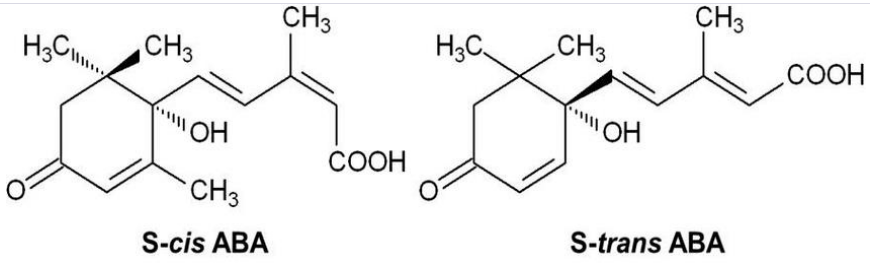
ABSCISIC ACID (A Seed Maturation and Antistress Signal)

Abscisic acid has been found to be a ubiquitous, plant hormone in vascular plants. It has been detected in mosses. Several genera of fungi make ABA as a secondary metabolite. Within the plant, ABA has been detected in every major organ or living tissue from the root cap to the apical bud. ABA is synthesized in almost all cells that contain chloroplasts or amyloplasts. ABA is a 15-carbon compound that resembles the terminal portion of some carotenoid molecules. The orientation of the carboxyl group at carbon 2 determines the *cis* and *trans* isomers of ABA. It appears to have a more limited range of specific effects than auxins, gibberellins, and cytokinins. The name is based on the once held belief that it was involved in the abscission of leaves and other organs. It now appears to have nothing to do with abscision, but the name has stuck.



S-*cis* ABA S-*trans* ABA

The conversion between cis-ABA and trans-ABA can be influenced by various environmental factors, including light conditions. In certain situations, such as exposure to light, cis-ABA can isomerize to trans-ABA, and vice versa. The specific biological activities of these isomeric forms are an active area of research, and their roles in plant responses to environmental stimuli are still being elucidated.

The primary functions of ABA are:

1. Prohibiting precocious germination and promoting dormancy in seeds.
2. Inducing stomatal closure and the production of molecules that protect cells against desiccation in times of water stress.

ABA Can Be Inactivated by Oxidation or Conjugation

ABA **biosynthesis** and **concentrations** can fluctuate dramatically in specific tissues during development or in response to **changing environmental conditions**.

1. In **developing seeds**, for example, ABA levels can **increase 100-fold** within a **few days** and **then decline** to vanishingly **low levels** as **maturation** proceeds.
2. Under conditions of **water stress**, ABA in the **leaves** can **increase 50-fold** within **4 to 8 hours**. Upon **rewatering**, the ABA level **declines to normal** in the same amount of time.

Biosynthesis is not the only factor that regulates ABA concentrations in the tissue. As with other plant hormones, the concentration of free ABA in the cytosol is **also regulated by degradation, compartmentation, conjugation, and transport**.

For example, cytosolic **ABA increases during water stress** as a result of **synthesis** in the leaf, **redistribution** within the **mesophyll cell**, **import from the roots**, and **recirculation from other leaves**. The concentration of **ABA declines after rewatering** because of **degradation** and **export** from the leaf, as well as a **decrease in the rate of synthesis**.

Inactivation of ABA:

1. A **major cause of the inactivation of free ABA** is **oxidation**, yielding the unstable intermediate **6-hydroxymethyl ABA**, which is rapidly **converted** to **phaseic acid (PA)** and **dihydrophaseic acid (DPA)**. PA is usually inactive, or it exhibits greatly reduced activity, in bioassays. However, **PA can induce stomatal closure** in **some species**, and it is as **active as ABA** in **inhibiting gibberellic acid**–induced α-amylase production in barley aleurone layers.

These effects suggest that **PA** may be **able to bind to ABA receptors**. In contrast to PA, **DPA** has **no** detectable **activity** in any of the bioassays tested.

