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Reproductive isolation that leads to speciation in flowering plants

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ABSTRACT

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In sexually reproducing organisms, two different species are always separated by reproductive isolating mechanism, which limits or prevents the interchange of genes between them. The focus here is on the reproductive isolation which is a key mechanism in the process of speciation or origin of species. Reproductive barriers or isolating mechanisms occur at both the pre-zygotic and post-zygotic levels. Pre-zygotic isolating mechanism may involve ecological, temporal and floral isolation and isolation through autogamy and incompatibility. The flower and pollination interaction that leads to speciation is also emphasized. However, post-zygotic isolating mechanism is discussed here along with embryo lethality due to genotypic interaction followed by hybrid mortality, sterility or hybrid breakdown.

Key-words: Pollinator, post-zygotic isolation, pre-zygotic isolation, reproductive barriers, hybrid sterility, autogamy.

INTRODUCTION

Reproduction is most important and necessary function of all life. In plants, though there are different modes of reproduction, sexual reproduction is always considered in species formation. Whenever we think about speciation in flowering plants, three concepts should be clear in our mind. These are: concept of species, concept of speciation and mechanism of reproductive isolation. These three are so closely related to each other that such studies in nature or in experiments cannot be possible without going through them. The origin of new species is relatively rare event and requires a succession of events that are usually distributed over a considerable time span. It takes place only when both ecological conditions and the genetic structure of a population are particularly favourable for it. Generally, the individuals of a group of plants that are morphologically looking similar are grouped under a species, which was the basis for Linneaus' original classification. It is still broadly accepted and applicable. Though taxonomists are routinely using this morphological species concept, it was criticized by the biologists as there were many examples in which members of a species are similar with each other but unable to interbreed. It means that in spite of their morphological similarity some biological barriers persist. Thus another concept, i.e. 'biological concept of species' was evolved. It has undergone a number of changes over the years. The earliest precursor was Du Rietz (1930). A few years later, Dobzhansky (1937) defined the species. Mayer (1942) gave final definition of biological species concept which attracted biologists and became widely adopted. According to him, "Species are groups of actually or potentially interbreeding natural populations which are reproductively isolated from other such groups". The

emphasis in this definition is on what happens in nature. However, when one such species gives rise to two or more species, as a result of change in gene pool, speciation takes place. Speciation is actually an evolution of reproductive isolating mechanism that essentially prevents gene exchange between newly arising species. In nature, when individuals of a population of flowering plants are prevented from freely exchanging genes by reproductive barriers, which may be operative prior to flowering, during flowering/pollination/fertilization and even after fertilization, speciation takes place. In the present study, main emphasis has been given on the evolution of different types of reproductive isolation in flowering plants and their role during speciation because reproductive isolation is the key mechanism in the process of speciation. Though reproductive isolation is a broad subject of debate and speculation, as it has still some unknown facts in nature, many forms of reproductive isolation exist that can block gene flow between populations in different ways. They may be of geographical or sympatric (in same population). The sympatric speciation is the most reliable and widely discussed with new approaches. There are several different types of reproductive isolating mechanisms, which are classified according to when in the life cycle of the organism isolation occurs. Isolation can occur before fertilization (pre-zygotic barriers) or after fertilization (post-zygotic barriers). Such speciation may be the consequences of some pre- and post-zygotic reproductive isolating mechanism, i.e. barriers to gene flow may occur before fertilization or after fertilization.

PRE-ZYGOTIC ISOLATION MECHANISM

The pre-zygotic mechanism includes all the factors associated with habit, habitat, floral behaviour, process of pollination and fertilization whereas post-zygotic mechanism includes hybrid sterility, mortality and hybrid breakdown. Pre-zygotic mechanism is divided into following types:

Ecological or habitat isolation: Formation of new species takes place when barriers to gene flow, i.e. reproductive isolation, evolve between populations as a result of ecologically based divergent selection. Here, reproductive isolation or barrier to gene flow evolved between the species as a consequence of adaptation to different environment or ecologically based divergent selection. Divergent selection may arise due to differences between populations in their habitat, climate, resources and competition with other existing populations (Schluter 2001). Though divergent selection between environment is common in allopatric speciation (geographical isolation), it may also arise between sympatric populations (growing in same population) with significant ecological variation within a single geographic area. The occurrence of ecotypic variation within species gives a promising lead on the possible step in speciation. Reduced gene flow by habitat isolation may lead to speciation, e.g. due to climate and soil differences. *Streptanthus niger* Greene (Tiburon Jewel flower – family Brassicaceae) is now geographically and genetically cut off from its nearest morphological congener *S. glandulosus* subsp. *pulchellus* (Greene) Kruckeb. (Kruckberg 1957). In the ecological speciation, reproductive isolation evolves as a by-product mechanism of adaptation to different habitats and ecologically isolated species, when crossed artificially, give rise vigourous fertile hybrids in both the F_1 and later generations, suggesting no internal isolating barrier between them. Since their natural habitats are very different, they never get crossed and remained as two distinct species.

