CHAPTER 1

EVALUATING COMMON DANDELION (*Taraxacum officinale* Weber) POPULATION DIVERSITY USING MORPHOLOGICAL CHARACTERISTICS AND DNA BASED GENETIC ANALYSIS.

Abstract: Population diversity of common dandelion was examined using morphological characteristics and genetic analysis. Seed from individual common dandelion plants were collected from multiple counties in Michigan and several states. Subsequent plants were established in field nurseries at Michigan State University research stations near East Lansing and Chatham to determine if common dandelion collected from different geographical regions exhibit phenotypic variability. Overall, plants at the East Lansing nursery tended to be larger and produce more seeds than those at the Chatham nursery. Individual collections that were larger and produced more seed at the East Lansing nursery were also the largest and most prolific at the Chatham nursery. Genetic diversity of common dandelion collections established in the field nurseries were also evaluated using randomly amplified polymorphic DNA analysis. Nine random primers amplified a total of 44 fragments that were polymorphic. Of the 26 populations screened, 24 were distinguishable from each other using the RAPD analysis. The diversity of polymorphic banding patterns observed suggest that there is a high level of genetic diversity in common dandelion in Michigan and the other states. Genetic similarity coefficients for all the populations evaluated ranged from 0.25 to 1.00. There was no discrete separation between common

dandelion from Michigan and the other collections. A survey of plants collected from a single no-tillage field in Michigan also revealed a high level of diversity. An additional study was conducted to verify the apomictic reproductive nature of this species. All progeny that were tested using the given random primers were genetically similar to the maternal plant. There does not appear to be a visible relation between morphological characteristics and the genetic similarity examined here. A greenhouse study was conducted to determine if differences in plant size would be observed between selected collections of common dandelion. Nine collections of common dandelion were grown in the greenhouse and the size of the plants compared 60 days after planting using leaf area and dry weight. Differences in plant size were observed; however collections that were genetically similar did not necessarily similar in size. With the high level of diversity documented in this study one could expect diversity in common dandelion response to certain herbicides.

Nomenclature: common dandelion, *Taraxacum officinale* Weber; red seeded dandelion *Taraxacum laevigatum* L.

Index words: apomictic, phenotypic variation, RAPD analysis, genetic variation, genetic similarity.

INTRODUCTION

Common dandelion (*Taraxacum officinale* Weber) has developed into a troublesome agronomic weed, especially in no-tillage crop production in Michigan and parts of the United States. Putatively originating in west central Asia (Richards 1970), this species can be found world-wide, primarily concentrated in temperate and cold regions (Solbrig and Simpson 1974). It was proposed that common dandelion was originally introduced to the Americas via the Alaskan ice bridge following the most recent ice age (Richards 1973). It is also presumed that early European settlers reintroduced common dandelion as an ornamental used to seed the roofs of sod houses to make them stand out on the prairie (Solbrig 1971; Stubbendieck et al. 1995).

Common dandelion, as it is collectively classified in the United States, is comprised of two similar *Taraxacum* species. Red seeded dandelion (*T. laevigatum*) is virtually indistinguishable from *T. officinale* except for the red coloration of the achene (GPFA 1996). Morphological and biochemical analysis comparing these two species showed no clear differentiation between them (Taylor 1987). A more comprehensive genetic analysis of ribosomal DNA (rDNA) and chloroplast DNA (cpDNA) supported this lack of a definite separation between these species (King 1993). Furthermore, it has been argued that morphological variation is by and large a response to the local environment (Taylor 1987). For these reasons, *T. laevigatum* and *T. officinale* are collectively considered common dandelion here.

The genus *Taraxacum* is comprised of both sexual and asexual species. *Taraxacum* species that reproduce sexually are diploid (2n = 16), whereas the asexual species are triploid (3n = 24) and reproduce via agamospermous apomictic seed production. The asexual species, which include *T. laevigatum* and *T. officinale*, are found primarily in North America where it is accepted that they reproduce via apomixis. The result of this apomictic mode of reproduction is progeny that are clones of the maternal parent. Despite the potential for populations in a given area to be genetically identical, differences in overall fitness and isozyme characterization have been documented (Solbrig and Simpson 1974). Additional studies of genotypic variation in rDNA have been reported in asexual lineages of common dandelion thought to be brought about by somatic mutations (King and Schaal 1990).

DNA-based molecular markers, such as randomly amplified polymorphic DNA (RAPD), are a powerful tool to examine genetic variation within a species (Williams et al. 1990). An advantage of using RAPD markers over other DNAbased markers (ie. SSR and AFLP) is that no prior knowledge of the species genome is required. Single oligonucleotide primers of arbitrary sequence are used to randomly amplify segments of template DNA. The simple presence or absence of an amplified DNA fragment represents a difference in the genome that can be used to compare individuals. The use of RAPD analysis has been utilized to examine genetic diversity in such weed species as leafy spurge (*Euphorbia esula* L.) (Rowe et al. 1997), wild mustard (*Sinapsis arvensis* L.) (Moodie et al. 1997), and hemp dogbane (*Apocyanum cannabinum* L.) (Ransom

et al. 1998), as well as economically important species such as tea plant (*Camellia sinensis* L.) (Jorge et al. 2003) and walnuts (*Juglan spp.*) (Orel et al. 2003).

