WHEAT Classification, Biodiversity and Issues for Conservation

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Wheat Classification, Evolution, Place of origin and Domestication and Morphology

Classification:

There are one quarter of a million species of higher plants; 3000 domesticated; 150 extensively. Ninety-eight percent of the food on earth is derived from these 150 plants. Wheat is the staple food for thirty five percent of the world and is grown on 240 million hectares annually. (Knott 1987).

Wheat is a member of the Grass family *Gramineae* (Poaceae) and the tribe *Triticeae (= Hordeae)* (Briggle and Reitz 1963) in which the one to several flowered spikelets are sessile and alternate on opposite sides of the rachis forming a true spike. Wheats (*Triticum*) and ryes (*Secale*) together with *Aegilops, Agropyron, Eremopyron* and *Haynalidia* form the subtribe Triticinae (Simmonds 1976).

Linnaeus in 1753 first classified wheat. In 1918 Sakamura reported the chromosome number sets (genomes) for each commonly recognized type; this was a turning point in *Triticum* classification for it separated wheat into three groups. Diploids had 14 chromosomes (n=7), tetraploids had 28 (n=14) and the hexaploids had 42 (n=21). Bowden includes *Aegilops* with *Triticum.*

Bread wheat is *Triticum. aestivum*. *T. durum* and *T. compactum* (club) are the other major species. All three are products of natural hybridization among ancestrals no longer grown commercially (Briggle 1967).

Evolution:

(Taken directly from Simmonds 1976) "The wild diploid species are presumably monophyletic in origin although they have diverged from each other. This is shown in the seed dispersal units and their ecological requirements and geographical distributions. Cytogenetic data confirms the taxonomic classification by showing that each diploid contains a distinct genome (Kihara 1954) The related chromosomes of the different genomes show little affinity with each other and do not pair regularly thus leading to complete sterility and isolation of the diploid species from each other. The polyploid species show classic evolution through amphidiploidy. They behave like genomic allopolyploids, so that their chromosomes pair in a diploid-like fashion and the mode of inheritance is disomic.

Each polyploid species can be unidentified as a product of hybridization followed by chromosome doubling. Since the different genomes are closely related (Morris and Sears 1967) polyploid wheats are segmental rather than typical genomic allopolyploids. The diploid like behavior of polyploid wheats is due to suppression of pairing of homoeologous chromosomes (related chromosomes of different genomes) by a specific gene. In hexaploid *T. aestivum* this gene is located on the long arm of chromosome 5 of genome B and is known as the 5B gene (Riley 1965). Plants deficient for this gene

behave like segmental allopolyploids; their homologous chromosomes pair and form multivalents. The development of the diploidizing mechanism has been critical for the evolution of the polyploid wheats and their domestication. By restricting pairing to completely homologous chromosomes, the diploidizing gene ensures regular segregation of the genetic material, high fertility and genetic stability. Synthetic polyploid wheats which do not contain this gene are partially sterile. In segmental allopolyploids permanent heterosis between homoealleles (homologous genes in different genomes) can be maintained.

Three groups of polyploids are recognized (Zohary and Fieldman 1962). Species in each group have one genome in common and differ in their other genomes. Polyploids of group A share the genome of diploid wheat, *T. monococcum*; those of group D share the genome of *T. tauschii* (=*Aegilops squarrosa*) and those of group D share the genome of *T. umbellulatum* (=*A. umbellulata*)".

The polyploids of each group resemble the morphology and seed dispersal characteristics of the diploid donor of the common pivotal genome. The cultivated polyploid wheats belong to group A. The arrowheaded shaped dispersal unit of wild diploid wheat *T. monococcum* var. *boeoticum* can be recognized in all the wild polyploids, and the non brittle ear of cultivated *T. monococcum* var. *monococcum* appears in all the cultivated polyploids. Group A polyploids contains the tetraploids *T. turgidum* (AABB), *T. timopheevi* (AAGG) and the hexaploid *T. aestivum* (AABBDD). *T. aestivum* (AABBDD) contains two genomes homologous with the A and B genomes of *T. turgidum*. The identification of the diploid donors of the B,G and D genomes has been the subject of intensive cytogenetic studies. The donor of D is identified as *T. tauschii* (Morris and Sears 1967).

The evolutionary advantage of polyploids over diploids is genetic flexibility and adaptability to a broad range of environments.

Classification of cultivated plants is complex because of the existence of two different classification concepts, namely botanical classification and agricultural classification which is based on agronomic characters of cultivars. From the breeder's point of view, classification of cultivated plants should reflect the degree of relationship between cultivated plants and wild or weedy relatives.(Hawkes 1980.)

In order to characterize relationships of cultivated plants, the variation and distribution patterns of wild, weedy and cultivated populations have to be studied (Vavilov, 1940). The cultivar concept was introduced in the first edition of the ICNCP (International Code of Botanical Nomenclature) and cultivar is now an established systematic category and are valid by simply being published with name and description in printed matter. If named landraces have to be considered cultivars, it is possible to select cultivars from cultivars by defining another mean and variation of characters and this has led to named selections.

Place of Origin and Domestication

Since the beginning of the twentieth century plant investigators have made expeditions to different parts of the world to study the geographic distribution of wild and cultivated species, subspecies and varieties of plants and to collect plants of potential use to agriculture. Russian scientist N.I. Vavilov travelled from 1922-33 and advanced the theory that the region of greatest diversity of varieties of a given species is probably the center of origin of that species. He did not consider the center of greatest varietal diversity as being an infallible index of its basic center of origin and he emphasized the necessity of distinguishing between primary centers of origin and centers of secondary differentiation. He regarded Ethiopia as a secondary center of differentiation of wheat.

Vavilov said Asia Minor was the origin of the diploid group, Abyssinia and North Africa of the tetraploid group and Central Asia of the hexaploid group. Gokgol also discovered in Anatolia an exceptional diversity of hexaploid wheats The place where *T. spelta* took its origin lies on the Upper Rhine. Vavilov said that where a great number of wheat types are found are held to be areas of wheat origin. In Asia Minor, wild forms of the diploid and tetraploid wheats are to be found.

Archaeological evidence shows wheat among the first cultivated plants and agriculture as a new way of living mostly founded on the cultivation of *Hordeum* and *Triticum*. The hybridization of *T. boeoticum* and *A. speltoides* is the presumed origin of the tetraploid group. Impressions of grains have been found in pottery excavated from Neolithic periods and carbonized wheats have been found at Jarmo (7th C. B.C.). (Peterson 1965). Emmer (*T. dicoccum,* syn. *T. turgidum* var. *dicoccum*) seems to be the main wheat that moved with people into Asia, Africa and Europe. (Renfrew 1973)

The domestication of crops refers to development of new forms of the plant which are more fitted for cultivation and threshing. Changes include adaptation of seed for easier sowing, often through the loss of hairs and spines on the seed coat, greater uniformity in crop growth and ripening resulting from uniform germination following loss of seed dormancy mechanisms, improved harvesting efficiency from the elimination of spontaneous shattering of the inflorescence or fruit with consequent seed dispersal, improved edibility from the elimination or reduction of toxic principles in that part of the plant which is used for food. Domestication has made the cultivated forms less fit for survival in the wild. Human societies, crop plants and domestic animals evolved together, and were interdependent in both their origins and development.

There are broad theories on evolutionary change in natural populations and a conservation program fails if it doesn't provide for continuing or future evolution of gene pools in natural habitats. Some programs are preservationist and others conservationist. In planning for evolution the mechanisms of evolutionary change should be considered. One is phyletic evolution, response to secular trends and environmental heterogeneity in space and episodic change giving rise to new phyletic lines. To delay the implementation of conservation and management until the complexities of evolution are understood spells doom (Frankel 1981). "Phyletic optimism" is the belief that natural populations have large reserves of genetic variation and given the need for rapid adaptation these

populations can adapt to all but the most catastrophic challenges by change, or phyletic evolution. (Frankel 1981).

Morphology:
(Taken directly from Schlomer 1960).

Inflorescence: A number of florets on a rachilla to form a spikelet; each spikelet is subtended by a pair of glumes; the spikelets collectively form a raceme or panicle. Each stem bears an ear which consists of a rachis around which some twenty spikelets close together. Each spikelet has 2-8 florets. The outer glumes protect the florets of the spicule. Each floret has one large bearded glume on the side away from the spikelet and a smaller one on the inside.

Flower: Bisexual. Compound pistil with two carpels and locules, superior ovary, fruit is a caryopsis. The ovary contains a simple ovule and bears two feathery stigmas. Three stamens. Two membranous scales called lodiculae cause the floret to open at flowering time. At the same time the stems expand pushing the anthers out of the floret. The pollen sacs dehisce and fertilize the stigmas. As the pollen is generally released when the glumes are open and as in warm sunny weather the florets open out still more it is possible on occasions for neighboring plants to be pollinated also. The flowering process begins just above the center of the ear and lasts about three days for each spikelet and up to six days for an entire ear and up to ten days for the plant.

