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Epigenomics: Approaches and Implications In Crop Improvement

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ARTICLE ID: 032

Introduction

An epigenome is the sum of all biochemical modifications in the biogenesis of nuclear DNA, histone proteins, and non-coding RNAs (ncRNAs) in a cell. At the molecular level, a gene's transcription is affected not only by its DNA sequence, but also by how genes are arranged within the chromosome's complex architecture. The DNA of eukaryotes is highly condensed and closely coupled with proteins called histones, and this DNA and histone protein complex is referred to as "chromatin." To start transcription at a particular gene or genes, the chromatin at that position must be relaxed so that transcription factors (TF) can bind and recruit RNA polymerase to start transcription. Thus, whether a gene will be “on” or “off” is determined by the chromatin state at particular gene(s). Epigenetics is the study of epigenetic changes in and around DNA that control genome function, and epigenomics is the branch of genomics that deals with epigenomic studies. Epigenetic modifications, such as DNA methylation and histone post-translational modifications, regulate gene expression by altering the chromatin state. The “epigenome” refers to the whole genome's chromatin landscape. The prefix epi (which means "over, outside of, or around") denotes that the characteristics are "in addition to" or "from outside of" the traditional genetic basis of inheritance. The epigenomes of cells are studied in this field of omics. Heritable information is not only tracked in the nucleus as DNA sequences, but it is also regulated in part by an epigenetic network. This network includes the three-dimensional structure of DNA, its methylation state, and histone (Margueron and Reinberg 2010) proteins (H2A, H2B, H3, and H4).

Not all phenotypic variation is caused by DNA sequence variation at the phenotypic stage. Some are caused by epigenetic changes in the genome, which include the transfer of gene expression in its "on" or "off" state to future generations. The term "epigenetic alleles" or

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"epialleles" refers to stable chromatin label variants. Heritable epigenetic variation has been shown to influence phenotypic and agronomic traits in both wild and crop plants (Quadrana *et al.* 2014; Zhang *et al.* 2012), and current and historically significant crop traits, such as those in the Green Revolution, have been shown to include epigenomic mechanisms. (Wu *et al.* 2020). However, the potential for using epigenetics to boost crops has not gotten the attention it deserves. A species' ability to adapt to novel or changing conditions is hindered by low genetic variation. Crop plants' genetic diversity has been eroded by domestication and intensive breeding, rendering them more vulnerable to disease (Smykal *et al.* 2018). To help address the challenge of insufficient genetic diversity, natural epigenetic variation and epigenomic processes should be studied as an alternate source of phenotypic variation. DNA methylation, phosphorylation, acetylation, histone and chromatin modifications, and changes in gene expression using siRNA (small interfering RNA) and miRNA (micro RNA) are all examples of epigenomics approaches. Shoot elongation and leaf development, flower initiation, and seed growth are both genetic and epigenetic processes in plants. Stress, paramutations, genome imprinting, silencing of introduced foreign genes, gene silencing caused by viruses, and transposable elements are all examples of plant biological phenomena that include epigenetics (Pikaard and Scheid 2014). The role of epigenomics in plant biotechnology, high-throughput approaches used in this field, and epigenomics applications in crop improvement are discussed in this article.

Approaches

Histone variants, modifications and methods for profiling:

Histones tightly package DNA, arranging, controlling, and protecting it from damage while still allowing replication and transcription. The nucleosome is made up of 146 bps of DNA wrapped around four core histones (H2A, H2B, H3, and H4), each of which contributes two copies to form an octamer. Histone H1, which binds to the nucleosome and short stretches of linker DNA, restricts DNA accessibility and preserves chromatin structure, stabilizes this structure. Epigenetic modifiers are another name for histone modifications. In comparison to transcriptionally inactive heterochromatin regions, histone modifications distributed on heterochromatin are typically associated with addition or removal of methyl group to cytosine

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(Grewal and Jia 2007), while euchromatins are evident with histone hyperacetylation and addition of more than one methyl group to H3 histone (Vaquero *et al.*, 2003 and Dou *et al.*, 2005).

Chromatin immunoprecipitation coupled with DNAmicroarray:

This is the most popular method for describing histone moderations, as well as the proteins that bind to these histones or DNA moderations (Lippman *et al.*, 2004, Robert *et al.*, 2011). The chromatin is broken down into small fragments using vibrations or digestion enzymes in this process. To separate chromatin fragments with histone modifications, ChIP with corresponding unique antibodies is used. Since histone proteins are conserved in eukaryotes, antibodies developed for them can be used to treat a wide variety of species, including animals and plants. Furthermore, microarray and sequencing techniques are used to correct the genomic positions of these modifications on DNA (Egelhofer *et al.*, 2010 and Robert *et al.*, 2011). ChIP-chip has been commonly used in eukaryotes, such as yeast (Barski *et al.*, 2007), where high-performance DNA sequencing techniques have been associated with chromatin immune precipitation. ChIP sequencing uses a similar technique to immune precipitation, but instead of purifying and hybridizing DNA fragments on a chip, DNA fragments are sequenced. This approach is better for analyzing histone moderation patterns at the genomic level because it has a higher resolution than the previous method (Gibson and Spencer 2009).

