



## An Overview of Breeding Objectives to Improve the Economically Important Traits in Oat

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### Abstract

The history of the modern oat may be traced to its intergeneric contamination of wheat and barley in southwest Asia, particularly in Mesopotamia and due to its use in only forage crop rather than a staple food in most of the countries it's been neglected. There are just a few economically cultivated species of this self-pollinated crop, including *A. sativa*, *A. nuda*, and *A. byzantina*. After rice, wheat, maize, sorghum, and barley, oat production comes in at number six and in terms of oat production (FAO 2019). The present review was undertaken in order to address the present scenario of oats utilization and its consideration in research. The work majorly focussed in present are  $\beta$  glucan (due to its nutritional status) and disease resistance (majorly powdery mildew, pyrenophora leaf spot and crown rust) throughout the world but some drawbacks is that oats is usually neglected because of its limited use as forage value and also of its hexaploid nature. The different breeding methods employed till date for crop improvement are described in the chapter viz, traditional methods, diversity studies, mapping strategies, Marker aided breeding Association mapping, Genomic assisted breeding etc. Some of the qtls mapped by various researchers for different traits is also been listed in this article. Apart from these studies, the novel techniques which are still lacking in oats has much scope now to get it utilized through various improvement programmes like in other cereal crops.



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### Introduction

Winter cereal crop oat (*Avena sativa* L.), a member of the genus *Avena* and family Graminae, has its origins in the Mediterranean region (Poaceae). The history of the modern oat may be traced to its contamination of wheat and barley in southwest Asia, particularly in Mesopotamia.<sup>1</sup> When main foodstuffs were

moved throughout the Neolithic era's agricultural expansion into Europe, it most likely did so.<sup>2</sup> There are just a few economically cultivated species of this self-pollinated crop, including *A. sativa* ( $2n=6x=42$ ), *A. nuda* ( $2n=2x=14$ ), and *A. byzantina* ( $2n=6x=42$ ). Oat production follows rice, wheat, maize, sorghum, and barley at number six (FAO STAT 2019), with the

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European Union leading the way, followed by Russia, Canada, the United States of America, and Australia, which are in fourth and fifth place, respectively.<sup>3</sup> Currently oats account for 9.26 million hectares of oat crops worldwide, with an output of 22.51 million metric tonnes and an average yield of 2.43 metric tons/ha (FAO STAT).

Oats are now more commonly consumed as human food as a result of its rising popularity as breakfast meal. Oat has highest amount of soluble fibre beta-glucan per 100 g varying from 5.0 g to 7.2 g compared to 0.5-2 in wheat and 1-2 in rye. Oat is higher in unsaturated fats like linoleic acid and has more lipids (5–9%) than other cereal grains. Along with tocotrienols and tocopherols, which are similar to vitamin E, oats also contain avenanthramides, which are distinct antioxidants.<sup>4</sup> Oats have a high concentration of soluble fibre, which lowers cholesterol and lowers the risk of heart disease. Oats are thought to have a protein content (11-15%) that is comparable to that of soya and egg proteins. In India, oat breeding started in the 1890s, and it is now farmed as the most significant grain fodder crop throughout North Western, Central, and Eastern India. India has several research facilities and agroecological zones that may be used to create varieties that are appropriate for various climates and production methods of fodder. India has a well-organized procedure for evaluating breeding lines before releasing them and notifying farmers so they can cultivate them.

It features an outstanding growth pattern, high-quality foliage, and rapid post-cut recovery. Oats are effectively produced in the plains and hilly regions of the nation because they need a lengthy, cold season for growth. Northern states among the Indian states has much of the area grown with numerous soil types are appropriate for the growth of oats. Oat is a staple in cattle feeding and is a superb source of minerals, fibre, and high-quality protein. However, the expected output of oats globally has decreased.