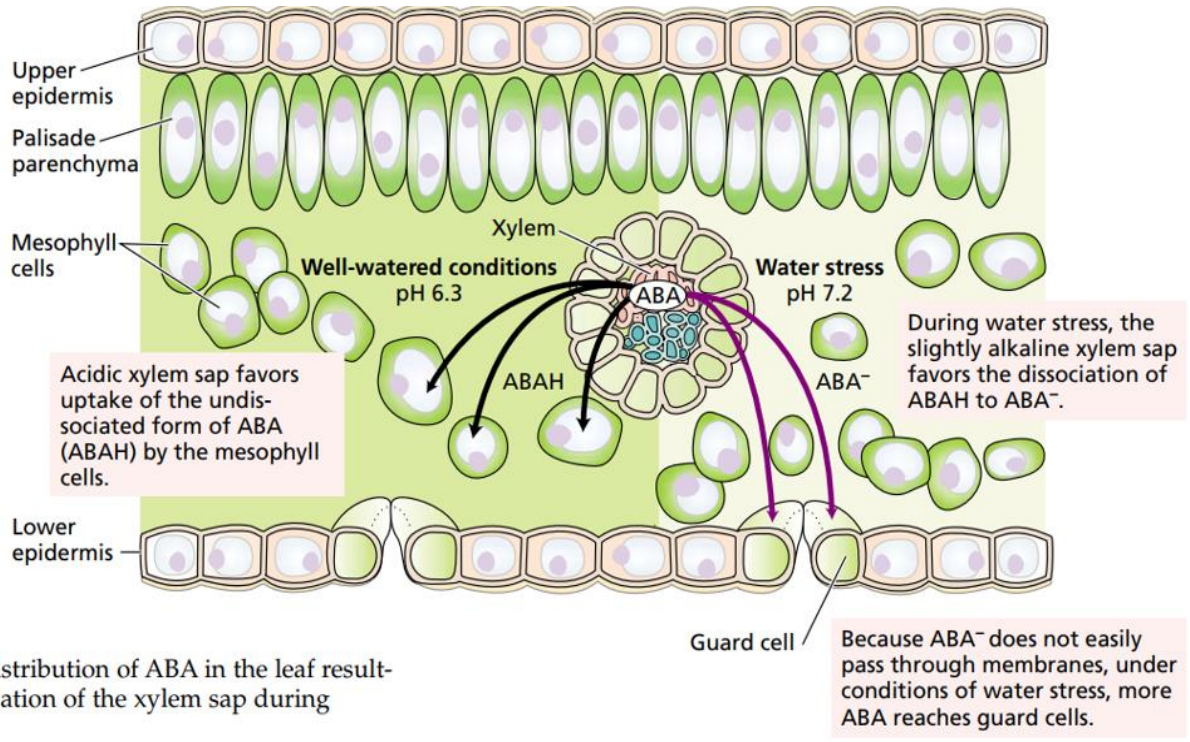
2. Free ABA is also inactivated by **covalent conjugation** to another molecule, such as a **monosaccharide**. A common example of an ABA conjugate is **ABA-β-D-glucosyl ester** (**ABA-GE**). Conjugation **not only** renders ABA **inactive** as a hormone; it also **alters** its **polarity** and **cellular distribution**. Whereas free ABA is localized in the cytosol, **ABA-GE accumulates in vacuoles** and thus could theoretically serve as a **storage form** of the hormone. **Esterase enzymes** in plant cells could **release free ABA** from the **conjugated** form. However, there is **no evidence** that **ABA-GE hydrolysis** contributes to the **rapid increase** in ABA in the leaf during water stress. When plants were **subjected** to **a series of stress and rewatering cycles, the ABA-GE** concentration **increased** steadily, suggesting that the **conjugated form** is **not broken-down** during water stress**.**

1.2 ABA Is Translocated in Vascular Tissue

ABA is transported by both the **xylem** and the **phloem**, but it is normally much **more** abundant in the **phloem sap**. When **radioactive ABA** is applied to a leaf, it is transported both **up** the stem and **down** toward the roots. **Most** of the **radioactive ABA** is found **in** the **roots** within **24 hours**. **Destruction** of the **phloem** by a **stem girdle prevents ABA accumulation in the roots**, indicating that the hormone is transported in the phloem sap. **ABA synthesized** in the **roots** can also be **transported to** the **shoot** via the **xylem**. Whereas the **concentration of ABA** in the **xylem** sap of **well-watered sunflower** plants is

between **1.0- 15.0 nM**, the ABA concentration in **water stressed sunflower** plants increases to as much as **3000 nM** (3.0 μM).

The magnitude of the **stress induced change in xylem ABA content varies** widely among species, and it has been suggested that ABA **also is transported in a conjugated form**, then **released by hydrolysis in leaves**. However, the postulated hydrolases have yet to be identified. As **water stress begins**, some of the **ABA carried by the xylem** stream is **synthesized in roots** that are in direct **contact with the drying soil**. Because this transport can **occur before** the **low water potential** of the soil **causes any measurable change in the water status of the leaves**, **ABA** is believed to be a **root signal** that helps **reduce the transpiration rate** by **closing stomata** in leaves. Although a concentration of **3.0 μM ABA in the apoplast is sufficient to close stomata**, not all of the ABA in the xylem stream reaches the guard cells. Much of the ABA in the transpiration stream is taken up and metabolized by the mesophyll cells. During the early stages of water stress, however, the **pH of the xylem sap** becomes **more alkaline**, increasing from about pH **6.3 to** about pH **7.2**.



