Seed Collection Strategy Rationale

A lot of the literature reviewed pertained to endangered plants and recommendations by the Center of Conservation for Endangered Plants. The methods and rationale in most cases will be relevant for the collection of the warm season grasses on Long Island. Although warm season grasses are rare as a plant community on Long Island the reproductive biology and ecology of these plants would not lead to their classification as endangered at the species level. Some of the recommendations developed were based on economy of labor and cost to insure the rapid conservation of the most endangered species. Some recommendations in the literature pertain to regions of variable elevation and rainfall existing in short linear distances which is found in the Western region of the US. The relative environment we are collecting from and selecting for is relatively more uniform than the Western region. Some of the information found in the Source-Identified Seed: the Iowa Road Side Experience was a collaborative effort with the Elsberry Missouri Plant Materials Center and other groups and would pertain to the Long Island Initiative except for references to longitudinal differences.

For *ex situ* samples to be of maximum conservation value they must be genetically representative, thus containing the allelic diversity and evolutionary potential of the original natural population from which they were collected.

Many endangered species survive in only one to five populations, (USFWS 1992) out of 91species the modal number of populations was one. The median population size of the endangered plants at the time of listing by USFWS was less than 120 individuals.

A first step is to find sites that are, to the best of our knowledge, not the result of or contaminated by non-native germplasm.

How many populations to sample per species:

CPC (Center of Conservation) recommended 1-5 populations/species, the degree of genetic difference among population would have a bearing on this number. In general, more population should be sampled from species with high diversity or when gene flow between populations is low (fragmented broken corridors) open pollinated species like warm season grasses are more diverse then self pollinators. Factors which suggest high among population diversity is noticeable ecotype or habitat differentiation, or isolated populations. We should collect from as many "pure" populations we can find.

Breeding systems: inbreeders (self pollinators i.e like wild ryes) typically have less total genetic variation than do outbreeders (open pollinatora), but more of what little variation they do have is distributed among populations. Thus to acquire a given proportion of a species total variation, a higher proportion of an inbreeders populations will probably have to be sampled. (Hamrick, J.L. 1989).

Out of 653 published studies across breeding systems of 449 sp. 165 genera 78% of allozyme diversity occurs within populations. (J.L. Hamrick and M.W. Godt, 1989)

In the LI situation the number of populations sampled will depend on the number of populations found that we have confidence are not contaminated by out of region germplasm. If enough are found than we need to assess labor and time requirements and choose those populations which have the greatest ecotype or habitat differentiation. It may be desirable to collect in additional years if labor and time is an issue or if other sites or permissions are obtained.

How many individuals should be sampled per population:

10-50 individuals, the allelic content of a sample is proportional to the logarithm of both the population size and the sample size. Therefore a strong law of diminishing returns is in operation. Monomorphic alleles and those in relatively high frequency are captured in early samples and the chance of obtaining rare alleles declines rapidly with increasing sample size. In a statistical sense, the first 10 individuals are as important as the next 90. Considerations are population size, and degree of genetic communication among individuals within the population. (A.D. Brown and J.D. Briggs, 1991).

In practice if there are 1000's of individuals it may be advisable to collect in groups keeping them in separate bags. Collect seeds from more individuals if there is a high diversity among individuals within the population. Indicators include: high microsite variation within population, self-incompatibility (outcrossing) or restricted genetic neighborhoods (fragmentation).

Collect from a distance away to avoid relatedness and vegetative reproduction and increase chance for more diversity.

Collect from differing microsites, soils, hydrologic conditions, position in landscape, aspect sun/shade and salt conditions, and but not in this case elevation.

Identify sites using simple GPS lat/long or UTM units to be able to later get soils information or to come back to collect more information or more seeds.

How much seed to collect per individual:

For endangered plants recommendations were for 1-20 seeds per individual. Common sense, driven by expected survivorship of the seed both in cultivation and in the wild, "Collect more if being used for reintroduction or restoration program." We can collect more since we are not dealing with endangered plants. Genetic variation is best maintained when each founder individual contributes equally to a new population. In practice, we can equalize the contribution of different female parents of seeds we collect simply by keeping seeds from each plant or cluster of plants separate. This also offsets the need for larger bulk collections.

Self fertilizing species collect from more populations, fewer per population less variation among individuals within each population.

Multi-year collection:

For rare and endangered species when sampling itself could have an effect on the short term survival prospects of the sampled population. Decision based on intuitive guides, based on life history, how many years of successful seed set and germination, whether the habitat is being improved or impaired. Multi-year collecting may be good for the LI initiative from the basis of getting more genetic material, effects of weather on seed set, and to collect seed at different time (this could also be done by collecting twice in same year). If we don't get all permissions and miss bulk of seed set. I don't think there is an endangerment issue with these species and the reproductive biology of the warm season grasses indicate good seed production, viability and an existing seed bank in some locations.