#### Floral Biology of Oats and its Progenitors

The flowers are arranged in distinctive structures known as spikelets and are encased in scales or bracts. A panicle is the structure used for oat spikelets. Each spikelet has a small joint axis, called a rachilla, that bears florets. Inside the spikelet are the florets. Two or three florets might

exist. When it contains three florets, they might be a primary floret that has opened, a secondary floret that hasn't opened, and a basic tertiary floret. Due of the absence of sepals and petals, oat flowers are incomplete.<sup>5</sup> Each floret consist of lemma, palea, stamens and stigma and two large glumes. Following hand pollination of both intraspecific and interspecific cross pollinations, oat breeders have observed inadequate seed set. Backcrosses for gene replacement and crossings for inheritance research have gone unperformed since it is challenging to get enough hand-crossed plants. Numerous reasons including temperature sensitiveness and stigma receptivity are most likely to blame for the poor seed set in contrast to other cereal flowers.<sup>6</sup> The oat blossom is delicate and readily damaged during the emasculation and pollination processes. Furthermore, a large amount of seed set is controlled by environmental conditions. Studies by some of the researchers showed that seed set as well as comparing the number of crossings produced in oats compared to others is low in percent.<sup>7</sup>

The common cultivated oat is hexaploid constitutes of three genomes i.e. AACCCDD. The A genome donor of the tetraploid and hexaploid oat is considered to be three species i.e. *A. canariensis*, *A. strigosa* and *A. longiglumis* viz morphological evidences relates to *A. carariensis*, cytological evidences are in favour of *A. longiglumis*. The possible donor of C genome is suggested to be *A. ventricosa* species. The tetraploid *A. insularis* is the most likely donor of CD genome, although in recent review<sup>8</sup> it is attributed as AACCC genome.

#### Important Traits Under Consideration in Oats Globally for Research

##### β Glucan

Breeding for increased levels of β-glucan in oat has been a part of variety development programme worldwide because when we consume oat as a food or breakfast cereal, it leads to positive and constant health effects. The economic and nutritional value of the crop can be increased by developing the varieties of oat with higher contents of β- glucan. As oat β-glucan trait is under the control of number of genetic loci with additive effects, so it is considered to be a polygenic trait.<sup>9</sup> Genetic × Environmental interaction has been reported for this trait by many but the relative significance of (G×E) is not clear yet. In certain cases, the G×E effect is thought to

be minor for genotypes which show similar  $\beta$ -glucan content ranking among the various environments.<sup>9</sup> In some cases, this effect was considered to be significant even when it was tested through mean square for year by variety<sup>10</sup> and there is no consistent correlation between  $\beta$ -glucan content and other agronomic characters. Because of huge genetic variability in germplasm for  $\beta$ -glucan content availability it can be used in breeding methods for its study.<sup>11</sup>

The presence of  $\beta$ -glucan and other agronomic characteristics may not always correlate with one another.<sup>9</sup> Long-term benefits via selection are obviously achievable because of the enormous genetic heterogeneity in germplasm for beta-glucan content.<sup>12,13</sup> Given that accurate measurements of  $\beta$ -glucan are challenging and expensive, molecular genetic deconstruction of the characteristic and subsequent application of genetic markers for marker assisted selection (MAS) might help to lower the cost and speed up the process of modifying the  $\beta$ -glucan concentration in oats. The ability to manipulate traits by genetic selection as compared to phenotypic selection, and maybe even the isolation and cloning of the specific genes involved, is made possible by molecular mapping, which is a crucial tool in genetic improvement. If there is diversity in the DNA sequence of the actual gene governing the characteristic, one may easily influence the phenotype even though it is determined by molecular markers. Genetic selection may be performed on seedling stage or even on half-endosperm samples thanks to molecular markers that are closely connected to or even present in the region around the locus of interest. Only because of genetic selection has high-throughput screening been made possible for features that are expensive or difficult to assess and/or are influenced by environmental factors. The identification of QTLs (quantitative trait loci) for several variables via marker-based characterisation is essential for their genetic modification because few commercially significant qualities in farmed oat are controlled by single genes. As a result there is need to identify qtls responsible for the increased  $\beta$ -glucan concentration in oats.