How genetic variation affects common dandelion management is currently unknown. Therefore studies were conducted to understand genetic variation of this species to aid in future management strategies for this weed. The objectives of this research were to utilize RAPD analysis to 1) determine the amount of genetic diversity of common dandelion in Michigan and the United States, and 2) identify unique populations of common dandelion.

MATERIALS AND METHODS

Plant material

To examine the population diversity of common dandelion, mature seed (as indicated by the presence of white pappus) was collected from individual plants from selected sites in 2001 (Table 1). Seeds were removed from the flower receptacle and stored at 4 C until planting. Seed were planted in 1000 ml pots filled with commercial potting soil¹ and maintained in the greenhouse. Seedlings were transplanted to individual 1000 ml pots filled with Spinks loamy sand (sand, mixed mesic Psammentic Hapludalfs) with pH of 6.8 and 2.4% organic matter. Plants were maintained in the greenhouse until they were transplanted to a field nursery.

Phenotypic variation among collections

Phenotypic variation of common dandelion was examined by establishing plants in field nurseries and observing a number of different morphological and reproductive characteristics. Common dandelion field nurseries were established at two sites in Michigan. A southern and northern nursery was established at the Michigan State University Agronomy Farm at East Lansing (42° N latitude) and the Michigan State University Upper Peninsula Research Station near Chatham (47° N latitude), respectively. Morphological differences were compared among common dandelion collections from 12 counties in Michigan, 11 states, and a collection obtained from the Beal Botanical Garden at Michigan State University that originated in Germany (Table 1). Single plants were randomly selected to represent that population in which it was growing. Common dandelion plants from agronomic fields and residential areas were selected for this analysis.

Common dandelion seedlings were established in the greenhouse and transplanted into 0.6 by 0.6 m plots at the nurseries in the spring of 2002. Plants were irrigated weekly for the first month and with natural rainfall for the remainder of the experiment. Plants grew free of competition by hand weeding around established common dandelion plants. Characteristics observed at each of the nurseries included winter survival, plant diameter, leaf shape, leaf pubescence, flowering date, growing degree days to flowering, total number of flowers produced, and seeds produced per flower. Winter survival was determined by observing plants in the spring of the year following establishment in the field nurseries. Plant diameter was recorded as the average of two perpendicular

measurements of the common dandelion rosette. Leaf shape was determined using a scale from 1-5, where 1 represented a leaf with deeply lobed leaf margins and 5 represented an entire leaf margin. Flowering date was recorded as the day in which the first yellow flower was present on the individual plant. Growing degree days were calculated using a 10 C base beginning on March 1, 2003. Total flower production was monitored weekly starting in May and continuing until flower production declined approximately one month later. The total number of flowers produced each week was recorded, the mature flowers were removed, and the seeds placed in paper envelopes. The number of seeds produced per flower was determined by randomly collecting four individual mature flowers per plant prior to seed dissemination. Mature flowers were dried at 70 C for 24 h and stored at room temperature until the seeds were counted.

Genetic variation among collections

Genetic analysis of common dandelion using RAPD analysis was conducted to assess the amount of genetic variation in this species. Population genetic diversity was examined for common dandelion collected from 16 counties in Michigan, 9 states, and the collection from Germany (Table 1). Genomic DNA was extracted from established plants in the common dandelion nursery at East Lansing.

Within-field genetic variation

The genetic diversity of common dandelion within a field population was examined using eight established plants that were collected from a 0.5 ha area of a no-tillage production field. Two plants were collected from each of four 3 m by 9 m plots from a field near Elsie, Michigan that had been in a no-tillage cornsoybean rotation for 10 years. Entire plants (above and below ground biomass) were randomly collected from each of the four plots and maintained in the greenhouse. Genetic analysis was conducted using the original plant collected from the field.

Among-progeny genetic variation

The apomictic nature of this species was examined by collecting mature seeds from a single flower and conducting the RAPD analysis on 10 sibling progeny and the maternal plant. Common dandelion seedlings were established and maintained in the greenhouse for this analysis. Collections selected for this evaluation included common dandelion from Michigan, Oregon, and Germany.