Seed: The seed capsule (pericarp) has three consecutive layers of cellular tissue, the parenchyma, the cross layer and the inner epidermis, enclosing the seed which is firmly united with these layers. The seed coat consists of the brown layer and the hyaline membrane. Underneath are the farinaceous tissues, with an outer layer of aleurone cells containing fine grained albumen and oil, but no starch. The gluten albumen is found together with the starch grains in the large underlying endosperm cells. These are transformed into storage capacity. The embryo germ lies with the scutellum at the side of the endosperm; on the outside it is surrounded by pericarp.

Root: There is no tap root, the feathery strands of roots spread out in the top soil. At germination the embryo puts out 3 - 8 rudimentary roots and after leaves have appeared the first primary roots develop simultaneously with the tillers on the tiller nodes. During the growth of the stem new roots continue to appear so eventually a network of roots is formed, 60 % which lie in the topsoil. The primary shoots which grow from the individual tiller nodes push further apart from each other when the grain is sown more deeply; each stem forms its own primary roots. The root-stock and the firmness with which the plant is anchored in the soil increases with the number of adjoining stems. The extent to which the roots penetrate to the deeper layers of the soil depends upon the supply of nutrition and the drainage of the topsoil and subsoil. The roots may go down 2 meters. Different varieties have different characteristics as regards the thickness of the root, the depth to which it goes down and the size of the root system; this is relevant to the sturdiness of the plant and the resistance to drought.

Stem: May grow up to 1.6 meters. There are nodes at points where the leaves branch out, and in most types the stem is hollow except at the nodes. Usually there are five nodes.

Tillering: Lateral shoots develop out of the axillary buds which are present in the embryo; the stem nodes of the second and third leaves become tillering nodes. Out of the lowest node on the stem of these first side shoots (secondary stems) grow tertiary stems which can branch out in the same manner. Each stem forms its own primary roots at the tillering nodes. The secondary and tertiary stems are somewhat shorter than the main stem and bear lighter ears. Sowing at too great a depth has an unfavorable effect on tillering as the reserves of the endosperm are largely exhausted in the task of pushing the sprout to the surface. The extent of tillering varies according to the type of wheat and can be modified by the environment. Hereditary differences between varieties can be attributed in part to differences in their photoperiodic characteristics since higher temperatures up to the flowering period can stimulate tiller formation. When winter or spring wheat has been sown too late and the need for coolness not sufficiently satisfied in the spring, stronger tillering is the result. Varieties which do not begin their growth till late in the spring tiller well. With spring wheat low temperatures after sowing promote tillering. Well nourished plants standing alone can produce up to 60 ear bearing shoots. Sown in drills under normal conditions winter wheat develops three stems per plant and spring wheat two.

Varieties: Spring and summer varieties are discerned by vernalization and photoperiodism. Temperature can accelerate growth. Among the different varieties it is possible to distinguish winter wheats, spring wheats and various stages of intermediate wheat, the morphological development of the latter showing the characters of a spring or winter variety or of an alternative variety depending on the temperature and length of day in the region in which it is grown.

Spring and winter wheats display fixed characters as regards their reactions to temperature during the period of vegetation (need for coolness or vernalization) to the length of day (photo-periods) and as regards their capacities for resisting low temperatures. Spring varieties have less or no need for coolness and their growth is not markedly restricted when days are short. Winter varieties must have coolness so the plant must be subject to temperatures of 0 - 8C (vernalization) during vegetative development. Alternative varieties, like spring varieties have no need for low temperatures during vegetative development. They are sensitive to reduced daylight and will not develop during short days. Most wheat varieties develop intermediate forms distinguished by varying degrees for need of vernalization, of reaction to length of day and of resistance to cold. There are differences in temperature requirements between varieties not only for the period of preparation for flowering (vernalization) but with all phases of development.

There are three distinct groups based on the number of chromosomes. The brittleness of the rachis which serves to indicate a wild variety and the glumes of the grain and the glumes of the grain are features upon which a systematic classification can be based.

Hexaploid wheats grow in :
1. Humid, climatic zones
2. Steppe climates
3. arid and semi-arid climates
4. damp, highland climates

Hexaploid wheats of humid climates:
a) West European winter wheats
b) Mediterranean winter wheats
c) Central European wheats
d) East Asiatic wheats
e) North, West and Central European spring wheats - lowest need for warmth, have thin stems, relatively few leaves and from small grains.

Steppe Wheats:
a) both spring and winter types, predominates in Russia, as winter wheat in the steppe regions of the Ukraine, North Caucasus and Kirgistan and as spring wheat in the east and north-east. They have typical xerophytic features, thin straw, moderate tillering, small leave volume. Resistance to cold. Spring varieties of the same type are grown in Canada and the US.

Wheats of arid and semi-arid climates:
a) grown in irrigated regions and very common in Central Asia, India, Afghanistan, West China and North Africa. Small to medium height, relatively stiff stem, coarse leathery leaves, broad ears and firmly fixed grains. All ripen very early and give good yields without irrigation.

Wheats of damp, high regions:
a) Very leafy and suited to damp conditions and found in the highlands of Central Asia, Plalmyra, Kashgar and Mongolia. According to Vavilov the varieties which have been produced by cross breeding in the new areas of South America, Australia and South Africa represent a special type of wheat.

North American wheat growing differs widely from that in the Old World as to types of farming and cultivation practices. (Schlomer 1960).

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Definitions of Biodiversity

Biodiversity is a concept that links life forms with each other and their ecosystems. Dictionaries do not have the word defined. Diversity shows the differences between things, including inter and intra specific differences. Biological diversity relates to living objects but when looking at biodiversity it is impossible to separate the influences of environment, human actions, evolution and mutation as factors affecting the dynamics of biodiversity. As life forms are constantly interacting and their DNA replicating, the possibility of change is always present. It is arrogant for humans to think that we can condemn a life form to extinction because of measurements and hypothetical observations of growth and interaction. Biodiversity will not be totally understood until we can look at science and the marvel of life holistically.

Economically, and clinically, biological diversity is a central component of the stock of natural capital on which all economic development is based. The intractability of the biodiversity loss is related to the irreversibility of many of the social costs of species deletion. Ecological systems are highly non-linear, non-convex dissipative systems, the dynamics of which are discontinuous in the neighborhood of system thresholds (Perrings and Pearce 1995).

While the global system has the potential to favor nature conservation as well as expand trade and production, difficulties raised for conservation of biodiversity by short-term economic crisis in deficits in country international payments, international capital flows and foreign aid have reduced conservation efforts. (Tisdell, 1994) The importance of political lobbying by nature conservation groups in developed market economies will be a means of ensuring correction of market failures. No economic system is likely to prove satisfactory in conserving biodiversity so political action by conservationists is always required. (Tisdell 1994).

Biodiversity and agroeconomic development are natural competitors. Biodiversity results from the successful struggle of organisms to survive in the same environment that humans struggle for survival. Patterns of biodiversity are influenced by the same factors that influence the distribution of human economic prosperity.(Houston 1995).

The production of varieties with characters or combinations of characters different from those in use in the farming systems of today will depend on the identification and incorporation of genes or gene-systems with the capacity to develop the required characters. (Holden 1993). These genes must be found in the gene-pool and must be incorporated into suitable genetic backgrounds, either by conventional breeding or by genetic engineering.

When cultivated wheat was essentially a mixture of landraces there was probably a wide range of variability to be found in any single farmer's field. This variation provided a buffer against fluctuating environmental and pathogenic selection pressures, but it was considerably reduced when selection for uniformity became the norm in man's earliest

plant breeding efforts. This enforced genetic uniformity led to desirable field and harvest characters but at the price of vulnerability to pests and diseases. (Kimber 1993).

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How biodiversity is measured and conserved

Shiva (1991) says there are many strands in the conservation of biodiversity. There is the deep green ethos of the democracy of all life; it is based on the ethical ground that all life forms have value in themselves independent of the value man puts on them. This ethical concern is strengthened with justice and equity; arising from the peasant movements biodiversity takes on the significance of a struggle for self reliance and decentralization. Diversity in this perspective needs to be incorporated into the logic of production processes in agriculture because diversity protects the livelihood of different communities. The emergence of new biotechnologies have transformed the richness of earth into a strategic raw material for industrial production of food, pharmaceutical, fibers, energy, etc. Biodiversity conservation becomes conservation of 'raw material' rather than conservation of 'means of production of life'.

There are different approaches to measuring diversity. One might distinguish between taxic diversity and genetic diversity which is at the population and infraspecific level for evaluation of germplasm. Taxic diversity are aimed at conservation of genetic resources of genetic areas. Most are based on straight inventories, species richness and a combination of richness and abundance. Phylogenetic (taxic) diversity measures have been used as a basis for setting priorities for conservation of geographic areas using cladograms of taxa to be assessed for conservation. Another phylogenetic approach does not require the full resolution of cladograms aims to minimize diversity lost and maximize diversity preserved. The group is evaluated relative to sister groups and the ranking provides a guideline.

Conventional plant classification employ a diverse array of approaches (phytochemical, anatomic, morphologic, etc.) and often offer a synthesis of these data sets. Many of these traditional characters are susceptible to convergent evolution by natural selection.

Quantitative genetics is a biological way of looking at biodiversity. Phenotypic traits that allow classification of individuals into discrete and unambiguous phenotypic classes. Most of the variation between organisms is quantitative. Each genotype has a norm of reaction that covers a wide phenotypic range and there may be many segregating loci whose alleles make a difference to the phenotype being observed. Population genetic involves the transformation of a species either naturally through evolution or by human intervention. It relates the heritable changes in populations of organisms to the underlying individual processes of inheritance and development. (Suzuki 1989).

