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## TWINS AND ITS ROLE IN CROP IMPROVEMENT

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**T**wins in animals, can be either monozygotic (identical) or dizygotic (fraternal), depending on whether they develop from a single fertilized egg or two separate eggs fertilized by different sperm cells. This phenomenon is well-known in humans and many other animal species. In plants, the occurrence of twins or polyembryony is less common but still fascinating. Just like in animals, plant twins can be either genetically identical or fraternal. The process of fertilization and embryogenesis in higher plants is indeed complex and tightly regulated, similar to animals. The role of dehydroascorbate reductase (DHAR) in producing twin and triplet seedlings in plants is an intriguing discovery. DHAR is an enzyme that plays a crucial role in recycling vitamin C in both plants and animals. Increasing the level of DHAR can result in higher levels of vitamin C in plants, leading to the production of multiple seedlings from a single seed (Chen and Gallie, 2012). This has significant implications for agriculture and crop production, as it could potentially be used to increase the yield of high-value crops. Additionally, having multiple seedlings from a single seed may also enhance the survival chances of some plant species, as it provides a form of redundancy and resilience in case some of the seedlings encounter unfavorable conditions. Overall, the parallels between the production of twins in plants and animals, as well as the potential applications of DHAR in agriculture, highlight the fascinating complexity of biological processes across different species and the potential for scientific discoveries to benefit various fields, including crop production and genetics.

### Biology of Twinning

Regarding spontaneous or natural monozygotic twinning, a recent theory posits that monozygotic twins are formed after a blastocyst essentially collapses, splitting the progenitor cells (those that contain the body's fundamental genetic material) in half, leaving the same genetic material divided in two on opposite sides of the embryo. Eventually, two separate

fetuses develop. Spontaneous division of the zygote into two embryos is not considered to be a hereditary trait, but rather a spontaneous or random event.

### Occurrence of Twinning in Plants

In the context of plant reproduction, twinning refers to the formation of twin seedlings. These twin seedlings can arise in two main ways:

a. **Division of Single Zygote:** Similar to how twins can develop in animals, one way twin seedlings can form is when a single zygote divides into two distinct embryos.

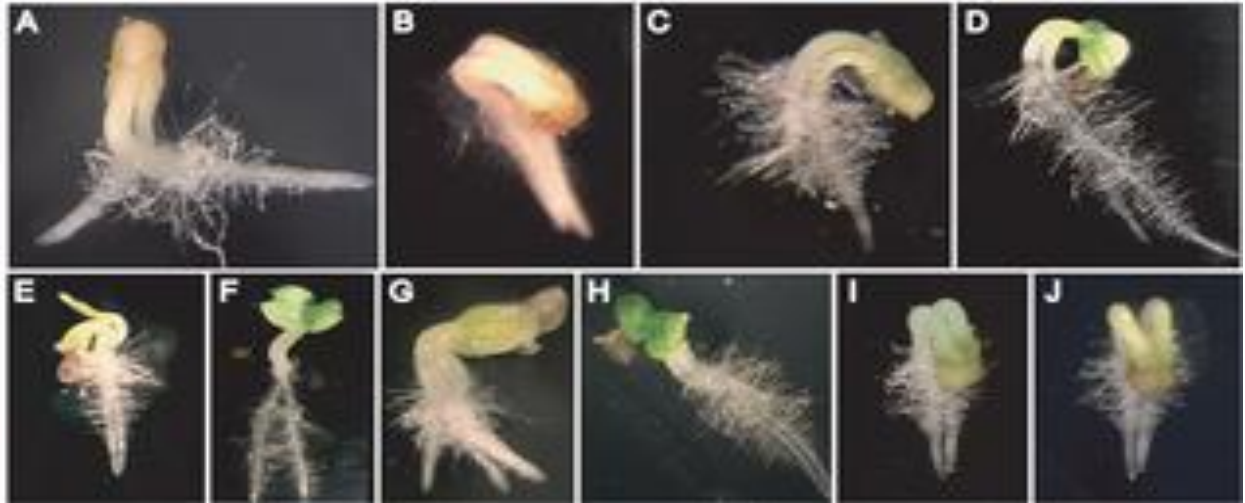
b. **Apomixis:** Apomixis is another way in which twin seedlings can develop. Apomixis is a form of asexual reproduction in plants where embryos develop without the need for fertilization.

**Embryo Development:** In plants, embryo development begins with the transverse division of a zygote into two distinct cells—an apical cell that becomes the proembryo and a basal cell that gives rise to the suspensor. Mutations or changes in cell division during this process can lead to the development of twin embryos.

**Role of Vitamin C (Ascorbic Acid, Asc):** Injecting plant ovaries with vitamin C (ascorbic acid) can induce twinning in some plant species (**Figure 1**). Vitamin C appears to regulate cell polarity during embryo development and can lead to the formation of genetically identical twin embryos. This process is similar to the role of the gene DHAR (dehydroascorbate reductase), which affects cell division and can induce twinning.

**Developmental Timing:** It's important to note that twinning induced by vitamin C or DHAR is developmentally limited to the early stages of plant growth, typically within the first two days after pollination.

