

Molecular Mechanisms of Auxin Action in Plants

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Molecular Mechanism # 1. An Auxin-binding Receptor has been Discovered:

A possible auxin binding receptor protein has been identified in plants which is called as auxin-binding protein 1 (ABP1). This protein appears to be a dimer made of two polypeptides of about 22 kD each. ABP1 is located in lumen of endoplasmic reticulum (ER), but it is believed to be active on surface of cell.

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Molecular Mechanism # 2. Auxin Causes Rapid Changes in Gene Activity:

The idea that auxin-induced cell growth may involve expression of certain genes had been given by scientists in early 1960s. Subsequently in early 1970s, it was shown that auxin could stimulate proton extrusion and growth with a lag time of 10-12 minutes only. At that time, the minimum lag time of 10 minutes was supposed to be too short for induction of protein synthesis i.e., gene expression and it was held that auxin induced growth was a result of direct interaction of auxin with membrane component rather than expression of certain specific genes.

Advancement of molecular biology techniques in recent past including the use of specific complementary DNA (cDNA) probes in hybridization studies and use of 'plus and minus' screening methods have enabled scientists to detect rapid changes in level of not only translatable m-RNA, but total or abundance of m-RNAs also with high degree of precision.

Studies made with such techniques have now clearly established that gene expression is involved in auxin-induced growth. The evidences indicate that the control is at transcription. Studies made with nuclear run off experiments which measure transcription directly have shown auxin to stimulate transcription of specific m-RNAs in 2-5 minutes only.

Molecular Mechanism # 3. Some Signalling Intermediates (Secondary Messengers) have been Implicated in Auxin Action:

Molecular mechanism of action of auxin (phytohormone) is not as clearly understood as those of animal hormones. However, studies of signal transduction pathways have implicated Ca^{++} , intracellular pH, cyclic AMP (cAMP) and Iyso-phosphatidylcholine as signalling inter-

mediates (secondary messengers) in auxin action. Cyclic AMP and MAP (Mitogen Activated Protein) Kinases are believed to be signalling intermediates in auxin induced cell division. (The primary messenger is the hormone auxin itself).

Molecular Mechanism #4. There are Two Categories of Auxin-Induced Genes-Early Genes and Late Genes:

After binding with its receptor, auxin initiates signal transduction pathway which in turn activates certain specific pre-existing transcription factors. The latter enter the nucleus and stimulate the expression of specific genes. Such genes whose expression depends upon preexisting transcription factors are called as early genes or primary response genes.

If the proteins required for the expression of early genes are already present (pre-exist) in the cell at the time of exposure to auxin treatment. Therefore, the expression of early genes is not blocked by inhibitors of protein synthesis such as cycloheximide (an antibiotic). The time required for expression of early genes may be as short as few minutes or it may extend to hours.

By analogy with animal hormones, auxin-induced early genes have basically three functions:

(i) Some of the early genes encode proteins which regulate transcription of late genes or secondary response genes.

Late genes are required for long term responses to auxin. Expression of late genes require de novo synthesis of proteins and therefore, it is blocked by inhibitors of protein synthesis such as cycloheximide.

(ii) Some of the early genes are involved in intercellular signalling.

(iii) Some of other early genes are involved in stress adaptation.

Five major families of auxin-induced early genes have been identified in plants

(i) Aux/IAA gene family.

(ii) SAUR (small auxin up-regulated) gene family.

(iii) GH₃ gene family.

(iv) Genes encoding glutathione-s-transferase (GST) like proteins

(v) Gene family encoding aminocyclopropane-l-carboxylic acid (ACC) synthase.

Auxin-induced early genes are believed to be negatively regulated by short lived repressor proteins. Auxin causes degradation of repressor proteins (through ubiquitination pathway) so that transcription factors become active and transcription of early genes is initiated.

Ubiquitination pathway:

This is very common non-lysosomal pathway of protein degradation that regulates cytosolic protein turnover in eukaryotic cells including plants. According to an estimate, more than 90% of short lived proteins are degraded in eukaryotic cells via ubiquitination pathway.

Protein degradation by ubiquitination pathway is accomplished by two main components, (i) A small highly conserved protein called ubiquitin that is a 76-amino acids polypeptide and (ii) A large proteolytic enzyme complex called proteasome that is an oligomeric 26S structure with a molecular mass of more than 1.5 megadalton. This itself consists of two parts, a 20S core proteasome and a 19S regulatory complex.

The function of ubiquitin is to 'mark' the target protein (by forming a covalent bond with the latter) after being activated by ATP and involving three enzymes viz., E₁, E₂ and E₃ for degradation by large enzyme complex proteasome which specifically recognises the 'marked' or 'tagged' protein molecule).

(Two Israeli scientists, A. Ciechanover and A. Hershko along with an American scientist I. Rose had done pioneer work in discovery and elucidation of the ubiquitination pathway for degradation of unwanted proteins for which they were jointly awarded the Nobel Prize of 2004 in Chemistry).

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