Phenotypes of most traits in nature and agriculture are continuous variables. This continuous distribution has been attributed to the collective action of many genes. They are termed quantitative trait loci (QTL) interacting with the environment. To date many agronomically and biologically important traits (e.g. resistance to biotic and abiotic stress, yield, nutritional quality) have been studied by means of molecular mapping in numerous crops. In most cases, the experiments have been conducted on balanced populations, backcrosses or F2s derived from pure lines, and have generally been able to identify minimal numbers of putative QTLs responsible for the character of interest and

to estimate the relative contributions of 'major' and 'minor' genes to the total phenotypic variation. Molecular markers have also allowed a partial insight into interlocus interactions (epistasis) and intralocus interactions (gene action) of loci controlling quantitative traits. (Styles 1986).

Adaptation to a particular ecological niche is likely to require some response to a number of interacting environmental factors and to achieve the full reproductive cycle will involve the coordination of physiological processes. This depends on the fine tuning of many gene sets and differences between ecotypes tend to be quantitative rather than qualitative. Diallel crosses between populations have allowed a better understanding of the nature and action of the genes which control population differences. In these crosses there is no evidence of any genetic isolating mechanism, since all crosses are fully fertile. (Styles 1986).

Genetic variation can be quantified using the concept of allele frequency, the proportion of all alleles at the locus that are of the same type among a group of individuals. The frequency is equal to twice the number of homozygotes for that allele plus the number of heterozygotes for that allele divided by the number of individuals. (Hartl 1980).

Molecular Genetics:
Electrophoretically discernible seed storage proteins have been used to assess variation in cereal populations, landraces and cultivars. Storage proteins of wheat endosperm are divided into two main groups, gliadins and glutenins on the basis of solubility criteria in different solutions. In both groups there is a high level of heterogeneity determined strictly by the genotype. This has aided in varietal identification, detection of off-types in pure seed production and related problems as well as showing migration of species from centers of diversity and genetic variation in collections. Allelic variation at each of the three complex loci *Glu-l* is the main reason for quality differences in bread wheat and the different alleles at each locus have been ranked according to their influence on bread making. Allelic variation at gliadin loci has been shown to be responsible for differences in baking quality and dough strength. Gliadins are responsible for low nutritional value of wheats and detection of mutants lacking entire clusters of gliadin components offers a different perspective to the improvement of whet nutritional value. (Lafiandra 1990).

The role of introgression (interspecific gene exchange) in plant evolution is debated. It could result in the breakdown of isolating barriers between two partially isolated taxa and their subsequent merger. Molecular markers provide a means of analyzing ambiguous cases of introgression, whereas morphological characters often converge when exposed to similar selective pressures. Organellar genomes (chloroplast and mitochondrial) are useful for the study of introgression because they often contain nonrecombinant, molecular markers. Nuclear markers are inherited in a simple Mendelian fashion and will be additively combined in hybrids or introgressants. The use of isozyme markers relative to RFLP is limited by the generally low number of diagnostic alleles distinguishing closely related forms and the difficulty of defining and polarizing isozymic character sites. (Riesberg and Brunsfield 1992).

Requirements for sampling at DNA level show that faulty phylogenetic hypothesis based on chloroplast DNA can be avoided if data from nuclear gene sequences are analyzed and after one devises comprehensive sampling strategies (Rieseberg, Choi and Ham 1991 in Baum 1996). Wheeler (1992 in Baum 1996) points out that drawing on characters from a single taxon to represent a large and diverse group could be viewed as a form of artificial extinction. Most molecular studies are based on a very limited sampling. The accuracy of a cladogram is most influenced by the number of taxa, more than the number of nucleotide sites so the most accurate results are ensured by inclusion of the highest number of taxa in a group.

Restriction fragment length polymorphisms (RFLPs) use certain restriction enzymes called endonucleases that cleave DNA at certain points. One ends up with a collection of different lengths of DNA. Associated DNA markers with traits are labeled with P32 and are used to identify the fragments of the RFLP. This technique maps the fragments of the chromosome.

RFLPs can be used in any plant species to obtain detailed maps of genetic linkage. RFLPs are codominantly expressed, do not have pleiotropic effects on agronomic traits and the number of possible markers is infinite. Hybridization patterns of all the available probes can be detected with RFLPs and they can detect more polymorphism than biochemical markers because many of the probes are non-coding, less conserved sequences. RFLPs could be exploited for obtaining very dense, saturated genetic maps for gene tagging and the dissection of quantitative loci which would be valuable for screening wheat germplasm and for monitoring the transfer of useful genes. RFLPs are expensive and require the manipulation of radio isotopes. Isozymes require staining and electrophoretic techniques. The isolation of a cDNA library will produce more probes. (D'Ovidio 1990).

Enzymes differing in electrophoretic mobility as a result of allelic differences in a single gene are allozymes and allozyme variation in a population is an indication of genetic variation. Estimates of polymorphism may be underestimated because routine electrophoresis fails to detect many amino acid substitutions. RFLPs are usually identified by using the Southern blotting procedure. Some probes identify sequence variation at variable sites known as minisatellites; DNA fingerprinting. Polyploidy refers to an organism having multiple sets of chromosomes. If a base substitution results in the loss of an amino acid in the protein product it is a nonsynonymous mutation and allozymes are the result. (Hartl 1989).

To obtain DNA sequences one starts with a defined fragment of DNA uniquely labeled at one end, then generating a population of molecules that differ in size by one base and being able to separate these molecules and identify the base in each case. A single stranded DNA is placed on a gel (acrylamide or agarose) and subjecting it to an electric current to separate the strands on the basis of their lengths. The mobility of the strand is inversely proportional to the logarithm of its length. This technique is so sensitive that fragments differing in length by only one nucleotide can be separated. DNA cloning and

DNA sequencing are related; the DNA cloning provides large amounts of DNA fragments. 3' end is labeled with 32P and cleaved and isolated, and then separated one strand for the other to yield a population of identical strands labeled on one end. The sample is divided into 4, then one or two specific bases are destroyed. This makes the sugar-phosphate backbone more likely to break at that point. It results in a mixture of different sized pieces carrying the 32P label. When these pieces are separated in the different lanes of a gel they can be arranged in order of length and the base destroyed at each site be determined by noting in which lane or lanes the band appears. Thus the sequence of bases in the strand can be read.

RAPDs use primers that find a sequence on double stranded DNA. The primers synthesize the DNA which is subjected to a polymerase which allows them to stick together and to reproduce up to a million times. The RAPDs can appear anywhere on the chromosome.

AFLPs use fragments from RFLPs then use RAPDs to attach the fragments. It is a proprietary technique which makes it expensive for non researchers.

Molecular markers can be used to map and characterize quantitative trait loci (QTLs) for several characters of agronomic and biological importance.(Grandillo 1996). Several regions on the genome showed effects on more than one trait; these appear to be scattered but appear to be in 'hot spots' so it is assumed they are tightly linked.

Chloroplast DNA (cpDNA) variation has proven to be immensely valuable in reconstructing phylogenies at the species level. One approach is to analyze the distribution of major structural rearrangements. Because of their infrequent occurrence, rearrangements usually can provide strong evidence of monophyly. (Downie 1992).

The chloroplast genome contains with few exceptions two duplicate regions in reverse orientation known as the inverted repeat. These repeated regions separate the remainder of the molecule into large single copy and small single copy regions. The expansion or contraction of the IR into or out of adjacent single copy regions and changes in sequence complexity due to insertions or deletions of unique sequences are largely responsible for variation in size of the molecule. (Downie 1992).

The cDNA is one of the most efficient probes for screening for cloned genes. The mRNA for the gene in question can be used as a probe; it is difficult to obtain sufficiently pure mRNA. The enzyme reverse transcriptase can be used to make a DNA copy of the partially purified mRNAs which can program the synthesis of a complementary strand. The remaining DNA duplex is termed complimentary DNA (cDNA). It is easy to purify individual cDNA fragments. The total population of cDNA fragments can be joined to plasmids so that each plasmid receives only one fragment and bacterial cells can be transformed so that each cell receives only one plasmid. Clones derived from single cells can be tested for which unique cDNA is present. Amplified DNA is purified and denatured and used to trap the corresponding mRNA on a nitrocellulose filter. The purified mRNA is used to program a cell free protein synthesis system to allow

identification of the protein encoded in the complementary mRNA and thus the gene present in the cloned mRNA. The purified cDNA can be used as a probe to screen colonies directly from a gene bank, since replica planting techniques have been developed that empty nitrocellulose filters to detect hybridization of DNA from colonies with specific probes. When partial digests are used to establish the gene bank, there will be fragments of different sizes containing the gene in question and the larger region surrounding the sequences contained in the probe can therefore be analyzed. (Downie 1992).

Data derived from cleavage points of various restriction endonucleases in all genomes (nuclear, mitochondrial and chloroplast) have been used for phylogenetic reconstruction. Mapped restriction sites which represent a sampling of a whole genome or of any specific sequence can be considered estimates of homologous characters and their transformations. Organisms evolve by character-state transformations. Diverse evolutionary processes can be examined from the framework of a cladogram. All individuals are part of a phylogeny so they are connected by inherited, homologous character-state transformations in a topological relationship that can be estimated by sampling within characters and across taxa. Phylogeny represents a process of descent with modification. (Albert 1992).

