshaar, H. Himmelbauer. 2013. The genome of the recently domesticated crop plant sugar beet (*Beta vulgaris*). Nature 505 (7484): 546-549.

Epstein, L., Kaur, S., Chang, P.L., Carrasquilla-Garcia, N., Lyu, G., Cook, D.R., Subbarao, K.V. and O’Donnell, K., 2017. Races of the celery pathogen *Fusarium oxysporum* f. sp. *apii* are polyphyletic. Phytopathology, 107:463-473.

FAOSTAT. 2018. Food and Agricultural Organization of the United Nations (FAO). Crops. <http://www.fao.org/faostat/en/#data/QC> . Accessed September 25, 2020.

Feng, C., Correll, J. C., Kammeijer, K. E., and Koike, S. T. 2014. Identification of new races and deviating strains of the spinach downy mildew pathogen *Peronospora farinosa* f. sp. *spinaciae.* Plant Dis. 98:145-152.

Feráková, V., 1977, *The Genus Lactuca L. in Europe*, Universita Komenskeho, Bratislava.

Fuentes-Bazan, S., P. Uotila, T. Borsch. 2012. A novel phylogeny-based generic classification for *Chenopodium* sensu lato, and a tribal rearrangement of *Chenopodioideae* (*Chenopodiaceae*). [Willdenowia](http://www.ars-grin.gov/cgi-bin/npgs/html/stdlit.pl?Willdenowia) 42: 5-24.

Galla, G., A. Ghedina, S. Tiozzo, and G. Barcaccia. 2016. Toward a first high-quality genome

draft for marker-assisted breeding in leaf chicory, Radicchio (Cichorium intybus L.). Plant Genomics, Ibrokhim Y. Abdurakhmonov, IntechOpen. DOI: 10.5772/61747. Available from: <https://www.intechopen.com/books/plant-genomics/toward-a-first-high-quality-genome-draft-for-marker-assisted-breeding-in-leaf-chicory-radicchio-cich> Accessed October 1, 2020.

Hayes, R.J., G.E. Vallad, Q-M. Qin, R. C. Grube, and K. V. Subbarao. 2007. Variation for resistance to Verticillium wilt in lettuce (*Lactuca sativa* L.). Plant Disease 91:439-445.

Koike, S.T., M. Cahn, M. Cantwell, S. Fennimore, M. Lestrange, E. Natwick, R.F. Smith, and E. Takele. 2011. Spinach production in California. University of California, Agriculture and Natural Resources, Publication 7212. <https://anrcatalog.ucanr.edu/pdf/7212.pdf> Accessed October 1, 2020.

Kwon, S. J., I. Simko, B. Hellier, B. Mou, and J. Hu. 2013. Genome-wide association of 10 horticultural traits with expressed sequence tag-derived SNP markers in a collection of lettuce lines. The Crop Journal 1: 25-33.

Lebeda, A., I. Dolezalova, V. Ferakova, D. Astley. 2004. Geographical distribution of wild *Lactuca* species (Asteraceae, Lactuceae). The Botanical Review 70(3): 328-356.

Lebeda, A., I. Dolezalova, M. Kitner, A. Novotna, P. Smachova, M.P. Widrlechner. 2011. North American continent – a new source of wild *Lactuca* spp. germplasm variability for future lettuce breeding. Acta Horticulturae 918: 475-482.

Lebeda, A., E.J. Ryder, R. Grube, I. Dolezalova, E. Kristkova. 2006. Lettuce (Asteraceae; *Lactuca* spp.). In: R. J. Siongh (Eds.). Genetic Resources, Chromosome Engineering, and Crop Improvement. CRC Press, New York, NY.

Li, M.,K. Feng, X. Hou, Q. Jiang, Z. Xu, G. Wang, J. Liu, F. Wang, and A. Xiong. 2020. The genome sequence of celery (*Apium graveolens* L.), an important leaf vegetable crop rich in apigenin in the Apiaceae family. Horticlture Research 7:9. https://doi.org/10.1038/s41438-019-0235-2

Liu, H.Y., J.L. Sears, and B. Mou. 2009. Spinach (*Spinacia oleracea*) is a new natural host of *Impatiens necrotic spot virus* in California. Plant Dis. 93:673.

Lucchin, M., S. Varotto, G. Barcaccia, and P. Parini. 2008. Chicory and endive. In: J. Prohens, and F. Nuez (eds.). Handbook of plant breeding, Vegetables I, Asteraceae, Brassicaceae, Chenopodicaceae, and Cucurbitaceae. Springer, New York, NY, p. 3-48.

Lukschal, Anna, Julia Wallmann, Merima Bublin, Gerlinde Hofstetter, Nadine Mothes-Luksch, Heimo Breiteneder, Isabella Pali-Schöll, and Erika Jensen-Jarolim. 2016. Mimotopes for Api g 5, a relevant cross-reactive allergen, in the celery-mugwort-birch-spice syndrome. Allergy, Asthma & Immunology Research 8: 124-131.

Morelock, T.E., and J.C. Correll. 2008. Spinach, p. 189-218. In: J. Prohens, and F. Nuez (eds.). Handbook of plant breeding, Vegetables I, Asteraceae, Brassicaceae, Chenopodicaceae, and Cucurbitaceae. Springer, New York, NY.

Mou, B. 2005. Genetic variation of β-carotene and lutein contents in lettuce. Journal of the American Society for Horticultural Science 130(6): 870-876.

Mou, B.2008. Lettuce. In: J. Prohens and F. Nuez, ed., *Handbook of Plant Breeding, Vegetables I, Asteraceae, Brassicaceae, Chenopodicaceae, and Cucurbitaceae*. Springer, New York, p. 75-116.

