The University of Reading

Investigations into the genetic and physiological basis of self-incompatibility in cocoa (*Theobroma cacao* L.).

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Abstract

The cultivation of cocoa (*Theobroma cacao* L.) occurs in almost fifty countries across the humid tropics of the world, dominating the domestic economies of some of its producing countries. The inherent heterozygosity of the species and the presence of self-incompatibility in genotypes conferring disease resistance have hampered the development of homozygous breeding lines and repress yields of self-incompatible clones. Investigations were performed into the genetic and physiological basis of the recognition and rejection reactions in the late-acting self-incompatibility mechanism. Three sites of possible physiological interaction were investigated: pollen-stigma recognition; sperm-egg delivery and fusion; and floral abscission. A series of pollination experiments to study pollen-stigma interaction suggests that recognition does not occur at the stigmatic surface. In addition, no homologues of genes implicated in *Brassica* pollen-stigma recognition were conclusively identified. Observations within the ovule using confocal laser scanning microscopy revealed a slower rate of sperm cell delivery to the egg nucleus and polar nuclei following sperm cell release in incompatibly pollinated ovules. In addition, semi-quantitative fluorescent staining suggests that DNA replication may be occurring within the egg nucleus following incompatible pollination. Recognition may occur within the embryo sac between the male and female cytoplasm or between gamete-specific gene products on the nuclear membranes, and may function through the failure of nuclear membrane coalescence or a loss of phase synchrony between gametes preventing karyogamy. Exogenous hormone applications suggest that the repression of abscission of compatibly pollinated flowers is regulated through endogenous auxin, although increased levels of auxin do not lead to fusion of incompatible gametes. Thus, it appears likely that the male gametophyte is recognised by the female sporophyte after pollen germination, causing an uncharacterised cascade that inhibits gamete delivery, synchrony and fusion, and this in turn fails to cause a secondary increase in auxin concentration to repress floral abscission.