### Biotic Stress

Stress is a factor that negatively affects crops within favourable effects which can be either abiotic (related to soil and environment) or biotic

(weeds, diseases, insects, and so on). A change in temperature of environment become stressful quickly in comparison, to soil minerals stress which might take time to become stressful.

### Important Diseases

In 1889, *Helminthosporium teres* (var.) *avenae-sativae*, the fungus that causes leaf spot of oats, identified for the first time in Europe. Warm temperatures and high humidity are excellent for the growth of the HLB disease pathogen, which has been well-established in oats and is mostly peculiar to that species.<sup>14</sup> The first part of the oat seedling to get infected is the coleoptile, resulting in one or more small and brown lesions in affected plants. The seedling's future development is determined by how severely the parasite infiltrated it, and in extreme cases, the underdeveloped small shoot is attacked even before it emerges from coleoptile. Such plants are stressed and weakened to the level that they are unable to enter the soil above them, and they take on a twisted appearance and later eventually die. Further, pathogen has been seen as a minor pathogen of barley and other species of grasses.<sup>15</sup> *Drechslera avenae*, the causal agent of HLB, is a sexual phase of *Pyrenophora chaetomioides*, a necrotrophic parasitic disease causing the brown spots with purple margins. These blotches lead to a fall in the amount of leaf greenness, which, in turn, causes a decline in photosynthesis and, consequently, a decrease in tillering. In 1984, it was reported as a significant issue in the southern United States, Western Europe, Japan, and India,<sup>16</sup> and during epidemics, estimates suggest that Germany and the southern United States suffered losses of 30–40 percent (<sup>55</sup>and<sup>56</sup>)

Crown rust, also known as *Puccinia coronata*, is said to be the most harmful oat disease in the world which can increase lodging, lower forage yields and quality and grain yields.<sup>17</sup> Rusts must constantly infect new hosts since they can only develop and replicate on live plants. They survive the summer by infecting wild and uninvited oats, and the following season they infect crops. Round to rectangular, orange to yellow pustules that occur 7–10 days after infection, mostly on leaves but sometimes on stems and heads, are the defining sign. The pustules' powdery spore masses are easily moved around. As they mature, the pustule regions turn black. Crown rust also lowers protein yield and seed

weight goat percentage.<sup>18</sup> One of the researcher<sup>19</sup> developed KASP markers tightly linked to crown rust resistance *pc39* using RIL population, which was mapped on chromosome 1C and linkage group Mrg11 using GWAS studies.

Similarly, barley yellow dwarf virus is the second-worst disease for oats in the world (BYDV). The aphid species *Rhopalosiphum padi* L., *Macrosiphum avenae* Fab., *Schizaphis graminum* R., and *Metopolophium dirhodum* are the most significant disease vectors for oats. It is possible to confuse the symptoms of the barley yellow dwarf virus (BYDV), which causes leaf yellowing, reddening, and striping, with those caused by nutrient deficiencies, waterlogging, or other plant stresses. Finally powdery mildew is one of the major disease threatening oats yield *i.e.* its lower leaves and sheaths develop fluffy white spots from powdery mildew. The cottony patches become powdery in colour as the condition worsens. If the outbreak is strong, leaves may die and the region around the spots may eventually become light yellow.

Oat genotypes' racial-specific and non-racial-specific resistance and tolerance to fungus infections are already identified. Oat race-specific resistance to fungi is influenced by a single either dominant nature, partially dominant, less recessive or incompletely recessive independently inherited gene as well as gene interaction like complimentary genes.<sup>20</sup> Recessive genes with additive effects predominantly influence racial non-specific resistance (slow disease spread) and tolerance. The effectiveness of disease resistance is predicted to rise with the presence of molecular markers linked to desirable qualities.