Chloroplast DNA is maternally inherited making it suitable for identification of maternal parentage species of polyploid crops. The identification of specific mutational events increases the confidence that the restriction fragments are in fact the same. The mitochondrial genome is maternally inherited. It is typified by a much higher rate of structural rearrangement generating higher levels of intraspecific RFLPs but lowering confidence that two restriction fragments with the same electrophoretic mobility are indeed the same. Without detailed restriction mapping it is nearly impossible to sort out restriction site mutations and structural rearrangements in mtDNA. Nuclear DNA offers opportunity to study repetitive sequences such as ribosomal gene and single copy or low copy number sequences or genes. Nuclear DNA sequences provide evidence for the maternal and paternal linkages and levels of polymorphism for nDNA are suitable for intraspecific analyses. For cpDNA crops and their relatives can be analyzed for the presence or absence of particular restriction sites and insertion or deletion events. These events can be used to construct phylogenetic trees using parsimony. For nDNA and mtDNA the higher levels of polymorphism create a more complex situation if the molecular basis of the differences in restriction fragments is not understood. The mtDNA RFLPs are apt to mix both restriction site changes and structural rearrangements distances between them will not provide an accurate estimate of the substitutions per site but will provide a degree of phenetic similarity. (Doebley 1992).

Methods:
Restriction enzymes provide a way of cleaving DNA from any source at a specific sequence thereby producing a heterogenous population of fragments with identical ends. The restriction enzyme target sites can be used as markers for DNA. DNA from a specific source is subjected to successive digestion by different restriction enzymes. The

map of the restriction sites can be made when the fragments are separated electrophoretically on polyacrylaminde gels.

Plasmids are circular DNA molecules that can be introduced into cells through transformation. They can confer antibiotic resistance to a cell. Single stranded phages can be used as cloning vectors; on infection of E. coli the single infecting strand is converted to a double stranded replicative form which can be isolated and used for cloning.

Expression vectors offer the opportunity to express the cloned sequences by fussing them to the appropriate transcription and translation start signals. This allows the foreign DAN to be expressed in the respective host.

Shotgunning is the nonselective method for cloning random DNA fragments from a higher organism into bacteria. The entire collection represents a gene bank or gene library which can be screened for specific genes. All the plasmids or phages comprising a gene library together represent the entire genome of an organism with each different plasmid or phage carrying a different small fragment from the genome.

Southern Blotting identifies cloned genes. Single stranded but not double stranded DNA can stick to nitrocellulose. During electrophoresis on agarose gels, DNA fragments from restriction digests will migrate according to their size. They can be transferred to nitrocellulose by a buffer flow, after denaturation and immobilized on the filters in a pattern that mirrors their positions in the agarose gel. P labeled DNA or RNA probes are then used for hybridization with the affixed DNA on the filters. The probes are denatured and allow annealing with the single stranded restriction fragments that are anchored on the gel. Unlabeled single stranded DNA from an unrelated sources is used to saturate the remaining sites on the nitrocellulose to prevent nonspecific binding of the single stranded probe which can now bind to the filter only by annealing to the complementary DNA fragments. The position of bands that anneal to the probes is revealed by autoradiography.

Isoenzymes:
Enzymes differing in electrophoretic mobility as a result of allelic differences at a single loci are called allozymes. Allozyme variation in a population is an indication of genetic variation. This technique makes use of the proteins in a molecule. (Hartl 1980).

Immunoassays:
Enzyme immunoassays (EIA) are analytical procedures based upon the specific binding of animal derived antibodies to a target molecule; the antibody or antigen is tagged with an enzyme. Diagnostic sensitivity means that a test is positive whenever the target antigen is present even at low levels. Equally important is specificity where the test is negative when the target antigen is not present. Specificity and sensitivity are used to determine the predictive value of a positive or negative test. The antibody is responsible

for the specificity and sensitivity of the immunoassay. Antibodies suitable for use in immunoassays may be produced by immunizing a laboratory animal with a preparation containing the antigen to be assayed. Additional materials called adjuvants are commonly injected with the antigen-containing preparation to enhance the production of antibodies. (Landlow 1987).

Measuring devices:
Coefficient of parentage (COP) values can quantify germplasm sources and their contribution to the gene pool, measure changes in genetic diversity through time and identify major groupings of related cultivars. (Mercado 1996) found that landraces and local cultivars contributed to the formation of the gene pool in spring wheat (64%). Similarity among released Canadian cultivars remained high until the 1970s when the introduction of new market classes resulted in a 50% reduction in similarity. From 1901-1920 very low similarities existed among cultivars. Marquis and cultivars of the 1930s were very similar.

If a trait is shown to have some heritability in a population then it is possible to quantify the degree of heritability. There are average differences between the genotypes and each genotype exhibits phenotypic variance because of environmental variation. The total phenotypic variance of the population can be broken into two portions; the variance between the means (genetic variance) and the remaining variance (environmental variance). The degree of heritability can be defined as the proportion of the total variance that is due to the genetic variance. The broad heritability tells what proportion of the populations' variation in phenotype can be assigned to variation in genotype. It does not tell what proportions of an individual's phenotype can be ascribed to its heredity and to its environment. An individual's phenotype is a consequence of the interaction between its genes and its sequence of environments. Genetic variance and heritability can be estimated in several ways. By making a number of homozygous lines from the population and crossing them in pairs to reconstitute individual heterozygotes and measuring the phenotypic variance within each heterozygous genotype an estimate of environmental variance can be obtained. Other estimates of variance can be obtained by considering the genetic similarities between relatives. (Suzuki 1989).

Wheat specifically:
Chromosome arm designations in wheat initially were designated L or S (long or short) on the basis of differential arm length within a chromosome. Now p is used for short and q for the long arms of wheat chromosomes (Gill 1987). Basing the system of homoeology causes problems in hexaploid wheat. The earliest method of genomic analysis done by Kihara using morphology measuring total amount of chromosome pairing. The recognition of the bivalents in multiples of the basic number is taken to indicate genomic homology. Mathematical methods allow the recognition of patterns of chromosome pairing in hybrids. Self fertility of alloplasmic lines and restriction enzyme analysis of cytoplasmic organelles show evidence of the maternal parent in allopolyploids. Immunochemical and electrophoretic studies often deal with a single nuclear loci and these can produce unequivocal results. They show evidence of evolutionary process at

the gene level but on the basis of differentiation at single loci cannot be used to draw evolutionary conclusions. Nuclear probes as with chromosome pairing still cause questions as to what is being compared or labeled. Just because two chromosomes have the same banding does not prove their homology, they just have the same pattern. They do not show that the same sequences appear at several locations throughout the genome, the same sequences can be found in distant nonlineal taxa and the bands that are observed represent replicated regions, and some 95 percent or more of the DNA may not be detected. Arm ratio provides no evidence of the genetic content of the chromosome but the relative position of the centromere. (Miller 1988).

In wheat, RFLPs find few sequences with restriction enzymes. RAPDs work for finding loci sites and do not require the use of radioactive materials. Isoenzyme work is now obsolete; it makes use of proteins.

The *Nor* and *5Sdna* loci are composed of tandem arrays of units that carry the genes coding for RNA products, as well as associated spacers. The spacer regions separating the genes at these loci have also been studied in considerable detail at both the restriction fragment length polymorphisms (RFLP) banding pattern level and sequence level. (Appels 1992).

The *Nor* loci of Triticeae have been studied for many years and are useful for studying the application of DNA analytic information to problems assessing the relationships between the species. The studies fall into two broad levels of resolution, that of variation in the DNA fragments generated by restriction enzymes as assayed by specific probes (e.g. RFLPs) and variation at the level of DNA sequences. The RFLP analysis of *Nor* loci is very extensive. (Appels 1992).

In contrast to RFLP analyses, the comparison of DNA sequences is at a very high level of resolution and is more complex. A basic prerequisite for a comparison is an appropriate alignment of the sequences based on either the primary sequence or secondary structure. Following alignment, numerical methods are used to infer phylogenetic relationships between the sequences under study. (Appels 1992).

Published studies on the *5Sdna* loci in species of the Triticeae have shown that within a species two separate loci for the *5SDna* can coexist and that these loci are characterized by certain size classes of unit. At each locus the respective size class of the subunit is tandemly arranged and different parts of the unit accumulate changes during the course of evolution at different rates. (Appels 1992).

Cultivated wheat (*Triticum* L. species) are autogamous, disomic polyploids characterized by phenotypic buffering and tight linkage inside terminal chiasmata. However wheat also has great evolutionary potential through diploidization of homomeric loci and alien gene transfers. Wheat improvement by conventional methods implies the search for a superior genotype or group of related genotypes for a given agroecological niche. This search may rely on the creation of genetic variation by means of simple convergent, or composite crosses, or by the introduction of mutations. Then the outcome mainly depends on the

efficiency or duration of the process of population management and selection. Alternatively the ambition may be to direct variation by means of strict backcrossing rather than relying on selection. It should however be very clearly realized that most efficient breeding systems will require alternating or simultaneous procedures for recombination and selection. The characteristic inheritance pattern of polyploid wheat and the impossibility of realization recombination potentials for more complex goals within reasonable plot or population size, calls for strategies accepting some kind of stepwise progress. (MacKey 1987).