Redistribution of ABA in the leaf resulting from alkalinization of the xylem sap during water stress.

Developmental and physiological effects of ABA

Abscisic acid plays primary regulatory roles in the initiation and maintenance of seed and bud dormancy and in the plant's response to stress, particularly water stress. In

addition, ABA influences many other aspects of plant development by interacting, usually as an antagonist with auxin, cytokinin, gibberellin, ethylene, and brassinosteroids.

1. **ABA Promotes Desiccation Tolerance in the Embryo**

An important function of ABA in the developing seed is to promote the acquisition of desiccation tolerance. Desiccation can severely damage membranes and other cellular constituents. During the **mid- to late stages** of seed development, specific mRNAs accumulate in embryos at the time of high levels of endogenous ABA. These mRNAs encode so-called **late-embryogenesis-abundant** (**LEA**) **proteins** thought to be involved in **desiccation tolerance**. Synthesis of many **LEA proteins**, or related family members, can be **induced by ABA treatment** of either **young embryos** or **vegetative tissues**. Thus, the synthesis of most LEA proteins is under ABA control.

2. **ABA Promotes the Accumulation of Seed Storage Protein during Embryogenesis**

Storage compounds accumulate during **mid- to late embryogenesis**. Because **ABA** levels are still **high,** ABA could be affecting the **translocation of sugars and amino acids**, the synthesis of the reserve materials, or both.

Studies in **mutants impaired** in both **ABA synthesis** and response showed **no effect** of ABA on **sugar translocation**. In contrast, ABA has been shown to **affect** the amounts and composition of **storage proteins**. For example, exogenous ABA promotes accumulation of **storage proteins** in cultured embryos of many species, and some **ABA-deficient** or -insensitive mutants have reduced storage protein accumulation. However, storage protein synthesis is also **reduced** in other seed developmental mutants with normal ABA levels and responses, indicating that ABA is only **one of several signals** controlling the **expression of storage protein genes** during embryogenesis.
ABA not only regulates the accumulation of storage proteins during embryogenesis; it can also maintain the **mature embryo** in a **dormant state** until the environmental conditions are optimal for growth. Seed dormancy is an important factor in the adaptation of plants to unfavorable environments.

3. **Seed dormancy and germination are controlled by the ratio of ABA to gibberellic acid (GA)**

Embryo dormancy is thought to be due to the presence of inhibitors, especially **ABA**, as well as the **absence of growth promoters**, such as **GA**. Maintenance of **dormancy** in imbibed seeds requires **de novo ABA biosynthesis**, and the loss of embryo dormancy is often associated with a **sharp decrease** in the ratio of **ABA to GA**. The levels of ABA and GA are regulated by their synthesis and catabolism, which are catalyzed by specific isozymes whose expression is controlled by developmental and environmental factors. In addition to the **ABA-GA antagonism** affecting seed dormancy, ABA inhibits the GA-

induced synthesis of hydrolytic enzymes that are essential for the breakdown of storage reserves in germinating seeds. For example, GA stimulates the aleurone layer of cereal grains to produce **α-amylase** and other hydrolytic enzymes that break down stored resources in the endosperm during germination. ABA inhibits this GA-dependent enzyme synthesis by inhibiting the **transcription of α-amylase mRNA**. ABA exerts this inhibitory effect via at least two mechanisms, one **direct** and one **indirect**:

4. **ABA promotes root growth and inhibits shoot growth at low water potentials**

Despite the traditional view of ABA as a growth inhibitor, endogenous ABA restricts shoot growth only under water stress conditions. Moreover, under these conditions, when ABA levels are high, endogenous ABA exerts a strong positive effect on **primary root growth by suppressing ethylene production**. The overall effect is a dramatic increase in the **root: shoot ratio** at low water potentials, which, along with the effect of ABA on stomatal closure, helps the plant cope with water stress. Furthermore, the **temporary inhibition of lateral root outgrowth** promotes exploration of new areas of soil, and permits replacement of dehydrated laterals following rehydration.

5. **ABA greatly accelerates the senescence of leaves, thereby increasing ethylene formation and stimulating abscission**

Abscisic acid was originally isolated as an abscission causing factor. However, it has since become evident that ABA stimulates abscission of organs in only a few species and that the hormone primarily responsible for causing abscission is ethylene. On the other hand, ABA is clearly **involved in leaf senescence**, and through its promotion of senescence it might **indirectly increase ethylene** formation and **stimulate abscission**. Leaf senescence has been studied extensively. Leaf segments senesce faster in darkness than in light and they turn yellow as a result of chlorophyll breakdown. In addition, the breakdown of proteins and nucleic acids is increased by the stimulation of several hydrolases. ABA greatly accelerates the senescence of both leaf segments and attached leaves.