### Pests

Different pests, some of which have particular agronomic value, attack the oat crop. There are six different aphid species that harm oat crops. These species include *R. Maidis*, *Rhopalosiphum padi*, *Diuraphis noxia*, *Metopolophium dirhodum*, and *Schizaphis graminum*. For their direct and indirect losses, two of the species known as Russian Wheat Aphid (*D. noxia*) and Bird Cherry-Oat Aphid (*R. padi*) are famous. In a climate that is conducive to them, they multiply in large numbers and significantly reduce agricultural yields.<sup>21</sup>

### Abiotic stress

Also Abiotic factors are responsible for 50% of the overall yield losses in comparison to all other pressures that affect the crops.<sup>22</sup> High temperatures (40%) and salinity (20%) are the most frequent environmental stressors, followed by drought (17%), cold temperatures (15%), and a variety of other stresses (*i.e.*, only 9% of the world's land is suitable for grain cultivation while 91% are subject to numerous stresses that limit crop output and quality). Agricultural productivity can be reduced by 50 to 70 percent as a result of abiotic stress, which has a substantial influence on the growth and development of crop plants.<sup>23,24,25</sup> The release of greenhouse gases from diverse sources (global warming) is one of the factors assumed to be responsible for the steady increase in global average temperature. This alters the pattern of precipitation, which in turn leads to irregular drought or flood stress.

### Breeding Methods – Traditional to Recent Approaches for oat Improvement

#### Traditional Approach

The selection of proper breeding programme for the development of cultivars that are of our interest are influenced by a number of factors, including the screening procedures, tolerance sources and mechanisms, gene action and heredity, and their relationships to agronomic qualities. Traditional breeding techniques have been used to develop varieties of peanuts, common beans, safflower, chickpeas, wheat, tall fescue, soybeans, wheatgrass, barley, and maize. In the conventional method, DNA is recombined by separate chromosomal assortment and crossing-over using both the sexual cycle and existing genetic variety. The oat genome is large and complicated, and DNA sequence information is now about to get over which causes oat molecular genetics research to lag substantially behind that of other cereal crops. However, there have been continuing efforts to increase the density of the map with various kinds and numbers of markers ever since published the first linkage-based QTL map in oat.<sup>26</sup>

Due to the hexaploid nature of oat, a small number of its genes have been discovered through diverse studies based on their phenotypic effects. The most recent experts to summarise these genes were.<sup>27</sup> Even if they were homozygous, the majority of

gene mutations leading to loss of function in the hexaploid oat would be covered by genes with the same function on homologous chromosomes in the other component genomes. Marker-trait associations have been identified for some oat genes and QTLs, though not to the same extent or density as in many of the major cereals. Marker-assisted selection has also been utilised or is now being tested for several economic factors. This work will go on as oat researchers make use of scientific developments to provide quicker, less expensive, and higher throughput markers, DNA sequencing, and bioinformatics analyses.

#### Use of Wild Species as the Main Source

Fundamental knowledge on the linkages and patterns of evolution among agricultural plant species, which are major sources of variety, is provided by wild species. The majority of the wild oat species are composed of several hexaploid species that belong to the core gene pool.

#### Diversity Studies

Genetic diversity analysis is a technique that allows for the testing or evaluation of genetic variation across people or germplasm lines using a variety of approaches, either separately or in combination. Different types of data have been employed to estimate the genetic diversity of crops.<sup>(28 29)</sup> However, it has been shown that genetic diversity studies are useful for categorising genetic stocks numerically into several genetically distinct clusters and understanding the genetic divergence in a sizable germplasm collection.<sup>(30 31 32)</sup> In evaluating phenotypic diversity using morphological genetic diversity analysis, genotype plasticity and genetic diversity are both significant factors.<sup>34</sup>

Since the beginning of plant breeding era, natural variation and diversity across crops have been used to enhance and improve crops. This naturally available variability has been much used again and again over time as a result of breeding practices *i.e* to improve only a few traits and using it for introduction of a few outstanding lines to many countries that led to increased genetic similarity between crops. Hence, identifying diverse lines, creating diversity and its utilization are the main goals of any improvement programmes. Morphological markers—variations of a specific plant species that spontaneously

developed and are still used today—were originally employed for diversity analysis and may be used in conjunction with cytological, biochemical, and molecular characterization to determine variety.