DNA extraction and RAPD analysis

Genomic DNA was extracted from the newest leaf material emerging from plants growing either in the greenhouse or field nursery. DNA was extracted from four 10 mm diameter leaf disks (approx. 45 mg fresh leaf tissue) using the protocol described with the PUREGENE[™] DNA isolation kit² (Appendix A1) and stored in Tris-EDTA (TE) buffer (pH 7.0). DNA concentration was determined by

visual comparison with a known quantity of DNA mass ladder³ on an agarose gel stained with 0.1 µg ml⁻¹ ethidium bromide. The presence of an unidentified PCR inhibitor required a 1:20 dilution (concentrated DNA:TE) of DNA be conducted prior to PCR amplification. This resulted in a DNA concentration of less than 20 ng µl⁻¹. The PCR primers utilized were 10-base pair (bp) random oligonucleotides from primer kit A⁴. Each PCR reaction was carried out in a 25 µl reaction volume consisting of 50 ng genomic DNA, 2.5 µg bovine serum albumin (BSA), 5.0 mM MgCl₂, 3.2 mM Tris-HCl (pH 8.4), 50 mM KCl, 0.2 mM each deoxynucleotide triphosphate (dNTP), 1.2 pmol 10 base pair (bp) oligonucleutide primer, and 0.1 units Tag DNA polymerase⁵. PCR reactions for each random primer were conducted at least twice for each plant sample in a heated-bonnet thermal cycler⁶ programmed for an initial denaturation temperature of 94 C for 5 min followed by 35 cycles of 1 min 15 sec at 94 C, 1 min 15 sec at 40 C, and 2 min at 72 C. The final cycle was followed by 3 min at 72 C, after which the temperature was held at 4 C until gel electrophoresis. A 10 µl aliquot of the PCR product was loaded with DNA loading dye [50% glycerol, 0.25% bromophenol blue, 10 mM Tris HCI (pH 8.0), 1 mM EDTA] onto a 2.0% (w/v) agarose⁷ gel stained with 0.1 µg ml⁻¹ EtBr. Amplified products were resolved at 80 volts for 3 hr in a 1X Trisacetate (TAE) buffer (40 mM Tri-acetate, 1 mM EDTA). A 100 bp DNA ladder⁸ was used as a size reference. The gel was viewed and photographed on an ultraviolet light box to confirm product amplification. Polymorphic PCR fragments were scored as either present (1) or absent (0). Only those fragment length polymorphisms that were repeatable and intensely amplified were scored.

Comparison of plant size

A greenhouse experiment was conducted to compare plant sizes of selected common dandelion collections. Common dandelion from eight counties in Michigan and one county in Illinois were selected for this experiment. Collections were selected based on the results from the RAPD analysis. Plant collections were selected to represent both genetically similar and dissimilar collections. Mature seed was collected from the respective collections in the field nursery and stored at 4 C until planted in the greenhouse. Common dandelion seeds were planted 0.25 cm deep and seedlings individually transplanted to 1000 ml pots containing commercial potting mixture approximately 2 weeks later. Greenhouse temperatures were maintained at 30/25 ± 3 C (day/night) with 14:10 h (day:night) photoperiod. Supplemental light intensity from sodium vapor lamps provided a total midday light intensity of 1,000 µmol m⁻² s⁻¹ photosynthetic photon flux at plant height. Common dandelion plants were watered as needed and fertilized with 50 ml of N, P₂O₅, K₂O (20%-20%-20%) at 20 ppm to promote optimum plant growth.

Comparison of plant size for 9 common dandelion collections was conducted 60 days after planting. Plant collections were compared by measuring total leaf area and plant dry weight. Leaf area was measured with a transparent belt conveyor accessory for a portable leaf area meter⁹. Dry weight was determined for the above ground biomass; harvested plant material was dried at 70 C for 24 h.

Statistical analysis

11

Common dandelion collections were established in the field nurseries in a randomized complete block design and each collection was replicated four times at each of the nurseries. Data were subjected to analysis of variance with SAS¹⁰ and means separated using Fisher's Protected LSD ($\alpha = 0.05$). Nursery by collection interactions were significant; therefore data from each nursery location were analyzed and presented separately.

Genetic similarity coefficients between common dandelion collections were determined using Nei and Li's (1979) calculation for qualitative data. Dendograms for genetic distance were created using the unweighted pair group method with arithmetic averages (UPGMA) cluster analysis. Genetic similarity calculations and dendograms were made using NTSYS-pc version 2.11L software¹¹ (Rohlf 2002). Collections were compared using calculated genetic similarity coefficients where 0.00 indicated no similarity and 1.00 indicated that the collections were identical.

The experiment to compare plant size was conducted as a completely randomized design. Each plant collection was replicated four times and the experiment was conducted twice. Data were subjected to analysis of variance with SAS and means were separated using Fisher's Protected LSD ($\alpha = 0.05$). Variances were determined to be homogenous, thus the experiments combined.

RESULTS AND DISCUSSION

Phenotypic variation among collections

Following the winter of 2002-03 it was observed that all of the plants established in the Chatham nursery survived, whereas mortality was observed for some collections in East Lansing. However, mortality was no more than one plant from any collection. The one exception was the common dandelion collection from Oceana Co. Michigan. As a result it was dropped from the analysis of the East Lansing nursery. At the Chatham station in the Upper Peninsula of Michigan, the mean annual snow fall is 380 cm. This snow cover insulated the nursery, allowing these plants to survive the winter. The lack of snow fall and extreme cold temperature at the East Lansing nursery in 2002-03 may explain the common dandelion mortality and lower plant vigor in the 2003 growing season.