Autogamy does not stimulate development of genetic isolating mechanisms and allopolyploidy offers a network of intergenic relationships. The polyploid wheats have a disomic mode of inheritance which allows allelic interaction to operate between homologous loci at homozygosity. The diversification of homologous loci in tetraploid emmer wheat (*T. turgidum*) has proceeded partly due to mutation and partly due to introgression. B genome has been involved in the latter, and the A genome maintains more the function of preserving vital genes. Gene reduplication increased considerably when the D genome from *Aegilops squarrosa* was added to form the hexaploid dinkel (*T. aestivum* L) wheats not more than 8000 years ago. These wheats are thus characterized by a fairly high phenotypic buffering a conserving trend enhanced by a tendency to form terminal chiasmata inside which tight gene blocks may be maintained. Such a constitution offers tolerance to various genetic deletions and additions. In no other crop plant has the aneuploidy technique (Kimber & Sears 1980) reached such a sophisticated level of development and had such wide ranging applications as in wheat. Together with the weak isolating barriers, the ability of wheat to tolerate chromosomal manipulation has proved very useful in alien gene transfers by the conventional method of sexual recombination (Knott & Dvorak 1976; Sears 1981). The Ph><ph switch regulating meiotic pairing offers an elegant transfer procedure (Riley *et al*; 1968; Sears, 1981) that is available as a consequence of the allopolyploid constitution of wheat.

Wheat is autogamous and thus mostly represented by fixed genotypes. One allelic gene pair is able to give two different homozygous genotypes but 25 pair are able to give over 33 million. (MacKey 1987). Because linkage and homomeric inheritance preserve the phenotype, lack of genetic knowledge has made it comparatively difficult in wheat to utilize unadapted germplasm sources for improving polygenic characters. Such material, both inter and intraspecific, has so far been explored mostly for oligongenic traits such as disease resistance, dwarfness, and a few others. A wider more systematic exploitation of primitive or unadapted sources is being done in breeding for high protein content and stress resistance (Avivi, 1977).

The most promising method of producing doubled haploids is through anther culture, but the success rate is low and highly variable from cross to cross. Regenerated plants form wheat tissue culture show a remarkable range of somaclonal variants. While some are unstable, others are stable and have commercial potential. Nitrogen fixation has been shown to result form associations between certain bacteria and wheat. (Knott 1987).

Embryo culture techniques, using radiation to transfer segments of chromatin from one chromosome to another developed by Sears (1956) and development of procedures that induce homoeologous pairing are three advances in wheat breeding. Three procedures have evolved: chromosome manipulation using nulli-5B tetra-D to obtain plants lacking *Ph,* the gene which prevents homoeologous pairing; the use of crosses with species such as *Aegilops speltoides* Tausch to suppress *Ph*, or the use of *ph* mutants. The first two methods result in more recombination, but also more problems with sterility. The use of *ph* mutants reduces problems with sterility but also gives less recombination (Sears, 1981).

Genome analysis is not a finely tuned experimental technique. There are differences in the treatment of species between cytogeneticists that confuse breeders. Other geneticists and biosystematists base species boundaries on either genetic or ecological reproductive isolation. If there are substantial barriers to gene exchange between two taxa in the field, irrespective of whether they share the same genome, they are considered separate species. If there is reproductive isolation between wild wheat taxa in natural populations there is the possibility of genetic differentiation. To use wild germplasm effectively a useful system of species classification must be developed.

The B genome donor contributed the cytoplasm of durum wheat and was presumably the female parent. Breeders write the female parent first in cross notation, so the genome formula of durum wheat is BBAA and bread wheat is BBAADD. If the *Aegilops* species is the female parent then the genome formula is SSAA. The nomenclature problem in wild wheats and their relatives derives from the group being placed historically in two genera T. L and *Aegilops* L. by morphological taxonomists. Floras use this system; the impetus to merged the two genera derives mostly from Morris and Sears (1967) who followed Bowden (1959) and the International Code of Nomenclature for Cultivated Plants (Fletcher *et al*., 1958). These authors assume that the B. genome (maternal genome)of tetraploid wheat is from an *Aegilops* species and therefore *Aegilops* and *Triticum* should be merged. Geneticists and cytogeneticists do not yet agree which *Aegilops* species is the donor of the B genome of the BBAA tetraploid wheats. The biosystematic and nomenclatural problems are a great constraint to germplasm evaluation. Most crosses of wild *Triticum* and *Aegilops* species as males with tetraploid and hexaploid wheat cultivars as females are successful if sufficient care is paid in setting up the cross. (Waines 1990).

Meiotic chromosome pairing and recombination in eukaryotic organisms are multistage processes and the premeioitic association of the pairing partners whose closer spatial relationship is necessary to achieve the correct sequence of events leading to crossing over and chiasma formation. In polyploid species an organization level between the different chromosomal sets is also operative. The genetic homoeology of their component genomes in natural allopolyploids chromosome association is almost invariably restricted to homologous bivalents at metaphase 1 so meiosis is mechanically and genetically efficient. Divergence between homoeologous chromosomes is not usually sufficient to achieve complete diploidization. Homoeologues have so little tendency to pair and the homoeologous pairing is suppressed by a gene *Ph1* on the long arm of

chromosome 5B. *Ph1* acts as a homoeologous pairing suppresser gene. Additional suppressers as well as genes exerting an opposite effect (pairing promoters) are present and active in the wheat genome. A second *Ph* locus is *Ph2* on the chromosome arm 3DS. (Ceoloni 1994).

Wheat microsatellites (WMS) were used to estimate the extent of genetic diversity among 40 wheat cultivars and lines and reveal alleles and results indicate that a small number of microsatellites can be used to estimate the genetic diversity and cultivar identification in elite material of hexaploid wheat (Plaschke 1995).

Collections:
Gene banks preserve some genetic diversity or cultivated crops and wild relatives, botanical gardens preserve some diversity of selected native plants, herbaria preserve expressions of morphological variations within and between species. There is no substitute for biodiversity present in natural habitats as sources of future germplasm.

For collections to be useful for research and conservation they must be truly representative of the genetic diversity. Sampling attempts to portray the real statistics of an area and in taxonomy this is genetic variability as in the study of crops and their wild relative and taxic variation for most groups of plants. (Baum 1996). Sampling approaches include random sampling, cluster and multistage sampling, and various combinations of approaches. The size of samples is often neglected or is a contentious issue. Most biosystematic research is indifferent to this aspect and yet to distinguish between hypothesis whether two groups are different at species level requires large samples.

The core collection concept (Frankel and Brown, 1984) consists of a subset of accessions which broadly represents the genetic diversity of the collection. Passport data, morphological and agronomic data collected by gene banks aids in the evaluation process. Predictive attributes could be extrapolated from existing data; in finding a wheat for pre-harvest sprouting the environment the sample was collected from would indicate which accessions originated in environments where there is rain during the maturity stage of growth. The data on the origin would be very valuable when combined with other attributes would be useful when selecting varieties for future evaluation. Tools for linking environments to accessions need to be developed, such as a database with soil types, climate and geographical regions. (Mackay, 1990).

Four functional classes were introduced by the International Biological Program (IBP) in 1966: landraces, advanced cultivars, wild relatives of domesticated plants and wild species used by man. To conserve landraces entails the conservation of the conditions of traditional agricultural ecosystems. Advanced cultivars have a narrow genetic base; many older cultivars are replaced by superior quality varieties and the genes are not conserved. Wild relatives hold a wealth of genetic material for crop improvement; the sampling strategies depend on the breeding system. A minimum of five hundred plants has been suggested as a minimum to capture the variation of each population (Qualset 1975 in Baum 1996).

One of the more controversial issues in genetic conservation is the definition of optimal sampling strategies. In order to develop a quantitative theory of sampling it is necessary to define an appropriate measure of genetic diversity, the useful genetic variation which is to have priority in sampling. Marshall and Brown (1981) argued that the number of alleles per locus provides the best measure of genetic diversity in the context of genetic conservation. It is a direct measure of genic variation in a population. It is more reliable than diversity indices based on variance in quantitative characters which measure only the portion of genetic variability expressed phenotypically. Allelic variants can be divided into four classes depending on their population frequency and distribution in the target plant. The variants in each population can be classed into those which are common (frequency less than 5%), and those rare (frequency less than 5%). Each variant with at least one common occurrence is classed as to whether it is common locally or widespread.

A wide range of variation can be observed for a number of morphological characters including size and arrangement of leaves on the stem, leaf color and pubescence, color of the awns, angle of arrangement of the awns, color of the ear-head and color of the grains. In addition awned, hooded or awnless heads are found. The main phenological characters are germination, days to heading, days to flowering and maturity and growth habit. The main morphological characters are plant type, spike density, lodging, glume hair, plant height, spikelets per spike and awn characteristics. Grain yield, reaction to diseases and pests and seed color and size were also recorded.