Profiling of DNA methylation:

DNA methylation occurs in both the uniformity of CG as well as CHG or CHH (H may be A, C, or T) sequence contexts in plants, and includes methylation of the cytosine ring (Kurdyukov and Bullock 2016). The addition of methyl to cytosine is essential for gene silencing via transposons and other epigenetic controls (Zhang *et al.*, 2006). Where the centromere and its neighboring part have more transposons, heterochromatin is more susceptible to methylation (Law and Jacobsen, 2010 and Fujimoto *et al.*, 2012). To maintain genome integrity, most transposons are silenced by DNA methylation. Restriction enzyme-based method, antibody-based method, and sequencing-based method are the three approaches used for DNA methylation profiling.

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Profiling of RNA:

The characterization of cellular RNA pools, which will reveal more RNAs present between genes, RNAs that do not code (ncRNAs), and isoforms of RNAs that occur during splicing, are all examples of RNA profiling (Lister *et al.*, 2008). The various sRNAs (siRNAs, miRNAs, tasiRNAs, and nat-siRNAs) found in large amounts in cells regulate gene expression, heterochromatin silencing, and virus defense (Zhai *et al.*, 2008). On a large scale, high-performance RNA sequencing is being used in combination with the microarray technique for RNA profiling. The use of strand-specific RNA and cDNA libraries is needed for RNA sequencing methods (Lister *et al.*, 2008).

Applications of epigenomics in crop improvement:

Plant breeders can use epigenomics to help them pick positive epigenes, regulate transgene expression, and create new pialleles, which can all help them develop their crops. A detailed understanding of the epigenetic aspect of quantitative trait loci (QTLs) can help predict a plant's physical make-up and lead to steady improvements through selection (Springer, 2013).

Improving Plant Stress Tolerance:

Under stress conditions of development, the accumulation of oxygen free radicals can disrupt various macromolecules such as DNA, RNA, and proteins. In the adversed region, repair mechanisms at the genomic level in plants could result in several differences at the genetic and/or epigenetic level. Abiotic stress causes a rapid and beneficial accumulation of reactive oxygen species (ROS) in plant cells. Recognition of miR398 in Arabidopsis, which acts as a suppressor of the Cu/Zn superoxide dismutases CSD1 and CSD2 genes, contributes to the discovery of the governing role of miRNAs in various stress reactions in plants. Overexpression of a miR398-resistant version of CSD2 enhanced the plant's stress response (Tsaftaris *et al.*, 2008).

Enhancing Viruses and Other Parasites Resistance in Plants:

Environmental conditions have been shown to affect progeny phenotypes. Roberts (1983) conducted a systematic study of the transgenerational effects of damage caused by animals

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(herbivores) and insect pests. In comparison to the offspring of healthy plants (Nicotiana tabacum), he discovered mediated resistance to TMV virus in the offspring of diseased plants (Nicotiana tabacum). More research on Brassica and oilseed rape showed that seeds from plants affected by animals or insect pests have higher levels of phenolics (a protective compound) than seeds from healthy plants (Lammerink *et al.*, 1984).

Role in Yield and Hetrosis:

Several studies have looked at how DNA methylation occurs in hybrids after they have been hybridized (Banaei *et al.*, 2010). According to He *et al.* (2010), the addition of a methyl group to the DNA altered gene expression in hybrids. In Arabidopsis hybrids, Greaves *et al.* (2012) discovered that methylation at the CG complex was higher and methylation at CHH was lower than in the parents, and that changes in DNA methylation occurred most frequently at loci where parental methylation levels were different.

Role in improving transgene stability:

In plant systems, the spontaneous silencing or variable expression of transgenes is a natural occurrence. Transgene silencing, whether caused by several copies of the transgene or endogenous genes, is known as transcriptional gene silencing (TGS) or posttranscriptional gene silencing (PTGS). TGS and PTGS are related to DNA methylation induced by siRNAs in RNA-directed DNA methylation (RdDM) (small interfering RNAs). During transcriptional gene silencing, RdDM inactivates transcription by methylating the promoter (Huettel *et al.*, 2007). It has been discovered that during post-transcriptional gene silencing, the cytosines of the transcribed region are methylated, affecting transcript stability and/or translation rate (Chawla *et al.*, 2007).

Conclusions:

The most advanced DNA sequencing technologies have aided in the investigation of the complex field of epigenomics. The three interacting mechanisms that can control gene expression or silencing are DNA methylation, histone alteration, and RNA interference. Resolving the relationships between these epigenetic components would lead to a slew of new ideas that would emerge quickly. Understanding plant developmental gene regulation,

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reaction to environmental stimuli, and better use of natural variation for crop improvement can all benefit from epigenomic knowledge. The epigenomic plays a direct role in reversing transgene silencing, which is a common issue in transgenic growth. The peculiar role of EIS in plant genetic responses to various biotic and abiotic stresses, as well as in preserving heterosis, cultivar yield stability, and resource utilization, should be thoroughly investigated.

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