Some of the characteristics measured, such as leaf shape and the presences of pubescence on the leaf were variable between plants as well as on an individual plant. Leaf shape on an individual plant was highly variable, resulting in difficulty identifying differences in leaf shape between collections (data not shown). Previous research using common dandelion leaf morphology not only found leaf shape to be highly variable but also found that it was influenced by the environment and even varied between seasons (Sturtevant 1886; Taylor 1987). The presence of pubescence on the leaf surfaces appeared to be related to the age of the leaf. Newly emerging leaf material for all of the

collections was typically pubescent. In contrast, the older leaf tissue lacked pubescence, regardless of the collection (data not shown).

The common dandelion plants at the Chatham nursery were much smaller in diameter compared with East Lansing (Table 2). A common dandelion collected from Tolland Co. Connecticut was the largest in diameter at both East Lansing and Chatham with 55 cm and 22 cm, respectively. At Chatham, common dandelion from Hall Co. Nebraska was among the largest; however it was one of the smaller collections in East Lansing. The common dandelion collection from Germany was the smallest in diameter at both of the nurseries.

The date at which common dandelion began to flower was determined to be different at each of the nurseries; however differences within the nurseries were not apparent. The common dandelion collections at the East Lansing nursery initiated flowering within 1 week of each other beginning on May 1, 2003 at an accumulation of 239 growing degree days (data not shown). This coincides with previous research that classified common dandelion as a day-neutral plant (Gray et al. 1973). At Chatham, flower initiation commenced approximately 3 weeks later.

There were no significant differences observed in the total number of flowers produced or the total number of seeds produced per collection (Appendix A2). However, there were differences observed in the number of seeds produced per flower among collections (Table 2). Common dandelion in the East Lansing nursery tended to produce more seeds per flower than at Chatham. Seed production ranged from 106 to 230 seed per flower at Chatham and 123 to 304

seeds per flower in East Lansing. This difference in productivity is likely due to the greater accumulation of growing degree days at the East Lansing nursery as compared to the Chatham nursery, which was located at a more Northern latitude. At both nurseries, common dandelion from Baker Co. Oregon and Cache Co. Utah were the most prolific producers of seeds. The Alger Co. collection, which was collected on the Chatham station itself, was one of the more prolific plant collections at both of the nurseries. The common dandelion collections from Hall Co. Nebraska and Germany were the least prolific at each nursery.

Genetic variation among collections

Successful amplification of PCR products was dependent on the random primer used. Of the 20 primers screened, 9 primers resulted in the amplification of a DNA fragment. The nine random primers amplified a total of 71 repeatable DNA fragments, of which 44 were polymorphic (Table 3). The number of polymorphic fragments amplified per random primer ranged from 1 to 12. Of the 26 populations screened, 24 were distinguishable from each other using the RAPD analysis. Common dandelion from Berrien and Calhoun counties in Michigan were indistinguishable from each other.

The diversity of RAPD banding patterns observed suggest that there is a high level of genetic diversity in common dandelion in Michigan and the other states. Genetic similarity coefficients among all collections ranged from 0.25 to 1.00 (Table 4). Within Michigan, genetic similarity ranged from 0.27 to 1.00.

There was no discrete separation among common dandelion collections from Michigan and those collected from other states or Germany. However, common dandelion from Michigan tended to be more similar to other Michigan collections than with collections from the other states (Figure 1). Most of the Michigan collections were grouped together in the dendogram, with a few of the counties appearing more closely related to common dandelion from other states. Clustering of collections within the dendogram indicate more similarity among those collections than others outside the cluster. The amount of genetic variation observed here is similar to that observed for other weed species. For example, RAPD analysis of wild mustard (Sinapis arvensis) was found to be highly variable (Moodie et al. 1997). In addition, analysis of wild mustard plants sampled over two consecutive seasons showed different levels of population diversity, suggesting the influence of environmental variability. Isozyme analysis of two perennial species of snakeweed (Gutierrezia spp.) indicated a high level of diversity both within species and between species (Sterling and Hou 1997).

Common dandelion from three Michigan counties and two states were identifiable with unique DNA banding patterns (data not shown). The presence of a single unique DNA fragment is associated with the collections from Stafford Co. Kansas, Benton Co. Indiana, and Presque Isle and Clinton Counties in Michigan using the random primers OPA-8, 9, 11, and 18, respectively. The absence of the 675 bp fragment amplified by OPA-18 was unique to the Luce Co. collection. Several additional collections shared either the presence or the absence of two random DNA fragments (data not shown). A single 1000 bp random DNA

fragment amplified using OPA-18 was exclusive only to the collections from Berrien, Calhoun, Hillsdale, and Ingham Counties in Michigan.

Geographical location and genetic similarity did not appear to be related when comparing common dandelion collections. Many of the Michigan collections, which are in relatively close geographic proximity, tended to be genetically similar. However, an exception to this trend included the Ingham Co. collection that was genetically more similar to Adams Co. Colorado (0.69) and Benton Co. Indiana (0.69) than to any collection from Michigan.