A statistic describing phenotypic diversity was necessary to compare average diversity between regions. Two commonly used measures of phenotypic diversity are Shannon's information statistic (Hutcheson, 1970; Bowman *et al.*, 1971) and the phenotypic polymorphism index (Kahler *et al.*, 1980).

Modern plant breeding has reduced the genetic variability in wheat. Programs to identify variation should maximize the efficiency of sampling strategies by using ecological-genetic factors and biochemical markers as predictors. When the representative electrophoretic patterns of populations plotted far from each other it was possible to detect a fairly large difference in protein bands with low electrophoretic mobility. The separation of populations by this method partially reflects their geographic distribution and populations can be classified by the altitude of the collecting site. (Benedettelli 1990).

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Summary of the wheat genome mapping project

The International Triticeae Mapping Initiative was formed in 1989 as an informal consortium of geneticists dedicated to developing recombination maps of the genomes of the species in the Triticeae tribe of the Poaceae family. Comparative mapping among the various genomes in polyploid species and in the diploid progenitors was a goal. Although the genomes and chromosomes are large, the large bank of aneuploid stocks of wheat and chromosome banding polymorphisms have made possible physical mapping within the Triticeae to a greater extent than in other plant species.

Recent advances and progress in plant genome mapping offer new opportunities for breeding for stress environments. Molecular markers should allow breeders to tag genetic loci controlling stress resistance without having to measure the phenotype thus reducing the need for extensive field testing over time and space. The responsible genes can be cloned via map-based cloning techniques and manipulated by plant transformation techniques for crop improvement.

The Canadian Wheat Genome Mapping Group is an information group of researchers. The germplasm pool from which Canadian wheat cultivars are bred is perhaps smaller than in most countries because of our continuing focus on the maintenance of a narrow set of parameters on quality. This has decreased the frequency of the detection of polymorphisms to a level below which significant progress could be made. Therefore considerable effort has been expended on developing DNA fragment separation techniques which will increase the level of polymorphism detected. Canadian breeders have been developing near isogenic lines differing for disease resistance or agronomically useful genes for the past half century. (McGuire 1995)

The Canada Wheat Genome Mapping Group has eight research centers where active cereal biotechnology program are in progress

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Work in Centers of Diversity on crop diversity

Based on the diversity he encountered Vavilov (1951) identified Abyssinia as the origin of tetraploid wheats. Harlan (1971) put forward information on the place of origin of cultivated plants elsewhere due to the absence of wild progenitors and near relatives in Ethiopia. Diploid ($2n = 2x = 14$) Einkorn wheat (*T. monococcum*) is not grown and hexaploid ($2n = 6x = 42$) wheat is a late introduction, wheat evolution in Ethiopia is restricted to the tetraploid (($2n = 4 x = 28$) wheats. Within the tetraploid group, emmer wheat (*T. dicoccum*) which itself arose from wild emmer (*T. dicoccoides*) was the first to be domesticated. Wild emmer has not been found in Ethiopia. According to Halbaek (1959) emmer was most likely introduced from the Near East (via Egypt) which is believed to be the center of origin for wheat. This does not rule out the possibility that durum (the first free threshing tetraploid wheat) came to Ethiopia together with Emmer or even before.

Farmers in Ethiopia believe that broad genetic variability is believed to buffer against adverse environmental conditions in a locality. Bulking of two or more superior pure lines may result in a cultivar mixture with better yield stability and disease resistance in the locality for which it is intended. Cultivar mixtures consist of superior pure lines possessing similar seed color, glume color, height and maturity. It is believed that farmers prefer such a mixture especially if the grain is white because it gets a higher price on the market than mixed seed colors.

Each year hundreds of durum wheats are received from CIMMYT, ICARDA and elsewhere in the form of pure lines and segregating populations. The pure lines are tested for adaptation while segregating populations are selected for local adaptation through a number of generations of selfing, using the pedigree or bulk method. The primary objective of introduction is to evaluate wheat grown in other parts of the world and select genotypes adapted to Ethiopian conditions as well as those that have special features, such as disease resistance for use as parents in breeding programs. (Gebre-Mariam 1991).

Ethiopian wheat is rich in both interspecific and intraspecific variability and its wide agro-ecological amplitude can serve as a basis for selection for specific areas of adaptation. Although the landraces are apparently less desirable in terms of yield, it is evident that they possess desirable traits that can be incorporated into high yielding, improved cultivars.

Various non government organizations (NGOs) in Ethiopia work with farmers and genetic resource conservation. The Seeds of Survival program sponsored by USC Canada links the farmer with the breeder and the gene bank. The role of the farmer in maintaining crop diversity alive is recognized and breeders assist in developing higher yields in selected varieties. The gene bank staff characterize and evaluate the accessions and store the materials.

Diversity and relationships among ten tetraploid wheat landrace populations were studied using isozyme markers and agronomic traits. This type of analysis is fundamental for designing optimal germplasm collection, management practices and for developing an index for parental selection. The populations differed in allelic frequencies. Much of the diversity was attributable to the within-population level. Cluster analyses of the isozyme and agronomic data produced different pattern and memberships of groupings. (Tsegaye 1996).

Seasonal variables - genotype x environment (G x E) interactions will appear from observations over a number of seasons. The objective of analysis is to minimize the variance of the varietal means across season in order to obtain a maximum likelihood estimation of the varietal means. It is assumed that each genotype occupies its own characteristic place relative to others within the range of variation of a particular trait. (G x E) interactions will be disclosed by the incidence of varietal means with relatively large variances. It should be possible to identify genotypes showing G x E interaction by screening the standard deviations of the means. (Giles 1990).

During the multiplication, characterization and preliminary evaluation work done in Ethiopia it is noted that accessions collected in the same locality are a mixture of two, three or more species with different botanical forms. In some fields, 8-10 different botanical forms were observed in a single sampling. The hexaploid wheats in Ethiopia are of limited diversity and were probably introduced by Portuguese explorers during the 18th century. The tetraploid wheats have been grown in the highlands and have evolved diversified forms and unique characteristics not known elsewhere. Genetically homogenous accessions of self-pollinating crops are easy to handle whereas heterogeneous accessions cause complications and are difficult to record variable characters adequately , it is not easy to regenerate variable populations without losing rare types. There are varieties with purple kernels in durum. (Mekbib 1990).

The Green Revolution of the 1960s had a highly beneficial effect on human food supplies but adverse effects on the habitats of many crop species. There was displacement of wild relatives and landraces in West Asia and Mediterranean by modern semi-dwarf varieties. Modern agriculture is characterized by extreme homogeneity in cultural practices and crop varieties.

Sixty collecting mission in the Mediterranean and Ethiopian centers of genetic diversity for wheat have yielded 2171 populations which distributed throughout the world have stimulated breeders to incorporate new sources of variability for agronomic traits, resistance to diseases and protein, nutritional and technological qualities into new wheat lines and varieties. The field data recorded during collection have not been used extensively for selecting samples to distribute to breeders but they could be used for studies of evolution or co-evolution with wild species and of the ecogeographical distribution of important traits. Germplasm can be used only if large collections are screened in different environments.

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Impact of Modern Agriculture on Wheat Genetic Diversity

The late 1950s affected wheat and agriculture profoundly. Norman Borlaug (1958) stated that loss from diseases could be reduced if great diversification was incorporated into the varietal make-up of wheat. He believed that a group of composite or multilineal wheat varieties would meet this need. The varieties would have the essential characteristics of a conventional 'pure line' such as uniform morphological appearance or plant and grain and uniformity of gluten quality. This concept encourages field diversity. However, Borlaug won the Nobel Prize for his "Green Revolution" work which seems to have deviated from his visions in his paper (1958).

The process of diversification seems to have reached its peak at the middle to end of the 19th century. This process is now in reverse. The diversity of landraces which supported agriculture for the past 9000 years is being rapidly eroded and for some temperate crops is now nearing completion. This has happened through the substitution of new genetically uniform cultivars which are grown in environments that are becoming more uniform through the application of increasingly sophisticated agronomic practices, including improved tillage, irrigation, artificial fertilizers and the chemical control of pests and diseases. (Holden 1993).

Landraces characteristically consisted of mixtures of plants differing in form, color, height, vigor, and yield. The outcome was the production and marketing of improved varieties which often bore the selector's name. Vilmorin in France and Shirreff in Scotland were two who produced new single plant selections as varieties of wheat which replaced the landraces from which they were derived. Towards the end of the 19th century a great surge occurred in the breeding of crop plants with scientists as well as farmers and gardeners. Confusion reigned about the merits and identities of the varieties; not infrequently the same variety was sold under different names in different areas. Agricultural production came to based on fewer high performing varieties and this trend has been reinforced in some countries in recent years by regulations limiting trade in seed to officially approved varieties. (Holden 1993).

Landraces are genetically heterogeneous. There can be much variation in length of straw, architecture of the ear and color of the grain. This diversity extends to characters which cannot be readily seen, such as in their genes for resistance and susceptibility to their various pathogens. Landraces present a genetically complex target to the pathogens moderating the more extreme expressions of disease.

Wheat cultivars were originally all heterogeneous populations. They offered reliability under stress conditions rather than high productivity. When agricultural science was introduced in the middle of the last century, productivity could be emphasized, resulting in the selection for pureline cultivars adapted to high seeding rates. (MacKey 1987). Since then attempts to maximize reliability and productivity has occupied breeders.