#### QTLs Mapping Carried Out in Oats for Various Traits

A popular method for examining the position and actions of the factors causing quantitative genetic variation is QTL mapping. Earlier, a variety of quantitative features have been investigated into Oat using traditional biometrical methods. But the advancement of mapping methods in recent years has created new opportunities for the discovery of chromosomal areas harbouring QTLs influencing significant features. Considerable breeding efforts are being made by various researchers for increasing the  $\beta$ -glucan levels in the oat cultivars because of its positive health effects associated with consumption of oats.

One of the researcher<sup>35</sup> made the selection within the families of population developed by crossing of the two inbred lines for identifying beta glucan genes inheritance pattern. Even though it was phenotypic selection, the genetic gain was significant, but molecular marker development in oat has enabled the selection which uses genotypic information of individuals. Similarly Two RIL population having a common parent (Kanota x Ogle and Kanota x Marion) were used by one of the researcher<sup>36</sup> in order to find the genomic regions that are responsible for increased  $\beta$ -glucan content in cultivated oat. Similarly<sup>26</sup> had created the first genetic linkage map in the diploid species *A. atlantica* *A. hirtula* (1992). Seven of the eight QTLs associated with MN841801-1 alleles had already been identified by,<sup>37</sup> and a novel QTL with the name Prq8 was shown on linkage group MN13 conferring resistance to crown rust disease. Also, some researchers (2013) generated SNP and KASP markers for marker-assisted selection of the crown rust resistance gene Pc91, located on oat chromosome 7C-17A.<sup>38</sup>

First study on mapping powdery mildew resistance in hexaploid oat using comparative mapping was done with single dominant gene regulating resistance to powdery mildew.<sup>39</sup> Similarly, researchers discovered five powdery mildew resistance genes (Pm1, Pm3, Pm6, Pm7, and Pm8) in four cultivated oat<sup>40</sup> Among



these five genes, Pm6 shows recessive mode of inheritance and rest genes showed dominant mode of inheritance and genetic map was prepared for Pm1, Pm6 and Pm7 using Restricted fragment length polymorphic markers. Few of the genes mapped for powdery mildew disease resistance are pc38, pc39, pc48, pc54 pc68 and pc 83 etc by various scientists<sup>41</sup>

### **Recent Approaches Paving the Way for Oats Improvement**

#### **Association Mapping Studies**

Association mapping is assembling a diverse group of individuals from both wild and breeding populations that may be utilized for mapping quantitative features involves using linkage disequilibrium (LD), which is a result of hybridization between the lines used as parents.<sup>42</sup> One study on AM in oat that have been undertaken is Genome wide association mapping of oat crown rust (biotic stress) by<sup>43</sup> which employed 2972 SNPs for genotyping on 631 oat lines for association mapping of quantitative trait loci (QTL)

One advantage of association studies is that this AM method is likely to find markers that are significantly closer to the genes of interest than classical linkage mapping can. This is expected since LD analysis takes into account any possible recombination events between the gene and the marker in the historical population used for AM. While AM has the potential to be extended to non-model organisms as well, linkage mapping only takes into account recombination events that occur between the gene and the marker after the two parents have hybridized. Efforts would also be made to adequately address issues like epistasis, GxE interaction effects, and phenotypic plasticity.<sup>44</sup> The AM also has some drawbacks, including the transition from candidate gene studies to GWAS based on whole-genome sequencing of everyone in the sample due to the current decrease in genotyping costs, and results being influenced by a variety of factors, such as population structure, kinship, and so forth, which can lead to incorrect associations between QTLs and markers

#### **Molecular Mapping of Resistance Genes**

The study of DNA markers has led to a new technological revolution in plant breeding. Due to this, a wide range of DNA markers have been

created, making it easier for breeders to select complex traits, especially those that are difficult to assess phenotypically. Marker-assisted breeding (MAB) benefits breeders by allowing them to conduct effective and speedy selection based on DNA markers. Additionally, it is now possible to utilise it to pinpoint characteristics that are influenced by both qualitative and quantitative genes, which facilitates the development of numerous DNA markers and genetic maps. However, the oat molecular marker system is less developed than that of other cereal crops (such as rice, wheat, maize, etc.).