The common dandelion from Presque Isle Co. Michigan, Stafford Co. Kansas and Germany were identified from their seed color as red seeded dandelion. No RAPD polymorphisms were identified that were unique to red seeded dandelion. Single polymorphic fragments from different random primers were amplified that were unique to the Presque Isle Co. and Stafford Co. collections. Random primer OPA-09 amplified a 450 bp fragment that was unique to Stafford Co., whereas OPA-18 amplified a 650 bp fragment in the Presque Isle Co. collection only. Random primer OPA-07 failed to amplify an 875 bp fragment in either the Stafford Co. or Presque Isle Co. collections. The Stafford Co. or Presque Isle Co. collections. The Stafford Co. or Presque Isle Co. collections. The Stafford Co. or Presque Isle Co. collection was genetically more similar to the collection from Germany than to Presque Isle Co., 0.54 and 0.37, respectively. Common dandelion collected from Stafford Co. and Germany were grouped together in the dendogram (Figure 1), indicating they shared more unique DNA fragment length polymorphisms. The

Presque Isle Co. collection was more similar to other Michigan collections of *T*. *officinale* then the other two *T*. *laevigatum* collections.

Within-field genetic variation

The common dandelions that were collected from the no-tillage production field near Elsie, Michigan demonstrated a broad range of genetic similarity. Coefficients of genetic similarity for the eight plants collected ranged from 0.30 to 1.00 (data not shown). Two of the plants were indistinguishable from each other. A coefficient of genetic similarity of approximately 0.80 was calculated for three of the collected plants from across the area. Plants that were collected from the same 3 m by 9 m plots were not necessarily more related to each other. The level of diversity observed here is similar to that previously reported. Using isozymes and plant growth competitiveness, Solbrig and Simpson (1974) identified the presence of at least four common dandelion biotypes within an area of 0.01 ha.

Among-progeny genetic variation

RAPD analysis conducted on ten progeny from each of the three collections selected did not reveal any genetic variation between the progeny and the maternal plant using the 9 random primers. This observation supports that common dandelion is apomictic, at least with the individuals tested. King and Schaal (1990) screened the rDNA of over 700 progeny from 26 different parental genotypes and observed 42 plants with nonparental rDNA. This rate is higher

than what would be expected by mutation alone. Results from this study indicate that a large number of individuals are needed to find genetic differences.

Comparison of plant size

The nine collections in this experiment varied in their respective rate of growth after 60 days. Overall the dry weights of the 9 common dandelion collections ranged from 0.84 to 2.0 g per plant (Table 5). Total leaf area ranged from 216 to 432 cm². The Michigan collections from Alger, Monroe, and Shiawassee Counties had the greatest leaf area and dry weights at the end of the experiment. Conversely, the collections from Berrien, Luce, and Newaygo Counties in Michigan were the lowest in terms of leaf area and dry weight. The collection from Alger Co. demonstrated the greatest growth rate as measured by leaf area and dry weight. This collection was genetically most similar to the Ingham Co. collection with a coefficient of 0.61 (Table 4). These two collections had similar leaf areas but were different in terms of dry weight. The collection from Alger Co. was the least genetically similar to the common dandelion from St. Clair Co. and had a higher leaf area and dry weight (Table 5). The Shiawassee Co. and Berrien Co. collections were genetically the most similar (0.87) but differed in their growth rates. Conversely, the Vermillion Co. and Ingham Co. collections were the least genetically similar (0.35) but were similar in size. The lack of an obvious relation between genetic similarity and plant size indicate that the polymorphisms identified were not associated with traits influencing plant development.

The common dandelion collections in this analysis demonstrated a high level of morphological and genetic variability. From the RAPD analysis we did not identify distinctly unique biotypes of common dandelion. Similarity was observed between collections within a geographical region but there were no discrete boundaries. Genetic similarity did not appear to be related to similarity in morphological characteristics. These characteristics measured in the common dandelion nurseries are likely to be quantitatively inherited traits that are controlled by more than one gene. In addition, the numbers of polymorphisms used here to quantify the genetic diversity were insufficient to identify a relation between phenotype and genotype.

The high level of variability in common dandelion observed in this research could possibly be a result of the method of seed dissemination in this species. Mature seeds attached to white pappus are capable of long distance travel, spreading the genetic diversity across a large area. In addition, common dandelion become established across a wide range of climates and geographical regions, as is evident from its distribution throughout the world. And finally, the high level of genetic and morphological diversity observed will make population-specific management of common dandelion unlikely.

Source of Materials

¹Baccto, Michigan Peat Co, P.O. Box 98029 Houston TX, 77098

² Puregene DNA Isolation Kit, Gentra Systems, Minneapolis, MN 55441.

³ Low DNA Mass Ladder, Invitrogen Life Technologies, Carlsbad, CA 92008.