6. **ABA accumulates in dormant buds, inhibiting their growth; it may interact with growth-promoting hormones**

ABA was originally suggested as the **dormancy-inducing hormone** because it **accumulates in dormant buds** and decreases after the tissue is exposed to low temperatures. However, later studies showed that the ABA content of buds does not always correlate with the degree of dormancy. As we saw in the case of seed dormancy, this apparent discrepancy might **reflect interactions between ABA and other hormones**; perhaps bud dormancy and growth are regulated by the balance between bud

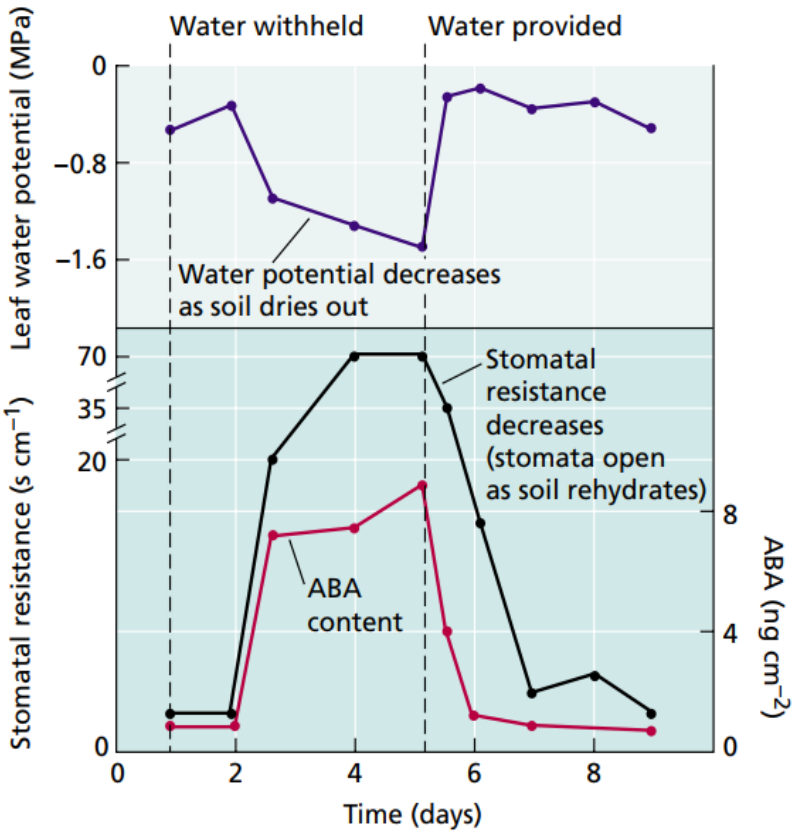
growth inhibitors, such as **ABA**, and growth-inducing substances, such as **cytokinins** and **gibberellins**.

7. Abscisic acid closes stomata in response to water stress

ABA accumulates in water-stressed (that is, wilted) leaves and exogenous application of ABA is a powerful **inhibitor of stomatal opening**. The precise role of ABA in stomatal closure in water-stressed whole plants has, however, been difficult to decipher with certainty. This is because **ABA** is ubiquitous (**everywhere**), often occurring in **high concentrations** in non-stressed tissue. Also, some early studies indicated that stomata would **begin to close before increases in ABA content could be detected**.

Stomatal closure does not always rely on the perception of water deficits and signals arising within the leaves. In some cases, it appears that the stomata close in response to soil desiccation well before there is any measurable reduction of turgor in the leaf mesophyll cells. Several studies have indicated a **feed-forward control system** that originates in the roots and transmits information to the stomata. In these experiments, plants are grown such that the roots are equally **divided between two containers** of soil.

Water deficits can then be introduced by withholding water from one container while the other is watered regularly. Control plants receive regular watering of both containers. Stomatal opening along with factors such as ABA levels, water potential, and turgor are compared between half-watered plants and fully watered controls. Typically, stomatal conductance, a measure of stomatal opening, declines within a few days of withholding water from the roots, yet there is no measurable change in water potential or loss of turgor in the leaves. Furthermore, **ABA is readily translocated from roots to the leaves** in the transpiration stream, even when roots are exposed to dry air. These results suggest that **ABA is involved in some kind of early warning system** that communicates information about soil water potential to the leaves.



Changes in water potential, stomatal resistance and ABA content in corn in response to water stress (source: Taiz L., Zeiger E., 2010)