Rationale of collection strategy for endangered plants and other comments:

"There is always a tradeoff to be made between the potential benefits resulting from better knowledge and the losses resulting from inaction in the absence of perfect knowledge"

The guidelines were designed to replace an expensive, information greedy, genetic inventory of each taxon (e.g. using allozyme or DNA sequencing data) with a set of standardized, expedient but scientifically based sampling procedures.

Populations need to be reestablished in a manner that minimizes the risk of rapid extinction due to demographic and environmental stochasticity (randomness). A reason to collect a lot of plants is to capture as much genetic diversity as possible.

Need to establish population large enough to avoid loss of alleles due to random genetic drift and increased inbreeding associated with even short bottleneck in population size. Population need to be larger than previously estimated.

Accumulation of even mildly deleterious mutations appears to be a much greater threat to small populations than previously thought. Theoretical model sizes smaller than 1000 are seriously threatened by mutational melt down after 100 generations.

Intentional subdivision of propagation material (seed) among reintroduction sites can offset the effects of genetic drift in a single closed population. A greater range of selection forces among sites and the improbability of drift having the same effect in all populations are likely to allow more of the total genetic variation to be preserved.

Issues during seed increase:

One simple way of avoiding selection in captivity (agricultural or garden setting) is to release propagules directly into protected prospective sites within historic range. The founding gene pool will be molded by conditions in situ instead of in garden. Another method is to harvest by hand each plant in the garden and use equal amounts of seed this doesn't take into account germination and pure live seed. Harvesting extra seed in situ to include with increased seed is a compromise.

Garden grown material can be used to create a desirable age or stage structure in a reintroduced population that may lower the probability of near term expiration more important for plants which are endangered and are having problems being established.

It is very important to keep the integrity of each collection (accession). A collection can be an individual plant or from a group of plants that are in close proximity

since they are probably genetically related. The number of plants to grow out from each collection/accession will depend on the size of the increase block and the number of accessions you are dealing with. Another consideration is the number of plants seed was collected from to make the accession if one accession came from 5 plants and another came from 50 there could be a need to grow out more from the 50 if we want to try and capture the same gene frequency as found in the wild. This is another reason not to collect from 100's of plants and put the seed into one bag and call it an accession. Also the size of the increase block based on availability of space, equipment and labor. The plants should be randomly spaced in the nursery to allow for random recombination during pollination. Another method is to block the seed increase block and randomize each subblock this is important when there is soil or moisture differences in the field.

The Iowa project seemed to have a lot more collection sites than I think we have and they used each population as a collection (accession) "collecting a small portion of seed from many individuals in each population" I think that there is more uniformity within their collection sites and there is even less topographical relief and trees breaking up wind pollination than what I saw at the Roosevelt Park Location. The other sources of information spoke of collecting at the individual or cluster level. The Iowa project used a minimum of 10 populations and usually obtained more than 20. They grew out 50 plugs from each accession. Additional plugs could be grown out if there is an interest to grow any plants on the landfill site if any selection by that environment is desired for use in a separate "improved cv." for reclamation. Any extra seed could be used for restoration purposes and some can be saved in a cooler in case there is a problem with the initial seed increase field. Surplus seed from the original collections together with seed from the nursery plots can then be further increased by foundation seed producer. If we only have 5 locations that have a lot of site variation we can separate them more finely at the individual or cluster level to hopefully capture more diversity. The distances called for to separate wind pollinated plants in a seed production system is 900-1320 ft. so we would definitely want to keep clusters of plants that were isolated by that much distance separate.

Collect the seed in small brown paper bags put collection information on it including the GPS coordinates which would then be placed in well ventilated greenhouse to dry.

In Summary

- Find sites/populations which contain plants that are genetically pure and uncontaminated.
- Collect from as many populations as you can. Its better to have more sites/populations than a lot of individuals from a couple of sites, as long as the sites are native.

• Depending on distribution and variability of the microsite you can collect based on microsite differences, if there is not much, than collect by distances (900-1320ft) grouping plants, there will be some overlap if plants are contiguous.

• Collect up to 25% of the seed from the plants especially if there are a lot of plants the seed can always be used for a restoration projects and the plants are not rare and endangered. We should have at least 200 seeds from each collection.

• Keep plants and clusters collected separately get good GPS reading collect as much information as you can without bogging down the operation can always go back if properly recorded coordinates can have back up topo map points.

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