⁴ Primer Kit A, Operon Technologies, Inc., Alameda, CA 94501.

⁵ Taq DNA polymerase, Invitrogen Life Technologies, Carlsbad, CA 92008.

⁶ PTC-225 Peltier Thermal Cycler, MJ Research Inc., Waltham, MA 02451.

⁷ Agarose, Invitrogen Life Technologies, Carlsbad, CA 92008.

⁸ 100 base pair ladder, Invitrogen Life Technologies, Carlsbad, CA 92008.

⁹ Portable leaf area meter, Li-Cor Inc., Lincoln, NE 68504.

¹⁰ SAS version 8.2, SAS Institute, SAS Circle, Box 8000, Cary, NC 27512-8000.

¹¹ NTSYS-pc ver. 2.11L software, Exeter Software, Setauket, NY 11733-2870.

LITERATURE CITED

GPFA (Great Plains Flora Association). 1996. Flora of the Great Plains. Lawrence, KS: University Press of Kansas. Page 1010.

Gray, E., E. M. McGehee, and D. F. Carlisle. 1973. Seasonal variation in flowering of common dandelion. Weed Sci. 21:230-232.

Jorge, S., M. C. Pedroso, D. B. Neale, and G. Brown. 2003. Genetic differentiation of Portuguese tea plant using RAPD markers. HortScience 38:1191-1197.

King, L. M. 1993. Origins of genotypic variation in North American dandelions inferred from ribosomal DNA and chloroplast DNA restriction enzyme analysis. Evolution 47:136-151.

King, L. M. and B. A. Schaal. 1990. Genotypic variation within asexual lineages of Taraxacum officinale. Evolution 87:998-1002.

Moodie, M., R. P. Finch, and G. Marshall. 1997. Analysis of genetic variation in wild mustard (*Sinapis arvensis*) using molecular markers. Weed Sci. 45:102-107.

Nei, M. and W. H. Li., 1979. Mathematical model for studying genetic variation in terms of restriction endonucleases. Proc. Natl. Acad. Sci. USA, 76:5269-5273.

Orel, G., A. D. Marchant, J. A. McLeod, and G. D. Richards. 2003. Characterization of 11 Juglandaceae genotypes based on morphology, cpDNA, and RAPD. HortScience 38:1178-1183.

Ransom, C. V., D. D. Douches, and J. J. Kells. 1998. Isozyme and RAPD variation among and within hemp dogbane (*Apocynum cannabinum*) populations. Weed Sci. 46:408-413.

Richards, A. J., 1970. Eutriploid facultative agamospermy in *Taraxacum*. New Phytol. 69:761-774.

Richards, A.J., 1973. The origin of *Taraxacum* agamospecies. Bot. J. Linn. 6:189-211.

Rohlf, F. J., 2002. NTSYSpc: Numerical Taxonomy System, Version 2.11L. Exeter Publishing, Ltd.: Setauket, NY.

Rowe, M. L., D. J. Lee, S. J. Nissen, B. M. Bowditch, R. A. Masters. 1997. Genetic variation in North American leafy spurge (*Euphorbia esula*) determined by DNA markers. Weed Sci. 45:446-454.

Solbrig, O. T., 1971. The population biology of dandelions. Am. Sci. 59:686-694.

Solbrig, O. T., and B. B. Simpson. 1974. Components of regulation of a population of dandelion in Michigan. J. Ecol. 62:473-486.

Sterling, T. M. and Y. Hou. 1997. Genetic diversity of broom snakeweed (*Gutierrezia sarothrae*) and threadleaf snakeweed (*G. Microcephala*) populations. Weed Sci. 45:674-680.

Stubbendieck, J., G. Y. Friisoe, M. R. Bolick. 1995. Weeds of Nebraska and the Great Plains. Lincoln, Nebraska: Nebraska Department of Agriculture. pp. 176-177

Sturtevant, E. L. 1886. A study of the dandelion. Amer. Nat. 20:5-9.

Taylor, R. J., 1987. Population variation and biosystemic interpretations in weedy dandelions. Bull. Torrey Bot. Club 114:109-120.

Williams, J. G. K., A. R. Kubelik, K. J. Livak, J. A. Rafalski, and S. V. Tingey. 1990. DNA polymorphisms amplified by arbitrary primers are useful as genetic markers. Nucleic Acid Res. 18:6531-6535.