Until the beginning of the current century the wheat crop under general cultivation in India comprised a mixture of different varieties, generally known as 'sorts'. Based on the

kind of grain such as white or red, hard or soft, the different 'sorts' were given common names. Almost all the commercial sorts are now out of cultivation. The work on the improvement of wheat in India was initiated in the beginning of the current century. The first phase concerned the improvement of the tall genotypes and the second is an era of the development of the dwarf and semi-dwarf wheats. Three phases exist for each process; the introductions from vastly different geographic areas, then selection and isolation from the introductions and local germplasm then as the variability decreased the step of hybridization was started. (Gill 1979).

North American varieties came from a variety of countries. According to Reitz (1958) the prototype of Red May became established before the American Revolution and was especially important in Virginia. A white wheat know as Goldcoin dates to 1768. Club, common white and durum wheats, probably of Spanish origin, were grown in California in early times. Mediterranean became known in 1819 and was adapted to the southern Corn Belt area. Purplestraw from unknown source, dates to 1822 and was the basis for the wheat industry of the SE states. White Australian later called Pacific Bluestem reached California from Australia by 1850 and was good in the west. Red Fife selected in 1842 was in the US in 1860. The introduction from Russia of Turkey winter wheats to Kansas, Iowa and Nebraska between 1873 and 1900, and of durums to North Dakota before and after 1900, and Federation from Australia in the early 1900's.

Genetic diversity is the foundation of all plant improvement programs. The use of specific cultivars and genetic stocks can be directly associated with major contributions in wheat improvement. Agronomically mediocre cultivars have combined to provide outstanding progeny. Much diversity is being lost with the ever increasing rate of genetic erosion. (Knott 1987).

N.I. Vavilov is generally credited with recognizing the role genetic diversity plays in plant improvement. He noted that plant breeding is evolution at the will of man and like all evolution is dependent upon variation. The sources of genetic variation collectively may be considered germplasm. Creech and Reitz (1971) define germplasm as individual donors of genetic traits at one extreme to large assemblages of breeding stocks at the other extreme, or as an array of plant materials, assembled or not, that serves as a basis for crop improvement or related research.

Genetic diversity is being reduced or lost by the very cultivars created from it (Hawkes 1981). The worldwide trend is reaching the primary and secondary centers of the origin of wheat. As yields have increased wheat breeders have been reluctant to make crosses with older landraces since the probability of resulting segregating populations containing desirable progenies is very low. Thus the development and release of new high-yielding cultivars is in fact contributing to a drastic narrowing of the genetic base. In many breeding programs plant breeders are discarding potentially valuable genetic stocks because such material does not fit their particular objectives at a given time. Furthermore, when personnel changes are made in breeding programs, valuable genetic stocks are often lost due to lack of documentation or lack of familiarity on the part of the new scientists with the desirable attributes of specific selections or cultivars. (Knott 1987).

Cultural practices are being introduced such as multiple cropping, intercropping or even minimum or no tillage systems that may have deleterious effect on maintaining landraces that because of their heterogeneity do not conform to a particular management practice. Disruption of wild habitats and destruction of wild species as well as farming and grazing land extend, logging and other activities that cause soil erosion alter native ecosystems. Modern farming methods involve weed control, the opportunity for introgression of genes from weeds to crops (e.g. From Einkorn to wheat through natural crossing) is greatly reduced, thus limiting this scope of genetic diversity (Zohary 1970).

Frankel (1978) has identified regions where genetic erosion has occurred, including the Mediterranean region where indigenous wheat cultivars present in Greece in the 1930's have virtually disappeared except in remote mountain areas. In Cyprus considerable genetic variation is still available for durum wheats and exploration could be productive.

In Near East countries primitive wheats of all ploidy levels are in jeopardy. A rapid erosion of genetic diversity is occurring among the indigenous wheats in Iraq. Iran and Pakistan still have many indigenous landraces of wheat growing under natural rainfall conditions in the mountainous regions. The great diversity of wheats once found in India has nearly disappeared.

A genetic survey by Frankel and Bennett (1975) raised questions concerning the status of genetic diversity represented by wheats within existing collections. It was noted that little is known about primitive cultivars even in large collections. In some instances accession do not represent the populations from which they were obtained because the collections were based on only a few plants. Questions could be raised about the genetic integrity of the material due to the probable occurrence of outcrossing, unsuitable growing conditions and resulting natural selection and human error during propagation. Often storage conditions are inadequate and records unreliable.

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Major Issues for the Future Conservation and Utilization of Wheat Genetic Diversity

Issue: Practically all ecosystems are threatened by human caused destruction and degradation including loss of ecosystems; these are the wombs of life. These include the Mediterranean basin of ecosystems, where many of the wheats and relatives originate and are very diverse. (Shiva et al. 1991). Rempel's reply: International agencies could target funds and expertise to the collection, evaluation and characterization of germplasm, as well as identification of areas for *in situ* conservation preservation efforts. This is not as glamorous as biotechnology research and funds are constantly being cut from national budgets to support basic ecosystem preservation work.

Issue: In the millennia following the establishment of wheat cultivation, farmers in many parts of the world have unwittingly created a wealth of wheat forms, domesticated from their wild ancestors. The reduction of the diversity of germplasm now facing the world has consequences for humankind which are far more profound than other, more widely publicized environmental dilemmas, such as the depletion of the ozone layer or the destruction of the rainforest. (Damania 1993). Rempel's reply: Kew Gardens in England has launched their Millennium project with a goal of collecting seeds from all over the world. Many scientists see this as a 'Noah's Arc' approach that will divert funds from national gene bank work.

Issue: Agricultural systems are characterized by human intervention which generally results in a purposeful reduction in the species richness of the system. On a global scale agricultural systems are not necessarily low in diversity. Sustainability is now considered along with maximizing biological or economic yield. The farm unit is linked in an area and economically because of shared markets and infrastructure. There are classes of regulators of the biological functions and the production system is divided into plants, animals and microbes. Farms are considered production units, not living and dynamic entities. Intercropping and low number multiple cropping systems are options in increasing biodiversity on farms. (Swift et al. 1994). Rempel's reply: Efforts at the United Nations are attempting to define the concept of Farmers Rights and begin to put humanism back into farming. This is an uphill struggle against regulations that restrict crop and seed choice by the farmer as well as great pressures to grow high input varieties. This affects the ecology of the farming system dramatically.

Issue: Biodiversity sciences represent the disciplines of whole organism biology, systematics, ecology, population biology, comparative biology. Each nation should establish a national biodiversity research program coordinated across all government agencies. (Cracraft 1995). Rempel's reply: Despite the Canadian Biodiversity Strategy existing on paper, the Environment Department Biodiversity Office will not become involved in Agricultural biodiversity conservation unless the Research Branch of Agriculture Canada endorses and funds the project. There is a large void between agriculture and environment in Canada and globally. Many scientists are still challenged by the concept of multidisciplinary research. There is little cross discipline work

happening in biodiversity conservation and little effort in educating people in agricultural biodiversity issues.

Issue: Although new technologies provide details of habitat loss estimates of future extinctions are hampered by our limited knowledge of which areas are rich in endemics. (Pimm 1995). Rempel's reply: Funds are not being allocated to do inventories and site assessments for conservation efforts.

Issue: Major collections of wheat are held around the world. They are repositories of the biodiversity available for each species and a source of genes. Workers face problems in the maintenance and exploitation of the germplasm because of large accession numbers. Core collections increase the efficiency of management by prioritizing representational subset of accessions for attention. Molecular markers identify variation and estimate biological diversity. PCR based marker systems, including RAPDs showing the differences between genomes. (Virk et al.1994). Rempel's reply: There is some hope that the glamour of biotechnology in genetic resource conservation will draw a few extra dollars to biodiversity conservation efforts. Developed countries are supposed to have the wisdom to give funds to their gene banks. This does not happen and labor and money dwindles daily in support of developing country work. The irony of this is illogical to most people involved in biodiversity conservation efforts in Canada. Canada's gene bank was rated 15th in the world and Greece 16th; having worked in the Greek gene bank I am aware of the degeneration of the collection day by day and inability of the workers at the bank to properly assess the collection. In Canada the emphasis on the gene bank is for industry support.

Issue: Lyons (1985) has identified the sources and nature of institutional barriers to biotechnological research in grains. His team concluded that first, there is declining support in grains research, and development of varieties remains heavily dependent on federal funding. There is a need to encourage greater participation in funding by the farmers, agribusiness, and provinces. The emphasis within federal funding agencies on biotech is aggravating the problems of under funding of grains research with a lack of recognition of the relevance of traditional plant breeding technology and resources are being transferred out of traditional plant breeding into the new research. Coordination between the new and traditional research is lacking. The third conclusion is that the licensing system as it is applied is restricting productivity and economic returns on the Prairies. Markets for Canadian wheat have changed in the direction of lower quality wheats. The market for high quality wheats will remain important but it is growing very slowly. Methods exist and will become available to reduce the need for visual distinguishability in new varieties. A change in this licensing requirement would allow lower quality wheats to be produced. The potential exists for production of lower quality wheats in area of the Prairies which are not suited to the higher quality varieties. Provided that the integrity of Canada's quality wheat marketing can be maintained, diversification in wheat production and marketing would benefit all segments of the grains economy. To achieve this result would require changes in the regulatory framework and plant breeding programs as well as increased support to wheat research.

