Receptor-ligand interaction demonstrated in *Brassica* self-incompatibility

Noni (V.E.) Franklin-Tong

There are very few examples in plants where both the receptor and the ligand that interacts with it have been identified. The self-incompatibility (SI) system, which involves the recognition and rejection of ‘self’ pollen, is controlled by the *S* locus.

Molecular analysis of SI in *Brassica* identified two stigmatic components, *SLG* and *SRK*, and a pollen component, *SCR*/*SPII*, at the *S* locus. Two recent papers demonstrated that *SCR* and *SRK* interact, providing not only a major breakthrough in our understanding of the SI response, but also in our knowledge about receptor-ligand interactions in plant cells.

Sex is always considered to be an exciting and risqué topic. It is also crucially important from the purely biological point of view of producing the next generation. Even though many plants can reproduce by asexual means, sexual reproduction remains vital to the general fitness of a population. Indeed, it is thought that at least part of the great success of angiosperms (flowering plants) as a group is owing to their evolution of complex strategies to prevent self-fertilization and to ensure out-breeding. This is especially important if one considers the physical proximity between the anthers producing male pollen and the stigma (the female receptive organ) in most plants (Fig. 1).

Self-incompatibility (SI) is one of the most widespread of mechanisms used by flowering plants. SI is usually determined genetically by a single *S* locus, with multiple *S* alleles. This locus is remarkably polymorphic; as many as 41 *S* alleles have been identified in a single population [1]. When pollen lands on a stigma, if it carries the same *S* allele as the stigma, it is recognized as ‘self’, and rejected. If it is ‘non-self’, it grows normally and will probably achieve fertilization (Fig. 1). This interaction was alluded to as a cell–cell recognition system long before the molecular basis for SI had been determined.

In the past few years in particular, major advances have been made in the identification of components involved in the rejection of self pollen in a variety of SI systems, including *Brassica* (which include many crop species such as cabbages, oil-seed rape, mustard and turnips), the Solanaceae (which include tobacco, potato, tomato and the ornamental *Petunia*) and *Papaver* (the field poppy, commonly found in cornfields). Interestingly, all three of these SI systems operate using very different mechanisms [2]. However, it is only relatively recently, as a result of considerable effort, that the *S*-locus components on both the pollen and stigma side in the *Brassica* SI system have been identified. This is, so far, the only SI system where both the male and female recognition components are known. Their identification has enabled the first steps to be made to investigate the nature of this very precise interaction.

**Brassica S-locus components**

Molecular analysis of SI in *Brassica* identified two stigmatic components encoded at the *S* locus: a secreted *S*-locus glycoprotein (SLG) and an *S*-locus receptor kinase (SRK). SRK, which functions as the female determinant of SI, has an

---

**References**


---

**Fig. 1.** Cartoon of how self-incompatibility (SI) operates. This plant carries two *S* alleles: *S1* and *S2*, so it has the genotype *S1S2*. The haploid pollen will carry alleles *S1* and *S2*. If *S1* is sporophytically determined (as it is in *Brassica*), the pollen from this plant will have the phenotype *S1S1*. If *S1* is gametophytically determined (as it is in *Papaver* and *Nicotiana*) the pollen will have the phenotype *S1* or *S1-*. (a) An incompatible scenario (red). Pollen from an *S1S1* plant, if it lands on a stigma of a flower from the same plant, or on that of another plant carrying matching *S* alleles (i.e. *S1S2*), will be ‘self’ or ‘incompatible’. Incompatible pollen is inhibited at a specific stage during pollination. In *Brassica* and *Papaver*, this is very early and occurs on the stigma surface; in the Solanaceae it is late, and occurs in the style. As a consequence, no seed is set. (b) A compatible situation (blue). Pollen from plants that carry different *S* alleles (e.g. *S1S2*) that land on a stigma of a *S1S1* plant are not ‘recognized’ since their *S* alleles do not match. This pollen can therefore hydrate, germinate and grow through the stigma and style, and fertilize the ovules to make seed.

---

**Claire A. Canning**

**Robin Lovell-Badge**

Division of Developmental Genetics, MRC National Institute for Medical Research, The Ridgeway, Mill Hill, London, UK NW7 1AA. *e-mail: rlovell@nimr.mrc.ac.uk*
ScR/ScP11 can trigger phosphorylation of the stigma receptor. This is interesting, as it contrasts with other studies of receptor–ligand interactions in plants, where an active kinase domain is required for the interaction of both CLAVATA1 with CLAVATA3, and FLS2 with flagellin [10, 11].