Collection	Site	Site Description ^a	Included in field nurseries	Included in genetic analysis
1	Alger Co. MI	dairy pasture	х	х
2	Berrien Co. MI	NT agriculture	х	×
3	Calhoun Co. MI	NT agriculture	x	×
5	Clinton Co. MI	NT agriculture		х
6	Hillsdale Co. MI	NT agriculture	х	×
7	Ingham Co. MI	residential	x	x
9	Ionia Co. MI	NT agriculture	х	х
12	losco Co. MI	state park	х	х
13	Leelanau Co. MI	state park		х
14	Monroe Co. MI	NT agriculture	х	х
15	Newaygo Co. MI	CT agriculture	х	х
17	Oceana Co. MI	fruit orchard	х	x
18	Presque Isle Co. MI ^b	state park		x
20	Shiawassee Co. MI	CT agriculture	х	x
21	St. Clair Co. MI	CT agriculture		x
22	Luce Co. MI	state park	х	x
23	Yolo Co. CA	fruit orchard	х	x
26	Adams Co. CO	sod farm	х	х
27	Tolland Co. CT	dairy pasture	х	х
28	Champaign Co. IL	wooded area	х	
29	Vermillion Co. IL	CT agriculture	х	x
30	Benton Co. IN	CT agriculture		х
31	Riley Co. KS	CT agriculture	х	
32	Stafford Co. KSb	CT agriculture		х
33	Hall Co. NE	residential	х	х
36	Baker Co. OR	pasture	х	
37	Elk Co. PA	road side	х	
39	Cache Co. UT	residential	х	х
40	Brazos Co. TX	residential	х	х
42	Germany ^b	unknown	х	х

Table 1. Location and site description of common dandelion collections included in the field nurseries for RAPD analysis.

^a Abbreviations: NT = no-tillage; CT = conventional tillage.
^b Plants collected from these sites were identified as red seeded dandelion (*T. laevigatum*).

	Number of	of seeds	Rosette d	liameter
Collection	East Lansing	Chatham	East Lansing	Chatham
	seeds pe	r flower	cn	ı
Alger Co. MI	275	200	42.2	15.6
Berrien Co. MI	232	208	37.8	20.6
Calhoun Co. MI	230	209	47.0	19.7
Hillsdale Co. MI	234	179	41.4	13.4
Ingham Co. MI	230	202	37.4	24.1
Ionia Co. MI	207	153	38.1	14.6
losco Co. MI	206	148	52.1	19.1
Leelanau Co. MI	223	198	44.5	17.8
Monroe Co. MI	187	153	31.8	19.4
Newaygo Co. MI	152	153	40.3	14.3
Oceana Co. Ml ^a	-	185	-	10.8
Shiawassee Co. MI	168	153	33.4	21.3
Luce Co. MI	183	142	36.2	11.5
Yolo Co. CO	173	132	31.7	17.8
Adams Co. CO	293	180	38.9	15.0
Tolland Co. CT	245	214	54.6	24.9
Champaign Co. IL	163	166	32.2	20.0
Riley Co. KS	212	230	38.8	16.5
Hall Co. NE	145	131	26.0	22.3
Baker Co. OR	304	222	38.5	19.4
Elk Co. PA	178	199	42.2	21.2
Cache Co. UT	261	215	35.6	14.9
Brazos Co. TX	168	145	33.0	21.3
Germany	123	106	22.0	12.2
LSD(0.05)	56	29	13.9	6.1

Table 2. Seed production and plant diameter for common dandelion at East Lansing and Chatham field nurseries.

^a Oceana County collection dropped from East Lansing nursery due to winter mortality

RAPD Primer	Sequence 5' to 3'	No. of bands	No. polymorphic
OPA-03	AGTCAGCCAC	4	1
OPA-04	AATCGGGCTG	14	8
OPA-07	GAAACGGGTG	10	8
OPA-08	GTGACGTAGG	12	11
OPA-09	GGGTAACGCC	5	2
OPA-10	GTGATCGCAG	6	2
OPA-11	CAATCGCCGT	3	2
OPA-18	AGGTGACCGT	10	6
OPA-19	CAAACGTCGG	7	4
Total		71	44

Table 3. RAPD primers used to evaluate the genetic diversity between common dandelion collections.