In the development of the current biotechnology thrust in agricultural research there appears to be a lack of comprehension of the process that has to be completed prior to the development, licensing and commercial use of a new grain variety.

As funds for traditional research shrink the demand for scientists and training at the university is reduced. This is further aggravated by the apparent lack of inter-disciplinary involvement between agriculture researchers, biologists and geneticists, at the government and university levels. In the normal course of events, the old biotechnology applied relative to the quality standards specified in the Canada Grain Act. The improved variety is then subjected to a testing process to be proven. Institutions such as the Canadian Grain Commission (charged with some for the regulatory aspects of grain marketing) and the Canadian Wheat Board (responsible for the marketing of specific grains) have argued for the maintenance of existing standards.

Revisions to patent policy in Canada have sought to achieve the appropriate compromise between providing the correct environment for creative activity and minimizing the monopoly effects of protection. It is in that context that compulsory licensing provisions are now available within patent policy.

The method of seed distribution with SeCan means distribution is through a select and restricted group of seed growers.

Significant disagreement exists on the basic questions of what wheats Canada should produce and for what market. There is little evidence available to support continue our historical approach. It is clear that the most important participants in the decision making prices are firmly committed to a position of no change.

Lyons (1985) states that Canadian grains are licensed and only a few of the most promising varieties are accepted for assessment in the Cooperative Test. The chosen new varieties are compared to several existing varieties at numerous locations across the Prairies. Three Expert Committees, Grain Breeding, Grain Disease and Grain Quality review the data. When all Committees recommend a variety be licensed, the breeder submits an application to the Plant Products Directorate of Agriculture Canada for a license. The Expert Committee on Grain Breeding consists mainly of breeders and geneticists who review results on agronomic qualities such as yield, time to maturity, resistance to lodging, height and test time. Membership on the Grain Quality Committee consists of chemists, marketing experts from the Wheat Board and Grain Commission and milling companies. Potential conflict of interest is great when plant breeders have a say in licensing their own varieties. New varieties must conform to a number of criteria that defines 'quality' and the standard is in the Canada Grain Act schedule of grades. New varieties are tested against minimum standards including protein content, gluten strength, flour yield and appearance, and general baking quality. No variety can be licensed unless it meets all the milling and baking criteria regardless of agronomic merit. Allowing lower quality non distinguishable varieties into the system could lead to a gradual erosion of the quality of the top grades of wheat. An alternative would be to come up with some other method of identification of variety. The visual distinguishability

requirement restricts the gene pool from which the parent breeding stock and this costs Canada in yield potential. Canada's wheat breeding program seems to disregard yield as a major selection criterion and is preoccupied with kernel characteristics, quality, etc. The official insistence that market potential for low quality Canadian wheat is limited restricts research. The Canada Grain Act and the Seeds Act as well as the Canadian Grain Commission and the Canadian Wheat Board positions should be questioned.

Concerns that the emphasis on high quality and visual distinguishability requirements cost farmers in yield and revenue and restrict biodiversity. The world grain market must be looked at as many countries demand medium quality lower priced grains. Licensing requirements in Canada largely exclude producers and breeders from this market. Rempel's reply: Little more needs to be said than what the authors have stated above.

Issue: There are options including research on landraces. Landraces are genetically heterogeneous. There can be much variation in length of straw, architecture of the ear and color of the grain. This diversity extends to characters which cannot be readily seen, such as in their genes for resistance and susceptibility to their various pathogens. Landraces present a genetically complex target to the pathogens moderating the more extreme expressions of disease A strategy for disease control attempts to combine the benefits of genetic diversity for resistance gene characteristically found in landraces with the advantages of uniformity and high yield, characteristic of modern high yielding varieties. This is done by creating varieties from artificial mixtures of a number of selections which differ only in the vertical resistance genes which they carry. Vertical resistance is usually qualitative in its effect and can be transferred from parents to progeny in breeding. These are known as multiline varieties. Component lines in theory differ only in the resistance genes which they carry but can lack uniformity in growing and harvesting and marketing and use of the crop. (Holden et al.1993). Rempel's reply: Breeding work should begin to acknowledge that landraces, mixtures and multilines could support biodiversity in the field as well as maintain yield productivity.

Issue: New crops are needed for different purposes including replacing those which are in overproduction, new crops to extend farming into hitherto marginal lands to increase food production and improvement of cultivated species which are scarcely different from wild plants. However, the dependence on high-input systems is more and more questioned and the need to develop sustainable systems based on new principles, techniques, varieties and crops may offer the incentives needed for the exploitation of a wider spectrum of the diversity of the plant kingdom. (Holden et al.1993). Rempel's reply: Major shifts will need to occur in the approach to agriculture nationally and globally. If sustainability is to be true, then global sustainability is to be considered. Politics prohibit people from eating, not crop yield. Human greed is a major concern. Poverty is a money making activity for some, and until attitudes change research will support high input high output farming and not bioregional self sufficiency.

Issue: The issue of crop biodiversity conservation is linked with the social and environmental impacts of farming methods, from pollution to debt and dispossession; they argue for radical changes in agriculture and food production (Clunies-Ross 1995). Rempel's reply: Chemicals have killed off many of the natural predators and have

allowed farmers to abandon crop rotations and mixed cropping practices in favor of monoculture. Since the Irish Potato Famine of 1846/47, plant breeders have sought to safeguard against similar tragedies by establishing collections of genetic material. But gene banks are not a substitute for diversity in the field and on the ground. A limited number of varieties dominate farms. A limited number of varieties are recommended by government agencies and although farmers are often aware of the need to diversify their hands are tied by regulations and legislation. By the 1970s it became illegal to sell seed not on the UK National List. In 1973 this list was harmonized with other EC countries to form a Common Catalogue. The high annual fee for maintaining varieties on the List mean that varieties that do not sell in sufficient volume are dropping, making it illegal to buy or sell the seed. Under Thatcher's government, Britain's publicly owned plant breeding institutes were sold off and privatization has placed plant breeders under further pressure to concentrate on developing varieties which sell in large volumes, enabling them to recoup their investment in research and development. As most farmers use chemicals to enhance yield and control disease breeders have concentrated on varieties that respond well to chemicals. Through competition the breeding companies have tended to develop varieties with similar characteristics. The big supermarket chain buyers, millers and processors exert influence on what is grown on the farm. Farmers who can avoid the supermarkets have been able to retain or increase diversity (Clunies-Ross). Funding has been diverted from the development of crops or varieties with minority end uses. Whole areas of research, including biodiversity work in Canada, have been abandoned because the economic return does not justify the development costs.

Over the past fifty years diversity has been progressively abandoned in favor of chemical control, and trapping farmers and breeders and chemical manufacturers on a treadmill which is going faster and faster. As long as marketing structures, processing techniques and seed legislation pressure farmers to grow a limited range of genetically uniform crops diversity cannot be maintained. Innovative agreements between consumers, farmers and political pressure to change the economic and regulatory framework one farms under. As the need to reduce chemical use in agriculture becomes apparent and accepted so does the need to put diversity back at the heart of farming if crop failures are to be avoided.

Issue: Population growth and changing dietary habits have led to an increased demand for protein; combined nitrogen and biological nitrogen fixation represents the major outputs of nitrogen for crop and protein yield. Wheat varieties have differing potential for adding nitrogen to the soil; a nitrogen balance study done from 1843-1967 in England showed an average annual gain of 34 kg/n/ha and 24 kgN/ha were removed in straw and grain. (Neyra 1977). Rempel's reply: Time to go back to the old agricultural manuals of the mid 1800's to the mid 1900's for many answers are already there and much of the research done. Politics are dictating the modern agricultural practices.

Issue: The short term goal of plant breeders exploits genetic diversity for multivariate organizations of characters and the long term objective of genetic conservationists must be to enhance genetic diversity for such adaptively organized multivariate characters in breeding populations. A progressive increase in the frequencies of several desirable character combinations is possible and desirable in maintenance of high genetic

diversity. (Jana et. al 1990) Rempel's reply: Breeders are finding funds cut in favor of biotechnology. Politics again.

Issue: The key to success in crop improvement for dry areas lies greatly in tapping the variability of existing plant genetic resources for use in breeding adapted germplasm. With increasing population pressures on natural resources, the challenge for the drylands will be to generate technologies which can increase yields through the judicious use of inputs without risking instability of production. The deployment of genetic resources to improve and stabilize crop production in the face of biotic and abiotic stress is a component I in ICARDA's strategy to develop appropriate technology for difficult environments. In addition to landraces and primitive forms, wild relatives can provide genes for disease resistance, high protein content, tillering, drought tolerance and other economically desirable attributes. So progenitors of cultivated species should be considered as an important source of variability for broadening the genetic bases of crop plants. The exploitation of collections is hampered because this material is scanty in collections and the material available is therefore not representative. Work on wild forms has concentrated on evolutionary and taxonomic studies and variability within populations of wild species has not been done and utilization has not begun. Data assembled after conserving, evaluating and documenting the genetic resources of primitive and wild wheat germplasm should be deciphered to show variability within populations, between populations and between countries and regions as a whole. (Damania et al.1990)

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