Receptor–ligand interactions identified in plant cells

There is very little information regarding receptor–ligand interactions in plant systems. Although a large number of putative transmembrane receptor-like kinases (RLKs) have now been identified in plants [12], the great majority are so-called ‘orphans’ without an identified ligand. Similarly, although a number of ligands have been identified, their cognate receptors have yet to be found. There are just a handful of examples where RLK receptor–ligand interactions have been demonstrated clearly in plant systems. These have emerged over the past year or so, and include interactions between the BRI1 receptor kinase with brassinolide, a brassinosteroid [13]; the CLAVATA1 receptor kinase (involved in meristematic determination) and CLAVATA3 [10]; the FLEGGELIN-SENSING 2 (FLS2) receptor kinase with flagellin [11].

The ScR/ScP11–SRK data therefore are quite significant, especially because the only interaction where binding affinities had been measured previously was the interaction of the BRI1 receptor with brassinolide [13], which has a dissociation constant ($K_d$) of 7.4 nm. The stoichiometric measurements of the interaction of ScP11 with its cognate receptor suggest that there are two binding sites: a high-affinity binding site ($K_d = 1.2$ nm) and a low affinity site ($K_d = 32$ nm) [8]. The data for ScR6 interactions with ScR6 [8] give a $K_d$ of 0.04 nm, which is considerably higher. However, there are several notable differences in the way that these studies were carried out, any of which could
potentially explain the discrepancy observed. One obvious difference is the fact that different *Brassica* species and different S-alleles were studied. It is, therefore, difficult to make any definitive statements or draw any real comparisons at present, except to say that these interactions appear to be in roughly the same range as some animal receptor–ligand binding affinities.

**SCR/SP11 also interacts with SLG**

Both labs tested the ability of SCR/SP11 to interact with the other stigmatic component, SLG, as the role of this component in the SI response is rather unclear. Both papers present data that, although not definitive, clearly indicate that SCR/SP11 also interacts with SLG, but not as strongly as to SRK [8,9]. Because both labs, using different approaches, came to the same conclusion, this seems a probable scenario. Furthermore, Takayama’s data [9], using cross-linked SP11 to immunoprecipitate SLG, suggest a very close association between SRK and SLG. They propose a model whereby SRK and SLG form a high-affinity receptor complex that interacts with SCR/SP11. The fact that SLG and SCR/SP11 interact is not altogether surprising, because the ‘ectodomain’ of SRK shares significant homology with SLG; indeed, this was how SRK was identified. However, the biological significance of the interaction between SLG and SCR/SP11 is unclear, especially as SLG is not necessary for the SI response, at least in certain haplotypes [14].

**SCR/SP11 can stimulate SRK autoprophosphorylation**

Finally, it has been demonstrated [9] that SP11 can induce autophosphorylation of SRK in an S-allele-specific manner. Although it was demonstrated previously that pollen coat proteins can elicit this response [7], this is the first time that it has been established that the pollen S receptor itself (and alone) can stimulate phosphorylation of this stigmatic receptor kinase. This is, therefore, an important observation, because it provides insight into the nature of the interaction between SP11 and SRK. It is assumed that the interaction triggers a signalling cascade as a consequence of the *Brassica* SI response.

**Conclusion**

The demonstration that SCR and SRK interact not only provides a major breakthrough in our understanding of the SI response, but also in our knowledge about receptor–ligand interactions in plant cells. This lays the foundation for a more detailed understanding of receptor–ligand interactions in general. It also will allow a detailed analysis of the signal transduction cascade assumed to be triggered by the SRK–SCR interaction.

**Acknowledgements**

Work in the author’s laboratory is funded by the UK Biotechnological and Biological Science Research Council (B.B.S.R.C).

**References**


Noni (V.E.) Franklin-Tong

Wolfson Laboratory for Plant Molecular Biology, School of Biosciences, University of Birmingham, Edgbaston, Birmingham, UK B15 2TT.

e-mail: V.E.Franklin-Tong@bham.ac.uk

**QTL for timing: a natural diversity of clock genes**

NeeraJ Salathia, Kieron Edwards and Andrew J. Millar

Conventional, forward genetics has identified several molecular components of circadian clocks. Many additional loci and genetic interactions have recently been implicated in rhythmic control by a major effort in mapping quantitative trait loci (QTL) in the mouse. Reconciling the QTL with previous results both from QTL and mutagenesis will be a challenge for rhythm researchers.

Most eukaryotes and some prokaryotes have evolved biological clocks that regulate behaviour and physiology rhythmically, in a daily sequence that can anticipate the environmental cycle. The clock is termed ‘circadian’, meaning ‘about a day’, because it does not keep exactly 24-hour days. In Nature, daily light and temperature cycles reset the clock and synchronize it with the 24-hour rotation of the Earth. Exactly when the daily round starts, relative to dawn (‘phase angle’), and which rhythms occur in what sequence, are probably affected by the array of selective pressures in particular habitats.

Many induced mutations have now been studied to bring some understanding of ‘clock genes’ (genes that are required to construct the circadian clock) in the five...