							Common dandelion collections	n dande	elion co	llections	10					
Collection	-	2	з	5	9	7	6	12	13	14	15	17	18	20	21	22
01 Alger Co. MI	1.00															
02 Berrian Co. MI	0.56	1.00														
03 Calhoun Co. MI	0.56	1.00	1.00													
04 Clinton Co. MI	0.50	0.61	0.61	1.00												
06 Hillsdale Co. MI	0.73	0.67	0.67	0.41	1.00											
07 Ingham Co. MI	0.61	0.67	0.67	0.38	0.65	1.00										
09 Ionia Co. MI	0.58	0.84	0.84	0.69	0.62	0.56	1.00									
12 losco Co. MI	0.42	0.50	0.50	0.29	0.45	0.40	0.52	1.00								
13 Leelanau Co. MI	0.93	0.60	0.60	0.53	0.71	0.58	0.62	0.36	1.00							
14 Monroe Co. MI	0.59	0.59	0.59	0.83	0.46	0.47	0.67	0.34	0.63	1.00						
15 Newaygo Co. MI	0.58	0.58	0.58	0.63	0.55	0.56	0.53	0.43	0.62	0.61	1.00					
17 Oceana Co. MI	06.0	0.58	0.58	0.57	0.69	0.56	0.60	0.35	0.97	0.67	0.60	1.00				
18 Presque Isle Co. MI	0.58	0.58	0.58	0.34	0.55	0.56	0.53	0.35	0.62	0.39	0.47	0.60	1.00			
20 Shiawassee Co. MI	0.60	0.87	0.87	0.65	0.64	0.58	0.97	0.55	0.64	0.63	0.55	0.62	0.55	1.00		
21 St. Clair Co. MI	0.50	0.57	0.57	0.56	0.46	0.55	0.67	0.50	0.54	0.61	0.52	0.52	0.44	0.69	1.00	
22 Luce Co. MI	0.53	0.53	0.53	0.47	0.43	0.52	0.48	0.27	0.57	0.51	0.69	0.55	0.48	0.50	0.54	1.00
23 Yolo Co. CA	0.55	0.69	0.69	0.55	0.52	0.53	0.71	0.57	0.59	0.65	0.64	0.57	0.50	0.74	0.64	0.59
26 Adams Co. CO	0.50	0.57	0.57	0.44	0.62	0.69	0.59	0.40	0.46	0.42	0.52	0.44	0.44	0.62	0.42	0.31
27 Tolland Co. CT	0.63	0.56	0.56	0.44	0.53	0.61	0.58	0.67	0.60	0.59	0.52	0.58	0.52	0.60	0.50	0.33
29 Vermillion Co. IL	0.56	0.51	0.51	0.79	0.49	0.35	0.58	0.39	0.59	0.86	0.58	0.63	0.42	0.54	0.57	0.43
30 Benton Co. IN	0.58	0.52	0.52	0.40	0.48	0.69	0.47	0.35	0.55	0.39	0.73	0.53	0.47	0.48	0.44	0.62
32 Stafford Co. KS	0.50	0.57	0.57	0.50	0.38	0.41	0.59	0.30	0.54	0.48	0.44	0.52	0.37	0.62	0.50	0.46
33 Hall Co. NE	0.61	0.56	0.56	0.80	0.53	0.38	0.63	0.36	0.65	0.88	0.63	0.63	0.40	0.59	0.63	0.47
39 Cache Co. UT	0.47	0.67	0.67	0.47	0.57	0.65	0.69	0.45	0.43	0.40	0.48	0.41	0.34	0.71	0.46	0.36
40 Brazos Co. TX	0.58	0.53	0.53	0.76	0.50	0.41	0.59	0.40	0.61	0.88	0.59	0.59	0.43	0.56	0.59	0.44
42 Germany	0 53	0 53	0 53	11	000		0	000	11	TL C		000			1	

Table 4. Matrix of genetic similarity coefficients for RAPD analysis of 26 common dandelion collections.

ZU

Table 4 (cont'd). Matrix of genetic similarity coefficients for RAPD analysis of 26 common dandelion collections.

				Commo	in dand	elion co	Common dandelion collections	(0		
Collection	23	26	27	29	30	32	33	39	40	42
01 Alger Co. MI										
02 Berrian Co. MI										
03 Calhoun Co. MI										
04 Clinton Co. MI										
06 Hillsdale Co. MI										
07 Ingham Co. MI										
09 Ionia Co. MI										
12 losco Co. MI										
13 Leelanau Co. MI										
14 Monroe Co. MI										
15 Newaygo Co. MI										
17 Oceana Co. MI										
18 Presque Isle Co. MI										
20 Shiawassee Co. MI										
21 St. Clair Co. MI										
22 Luce Co. MI										
23 Yolo Co. CA	1.00									
26 Adams Co. CO	0.48	1.00								
27 Tolland Co. CT	0.62	0.50	1.00							
29 Vermillion Co. IL	0.50	0.34	0.56	1.00						
30 Benton Co. IN	0.50	0.52	0.52	0.37	1.00					
32 Stafford Co. KS	0.48	0.25	0.50	0.46	0.44	1.00				
33 Hall Co. NE	0.55	0.38	0.56	0.93	0.40	0.50	1.00			
39 Cache Co. UT	0.52	0.85	0.47	0.38	0.48	0.38	0.35	1.00		
40 Brazos Co. TX	0.57	0.35	0.63	0.93	0.38	0.47	0.95	0.33	1.00	
42 Germany	0.59	0.31	0.40	0.43	0.48	0.54	0.41	0.36	0.39	1.00

Collection	Leaf area ^a	Dry weight ^a
	cm ³	g
Alger Co. MI	432 a	2.02 a
Berrien Co. MI	244 cd	0.84 c
Ingham Co. MI	394 ab	1.56 b
Luce Co. MI	216 d	0.87 c
Monroe Co. MI	387 ab	1.79 ab
Newaygo Co. MI	266 cd	0.97 c
Shiawassee Co. MI	409 a	1.74 ab
St. Clair Co. MI	319 bc	1.57 b
Vermillion Co. IL	310 bc	1.51 b

Table 5. Comparison of leaf area and dry weight for 9 collections of common dandelion grown in the greenhouse.

^a Means followed by the same letter within column are not significantly different according to Fisher's Protected LSD (α =0.05).