Mapping populations often include primary and secondary populations. In other words, primary mapping populations are produced through hybridization between two homozygous lines that differ in the desired trait. On the other hand, by crossing two lines or individuals picked from a mapping population, secondary mapping populations are produced, which are typically used for fine mapping of the genomic area of interest. The primary mapping populations are of the following different types F2, F2-derived F3, backcross (BC), backcross inbred lines (BILs), doubled haploids (DHs), recombinant inbred lines (RILs) and near-isogenic lines (NILs) etc.<sup>46</sup>

#### **Marker Assisted Breeding**

Genetic markers are crucial tools for analysing variation in the DNA sequences of various genotypes because they can characterise genetic resources without being confounded by environmental impacts. Because of their better repeatability, co-dominant inheritance, simplicity, low cost, and frequently high polymorphism, microsatellites are excellent markers for molecular study. These can be used in a range of investigations, such as marker-assisted selective breeding, genetic mapping, QTL analysis, variety protection, genotyping, and studies on diversification. Fewer works related to MAB carried by<sup>47</sup> and<sup>48</sup> on marker-assisted breeding in oat. Hence there is need to open up MAB scope for oat improvement as like other cereals.<sup>49</sup>

#### **Map-Based Cloning of Genes**

It is necessary to clone a gene using chromosome walking or other genomic techniques in order to comprehend its functions and explore its possible agricultural applications. To conduct chromosomal

walking on model crops and extract genes for large genome crops, high-resolution maps are required. The use of probes built from heterologous DNA markers that are closely related to the target genes can be used to assess YAC or BAC libraries made from model crop genomic data. DNA. The steps in map-based cloning include creating a mapping population, genotyping with genome-wide markers for coarse mapping, going on to fine mapping using saturated areas around the markers, and finally confirming the outcomes using transformation method or other techniques. *Avena* has enormous genomes, with a hexaploid Oat genome of 11,315 Mb and a diploid oat genome of 3,770 Mb per haploid genome, although it has received little research. In order to take advantage of the vast amount of data from other plant species that is currently available and find the orthologous link between genes in other species, a precise syntenic relationship between oat and other species must be built.

### Genomic Assisted Breeding

A replacement for the conventional MAS is genomic selection (GS), also known as genome-wide selection and initially put forth by various researchers,<sup>50</sup> forecasts an individual's overall genetic value using all of the markers that are evenly spaced throughout the genome. The goals of GS are to estimate accuracy as marker density and training population size increase, to measure accuracies when data is divided over time, and to investigate accuracy in the presence of population structure. However, the constancy of LD across populations, training population size, genetic linkages of training and selecting candidates, and marker density all have an impact on how accurate GS is.<sup>51, 50, 52, 53</sup>

### Conclusion

Conventional breeding methods for creating agricultural plants with great performance are typically given less weight.<sup>54</sup> The following are some of the causes for this: (1) the focus is on yield rather than particular attributes; (2) the difficulties in breeding for tolerance traits, which include issues caused by genotype by environment or G x E interactions. (3) the negligence of employing fundamental physiological characteristics as indications of tolerance when they may be less vulnerable to G x E interactions. Traditional breeding has been slow to produce high-yielding plants that are resistant to biotic stress conditions due to difficulties in accurately defining the target environment, complex interactions between drought tolerance and surroundings, and a lack of adequate screening methods. However, hexaploid oat genome sequencing (*Avena sativa*) has recently been completed, and its genome size is to be over 11 Gb composed of two different ploidy species as *Avena longiglumis* (AA) (3.7Gb) and *Avena insularis* (CCDD) (7.3Gb) which aids in quickening the process of improving the oat for a number of qualities.

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### Conflict of Interest

No potential conflict of interest relevant to this article was reported.

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