Temporal isolation: Isolation of two populations due to seasonal or timing differences in flowering periods is often responsible for speciation. Populations may flower at different seasons or different times of the day, e.g. the three tropical orchid species of the genus *Dendrobium*, each flower for a single day. The flowers open at dawn and wither by night fall. The flowering occurs due to some meteorological stimuli such as sudden storm on a hot day. One species flowers after 8 days, another after 9 days and third flowers after 10 or 11 days of such stimuli. However, when one species flowers, another withered which prevents interspecific fertilization. Thus difference in flowering periods acts as a reproductive isolating mechanism which leads to speciation (Encyclopedia Britannica 2007). Similarly, some taxa may achieve a high degree of isolation by

opening their flowers at different times of the day, e.g. in desert species of *Oenothera* (Onagraceae), some species open their flowers in the morning and other in the evening (Raven 1962). These differences develop reproductive isolation among the species, which prevents interbreeding. Sometimes, lack of synchrony in pollen release and female receptivity also acts as a reproductive isolating mechanism in anemophilous (wind pollinated) species, e.g. *Pinus radiata* D. Don (Monterey Pine) and *P. muricata* D. Don (Bishop Pine) are sympatric but shed their pollen at different times and do not interbreed (Stebbins 1950).

Among flowering plants, evolution of floral characters has long been considered as the important factor for achieving reproductive isolation and species diversity. Generally, entomophilous (insect pollinated) plants exhibit diversity in their floral forms, colour and size. The floral characters may affect reproductive isolation through pollination by different pollinators as pollinator selection always depends on the different floral characteristics, e.g. genus *Aquilegia* L. (Ranunculaceae) has wide variation in its species according to habitats, floral morphology, colour and pollinators. The two important species of *Aquilegia,* viz. *A. formosa* Fisch. ex DC. (Western columbine) and *A. pubescens* Coville (Sierra columbine) have differences in their floral morphology as well as pollinators. *A. formosa* has red flowers and exerted style and visited by hummingbirds whereas *A. pubescens* has white scented flowers with erect style and stamens and hawk moth is its pollinator. Between these two species of *Aquilegia*, genetic or reproductive isolation seems to be maintained by the adaptation of their flowers to two different pollinators (Grant 1952, Hodges et al. 2002). In such cases, barrier to interbreeding or reproductive isolation can only be maintained because of two different visiting pollinators. A common vector for both the species can break the isolating mechanism but it is not possible because the structure and behaviour of a particular pollinator, which is comfortable with a particular flower structure /colour, is not suitable for other. Pollinators are potentially powerful force in plant speciation. This is the reason that in spite of growing in the same population the two cannot interbreed and remain as separate species. It is obvious that pollinator preference can lead to reproductive isolation between flowering plant species, and in some cases, pollinator may provide good example of evolutionary origin of two flower features that lead to new species formation. Such type of co-evolution was reported in two closely related species of *Mimulus* (Monkey flower - Scrophulariaceae) where *M. lewisii* Pursh (Lewis's monkey flower / Purple monkey flower) is bee pollinated whereas *M. cardinalis* Douglas ex Benth. (Scarlet monkey flower) is pollinated by hummingbirds. Species pollinated by hummingbird have repeatedly and independently evolved tubular flowers, often red or orange coloured and different from their congeners (Charlesworth & Charlesworth 2000). Thus plant and pollinator interaction plays a central role in isolating plant species reproductively, which derives speciation. The most specialized correlation between pollinator and flower, responsible for extensive speciation, is seen in orchid family. The flowers of this family are highly specialized and the pollination takes place by a particular pollinator having a specific mode of pollen transportation according to their body characteristics. In most of the orchids, an extraordinary but efficient reproductive isolating mechanism is found. In such species, the flowers mimic in shape the females of certain species of wasps and bees (Dodson 1967, Ledford 2007). The various species of genus *Orphrys* L. has good example of such type of pollination. The flowers of these species have resemblance to the female of different species of bees of the genera *Andrena* and *Scolea*. The male bees are attracted to the particular flowers of their female appearance and take part in the pollination of particular species of orchid (Stebbins 1950). Such co-evolution of flowers and pollinators is responsible for pollination of specific flower by specific pollinator and makes the species reproductively isolated and initiates speciation.

Isolation through flower odour: Flower odour also plays a major role in reproductive isolation. Some insects are very picky about the flower odour/scent. They respond to a particular scent of a flower and pollinate it. Some orchids have a quality to change their pollinator by changing their flower odour /scent.

Chiloglottis trapezeformis Fitzg., an Australian orchid (Broad-lip bird orchid) which tricks male thymine wasp (*Neozeleboria cryptoides*), produces an entirely different chemical 'Chiloglottone' which is a volatile sex pheromone released by female wasp to attract their male partners. Some species of *Chiloglottis* growing together are similar in appearance, location and flowering phenology but can not interbreed naturally because both the species produce two different chemicals (pheromones) to attract two different species of wasps, thus preventing gene flow between two species (Ledford 2007).

Isolation through incompatibility: Self-incompatibility may not only isolate population system through restricted gene dispersal but may stimulate the development of gene flow barriers (Levin 1971). Incompatibility initiates reproductive isolation between species in several ways, e.g. pollen and stigma possess certain barriers, mainly surface proteins, that reject the union of each other by preventing pollen germination or blocking the pollen tube growth.