Mou, B. 2019. ‘USDA Red’ spinach. HortScience 54: 2070-2072. <https://doi.org/10.21273/HORTSCI14308-19>

Mou, B., K. Richardson, S. Benzen, and H.Y. Liu. 2012. Effects of *Beet necrotic yellow vein virus* in spinach cultivars. Plant Dis. 96:618-622.

NASS. 2001. National Agricultural Statistics Service. U.S. Dept. of Agriculture (USDA). Quick Stats. <https://quickstats.nass.usda.gov/>. Accessed September 20, 2020.

NASS. 2020. National Agricultural Statistics Service. U.S. Dept. of Agriculture (USDA). Vegetables 2019 Summary (February 2020). <https://usda.library.cornell.edu/concern/publications/02870v86p?locale=en>. Accessed Sept. 4, 2020.

Quiros, C. F. 1993. Celery *Apium graveolens* L. In: G. Kalloo and B. O. Bergh (Eds.), Genetic Improvement of Vegetable Crops. Pergamon Press, Oxford, England.

Reyes-Chin-Wo, S., Wang, Z., Yang, X. *et al.* 2017. Genome assembly with *in vitro* proximity ligation data and whole-genome triplication in lettuce. *Nat Commun* **8,**14953. doi:10.1038/ncomms14953

Ribera, A., Y. Bai, A. Wolters, R. van Treuren, and C. Kik. 2020. A review on the genetic resources, domestication and breeding history of spinach (*Spinacia oleracea* L.). Euphytica 216:48. https://doi.org/10.1007/s10681-020-02585-y

Ryder, E. J. 1979. Leafy Salad Vegetables. P. 195-227. AVI Publishing Co., Westport, Conn.

Ryder, E. J. 1999. Lettuce, Endive and Chicory. CABI Publishing, Oxon, U. K.

Sharma, S., Pethybridge, S., Buck, E., and Hay, F. 2019. First report of Leaf Curl on celery (*Apium graveolens* var. *dulce*) Caused by *Colletotrichum fioriniae* in New York. Plant Disease 103: 1791

Smith, R. 2012. Changing vegetable crop production practices. <http://www.growingproduce.com/article/32166/changing-vegetable-crop-production-practices>. Accessed October 1, 2020.

Sowbhagya, H.B. 2014. Chemistry, technology, and nutraceutical functions of celery (*Apium graveolens* L.): An overview. Critical Reviews in Food Science and Nutrition 54: 389-398.

USDA-ERS. 2011. Economic Research Service. U.S. Lettuce Statistics 2011. <https://usda.library.cornell.edu/concern/publications/cc08hf60z?locale=en> . Accessed October 1, 2020.

USDA-ERS. 2020. Vegetables and Pulses Yearbook Tables. U.S. per capita use of fresh and processing vegetables, dry pulse crops, and potatos; U.S. vegetable trade. <https://www.ers.usda.gov/data-products/vegetables-and-pulses-data/vegetables-and-pulses-yearbook-tables/> Accessed October 1, 2020.

Wehner, T. C. and B. Mou (Ed.). 2013. Vegetable cultivar descriptions for North America, List 27. HortScience 48: 245-286.

Whitaker, T.W., E.J. Ryder, V.E. Rubatzky, and P.V. Vail. 1974. Lettuce production in the United States. U. S. Dept. Agric., Agric. Res. Serv. Agric. Hdbk. No. 221.

Xu, C., C. Jiao, H. Sun, X. Cai, X. Wang, C. Ge, Y. Zheng, W. Liu, X. Sun, Y. Xu, J. Deng, Z. Zhang, S. Huang, S. Dai, B. Mou, Q. Wang, Z. Fei, and Q. Wang. 2017. Draft genome of spinach and transcriptome diversity of 120 *Spinacia* accessions. Nature Communications 8: 15275. doi: 10.1038/ncomms15275

**7. Appendices (number and lengths at the CGC’s discretion)**

**Table I. Leafy vegetable germplasm collections in the United States**

|  |  |  |
| --- | --- | --- |
| Location | Description of Collection | Storage Conditions |
| Lettuce |  |  |
| Salinas, CA | 2,227 cultivars, 1,275 PIs, 289 other varieties, more than 10,000 breeding lines and genetic stocks; base and working | -18oC, RH not controlled |
| Davis, CA | 1,600 cultivars, 1,500 PIs, 1,100 breeding lines; working collection | 5oC, RH controlled |
| Pullman, WA | 2,670 accessions | Distribution seeds ½ at 4oC and ½ at -18oC; original seed lots at -18oC. RH controlled at 4 ℃ |
| Ft. Collins, CO | 1,502 accessions (backup) | -18oC, 5-7% RH |
| Celery |  |  |
| Davis, CA | 250 PIs | 5oC, RH controlled |
| Geneva, NY | 248 accessions | Distribution seeds at 4oC, 25-27% RH; additional seed lots at -20oC |
| Ft. Collins, CO | 87 accessions (backup) | -18oC, 5-7% RH |
| Spinach |  |  |
| Ames, IA | 424 accessions | Distribution seeds at 4oC, 25-27% RH; original seed lots at -18oC |
| Fort Collins, CO | 396 accessions (backup) | -18oC, 5-7% RH |
| Fayetteville, AK | 600+ PIs and cultivars | -18 oC, RH not controlled |
| Salinas, CA | 779 accessions; 80 cultivars and hybrids | -18 oC, RH not controlled |
| **Chicory/Endive** |  |  |
| Salinas, CA | 222 chicory, 191 endive accessions, 19 PIs | -18oC, RH not controlled |
| Ames, IA | 285 accessions | Distribution seeds at 4oC, 25-27% RH; original seed lots at -18oC |
| Ft. Collins, CO | 259 accessions (backup) | -18oC, 5-7% RH |