**Genetic Factors and Mutations:** Mutations in genes like TWIN can result in the formation of additional embryos through the transformation of suspensor cells early in embryogenesis (Gunaga and Vasudeva, 2008). Abnormalities leading to twinning or polyembryony in plants can be attributed to various factors, including developmental errors during ovary development, fertilization, genetic factors, or mutations.



**Fig. 1:** DHAR induced twinning in tobacco

Depending on their origin, twin seedlings may be diploid/diploid, diploid/haploid or haploid/haploid. The frequency of observed polyembryony is extremely low and species-dependent; for instance, the frequency was reported at 4% in soybean, 0.37% in Capsicum, 0.10% in maize, and 0.054% in grape (Chen and Gallie, 2012). Twin seedlings usually show one of the following combinations: diploid-diploid, diploid-haploid, haploid-haploid, and diploid-triploid. So far many reports recorded the polyembryony in several tropical tree species like *Acacia farnesiana*, *Robinia pseudocasia*, *Terminalia Arjuna*, *Tectona grandis*, *Santalum spicatum*, *Mangifera indica*, *shorea robusta*, *Dalbergia sissoo*, *Bombax ceiba*, *Putranjiva roxburghii*, *Nathopodytes nimmoniana*, *Saraca asoca*, *Garcinia indica* and *Mammea suriga*.

Another study, the loss of AMP1 (ALTERED MERISTEM PROGRAM 1) function; a gene required for synergid cell fate during Arabidopsis female gametophyte development leads to supernumerary egg cells at the expense of synergids, enabling the generation of dizygotic twins (Kong et al. 2015). Alternatively the artificial delivery of supernumerary sperm cells in tetraspore (tes) pollen, enabling the formation of twin plants. The twin-embryo phenotype of amp1 mutants also sheds light on the boundary conditions for double fertilization in plant reproduction, which involves two sperm cells and the four cells at the micropylar end of the female gametophyte: two synergids, one egg cell, and one central cell. Ovules with twin embryos but no endosperm as well as ovules with one developing embryo and endosperm plus one persisting unfertilized egg cell strongly suggest that the two sperm cells of a fertilizing pollen tube are free to choose their mating partners.

Recently, the researchers discovered that the injecting plant ovaries with vitamin C is sufficient to produce twins or triplets in many plant species (**Figure 2**). In tobacco, the vitamin C causes the zygote, the fertilized egg, to divide into two or even three fertilized egg cells before these cells proceed through subsequent stages of development to produce twins or triplets. The twins and triplets produced in tobacco plants when vitamin C was increased were true twins or triplets as they were genetically identical. Because the early stages of embryo development are so conserved among plant species, the vitamin C will have a similar effect in almost any plant. These study suggested that the critical effect of vitamin C is on the very first stage during cell division.

The formation of embryo without sexual process is called apomixis and seed is called apomictic seed, the condition is called polyembryony. Polyembryony is the occurrence of more than one embryo in a seed which consequently results in the emergence of multiple seedlings. The additional embryos result from the differentiation and development of various maternal and zygotic tissues associated with the ovule of seed. Polyembryony usually associated with nucellar embryony because it frequently results in polyembryonic seeds from which multiple seedlings germinate. Based on genetic composition the polyembryony classified into I. Gametophytic: multiple embryos arises from the gametic cells of the embryo sac (synergid, antipodal) after or without fertilization. In this case haploid embryos are formed. The second one is Sporophytic where the multiple embryos arise either from zygote or from sporophytic cells of ovule (nucellus, integument) and the resulting embryo will be diploid.



a. Normal seedling

b. Twin seedling

**Fig. 2:** Vitamin C induced twinning in Terminalia

## Role of Twins in Crop Improvement

**1. Genetic Uniformity:** The ability to produce genetically identical seedlings from a single seed can be highly advantageous in crop improvement. It ensures that desirable traits in a crop can be preserved and propagated consistently across generations. This is especially important for maintaining the quality and characteristics of high-value crops.

**2. Increased Fertility:** Polyembryony can be particularly valuable when the natural fertility rate of a crop is low. By producing multiple embryos in a single seed, the chances of successful germination and plant establishment are enhanced. This is beneficial for crops where increasing yield or maintaining consistent production is a priority.

**3. Nutrient Enhancement:** In cases like corn, where the production of multiple embryos can significantly boost protein content, polyembryony can be instrumental in increasing the nutritional value of the crop. This can have a direct impact on the crop's suitability for human or animal consumption.

**4. Survival Chances:** Having multiple seedlings germinate from a single seed can also improve the per-seed survival chances for some plant species. It provides a form of insurance against adverse environmental conditions or pest attacks, as not all seedlings may be affected in the same way.

**5. Crop Resilience:** In situations where crops face challenges like diseases or pests, having genetically identical seedlings can make it easier to develop resistant varieties. This can aid in crop resilience and reduce the need for chemical interventions.

**6. Resource Efficiency:** Producing multiple seedlings from a single seed can be more resource-efficient than planting individual seeds. It can reduce the amount of seed required for planting and lead to cost savings in agriculture.

## Conclusion

In conclusion, twin seedlings or polyembryony offer various benefits in crop improvement, including genetic uniformity, increased fertility, enhanced nutrient content, improved survival chances, and resource efficiency. These advantages can contribute to higher crop yields, improved crop quality, and more sustainable agricultural practices.

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