Isolation through autogamy: Genetic isolation of autogamous races is due to their lack of out-crossing and morphological characters. In sympatric congener species of flowering plants, autogamy could operate as a reproductive isolating barrier (Levin 1971) which restricts gene flow and initiates speciation. The paucity or unreliability of pollinators generally promotes autogamy. *Solanum lycopersicum* L. (Tomato) is its best example, which is a native of Peru and crossable in wild, but when introduced in other parts of world, it showed autogamy. The reason behind it is the pollinator, which is available in its native country but not available elsewhere in the world. Thus the reproductive behaviour of the later is entirely different to its congener. In such case, autogamy acts as a reproductive isolating mechanism. Thus a shift to autogamy not only restricts gene flow but promotes modifications in floral characters like small light coloured flowers, with less or no scent. However, autogamy persisting for a long period of time would also offer with many distinct morphological characters. Reproductive isolation due to autogamy or selfing was observed in many plant species, e.g. *Orchis*, *Linum*, *Pitcairnia* by many investigators (Dodson 1967, Ockendon 1968, Wendt et al. 2002).

POST-ZYGOTIC ISOLATING MECHANISM

Post-zygotic isolating mechanisms are those in which hybrid zygote fail or develop abnormally or cannot self-reproduce and establish a fertile generation in nature. Such reproductive isolation mechanism present in plants of a particular group initiates speciation. Post-zygotic reproductive isolation mainly evolves from genetic incompatibilities between inter-specific plant species during hybrid formation (Rundle & Whitlock 2001, Coyne & Orr 2004, Howard & Nosil 2005). These incompatibilities can result hybrid mortality, hybrid sterility and hybrid breakdown. Post-zygotic isolation is an important cause of ecological isolation because hybrids are generally not well adapted to either parental environment. In that case, hybrid mortality, hybrid sterility and hybrid breakdown act as reproductive isolating mechanism.

Hybrid mortality: It occurs in those cases where interbreeding is possible but development of zygote takes place abnormally and results into embryo abortion. It may be caused by disharmony either between the parental sets of chromosomes or between the developing embryo and the endosperm, which surrounds it (Stebbins 1950). A well known example of hybrid mortality was observed during the hybridization between *Phlox drummondii* Hook. (Annual Phlox) and its wild relatives (Levin 1967, 1975).

Hybrid sterility: In this case, hybrids remain healthy but unable to produce fertile seeds. Irregular behaviour of the chromosomes at later stage or meiosis during gamete development leads to complete sterility of F_1 hybrid (Stebbins 1950).The classic example of hybrid sterility is hybridization between *Spartina alterniflora* Loisel. (Smooth cordgrass / Saltmarsh Cordgrass - Poaceae) and *S. maritima* (Curtis) Fernals (Small Cordgrass), which had produced genetically isolated sterile hybrid (Groves & Groves 1880) still growing by vegetative means. Another example of hybrid sterility was observed in *Jatropha tanjorensis* Ellis and Saroja, a natural interspecific sterile hybrid (*J. curcas* x *J. gossypifolia*) reported from Tamil Nadu (Ellis & Saroja 1961, Prabhakaran & Sujatha 1999). During the study of hybrid species (*Jatropha*

tanjorensis), variety of abnormalities were found in the male and female flowers, e.g. pseudostamens in the pistillate flowers, callus like structures attached on ovary and stigma, anthers irregular in number and shape with empty anther lobes and presence of double, triple or multiple pistils (Sahai et al 2009). These hybrids occasionally bear inferior fruits with no seeds inside.

Hybrid breakdown: When reproductive isolating mechanism or barrier to gene flow between species is effective only after the F_2 generation, hybrid breakdown takes place. However, the first generation (F_1) of offsprings remains healthy and fertile but its offsprings (F_2) show sterility and weakness of vegetative growth. Thus hybrid breakdown is actually later generation breakdown. The F_1 hybrid between *Gossypium arboreum* L. (Tree cotton) and *G. herbaceum* L. (Levant cotton) is likewise vigourous and fertile (Webber 1935, Hutchinson 1940, Silow 1944) but F_2 progeny are rarely found as they are too weak to survive. Similarly in two cultivar groups 'Japonica' and 'Indica' of rice, F_2 individual showed symptoms of breakdown, poor growth and sterility (Okuno 1999, Kubo & Yoshimura 2002). In this case, only those F_2 individuals that have become homozygous for the recessive allele at both loci manifest the symptoms of breakdown, poor growth and sterility.

In particular, speciation is directly linked with genetic divergence between plant species or populations. The greater the genetic distance between two species or populations the lower the pollination or fertilization chances that leads to lower reproductive success of hybrid.

It may therefore be concluded that species in nature are kept genetically apart by diverse isolating mechanism that may arise from simple ecological separation, temporal isolation, pollinator preferences to hybrid sterility or hybrid breakdown. Reproductive isolation is therefore a reproductive incompatibility between populations that prevent interbreeding as a result of which the origin of species may take place.

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