Incongruity and incompatibility in intimate partner relationships

N. G. Hogenboom

Stellingen

1. Normaal functioneren van de stamper-pollen relatie berust op hetzelfde principe als dat van de waard-parasiet relatie.

2. Bij het onderzoek naar de gebruikswaarde van rassen dient de invloed van veredelingsbedrijven te worden uitgeschakeld.

3. Het overbrengen van effectieve zelfincompatibiliteit uit wilde soorten in zelfcompatibele cultuursoorten is onmogelijk.

4. De evolutie in de kruisbaarheid van zelfcompatibele populaties met andere populaties kan niet worden verklaard met een serie S-gen mutaties.

5. De stabiliteit van een resistentie wordt voornamelijk door andere factoren dan haar genetische basis bepaald.

6. Exploitatie van natuurlijke incongruentie levert interessante mogelijkheden voor gereguleerde bevruchting.

7. Zonder ingrijpende veranderingen heeft de Nederlandse tomatenteelt geen toekomst.

8. Resultaten van plantenveredelingsonderzoek door overheidsinstituten dienen vrij beschikbaar te zijn.

9. De voedingsveiligheid van nieuwe rassen dient meer aandacht van de overheid te krijgen.

10. De wilde tuin heeft een grote opvoedende waarde.

11. Het S-gen is geen supergen; aan het S-gen zijn veel eigenschappen toegeschreven welke op andere genen en andere principes berusten.

12. Het functioneren van een intieme relatie kan worden belemmerd door ten minste twee mechanismen: incompatibiliteit en incongruentie.

13. In interspecifieke stamper-pollen relaties treedt geen incompatibiliteit op.

Proefschrift van N. G. Hogenboom
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Dit proefschrift met stellingen van Nicolaas Gerardus Hogenboom, landbouwkundig ingenieur, geboren te Uithoorn op 20 december 1937, is goedgekeurd door de promotor, dr. ir. J. Sneep, hoogleraar in de leer van de plantenveredeling.

De rector magnificus van de Landbouwhogeschool
H. A. Leniger

Wageningen, 31 juli 1973

N. G. Hogenboom

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Abstract

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Non-functioning of an intensively studied relationship, the one between pistil and pollen in flowering plants, has generally been associated with incompatibility. An evolutionary study in *Lycopersicon* and related research demonstrated the existence of an independent second mechanism for non-functioning.

The nature of this second mechanism is explained by a model in which the pistil-pollen relationship is based on matching genic systems. A lack of genetic information in the pollen about some relevant process in the pistil will cause non-functioning of the pistil-pollen relationship. For this phenomenon the term incongruity is proposed. Evolution and genetics of incongruity are different from those of incompatibility. Whereas incompatibility is an exception and compatibility the rule, incongruity is the rule and congruity an exception.

Reinterpretation of earlier research is necessary. It shows that in interspecific crosses incongruity plays a major role, whereas incompatibility is secondary or absent. Evolution of self-compatible species, genetics of crossability in interspecific hybrids and later generations, complex patterns of crossability between populations and *S*-gene polymorphism can all be reinterpreted on the same basis: the principle of matching genic systems. It is shown that properties have been ascribed to the *S*-gene, which are in fact based on other genes and other principles.

The present model has general applicability for intimate partner relationships as, for example, the host-parasite relationship. Naturally occurring incongruity can be exploited in different ways.

Enige biografische gegevens

De auteur werd op 20 december 1937 te Uithoorn geboren. Hij volgde het MULO te Uithoorn en bezocht de HBS aan het Ignatiuscollege te Amsterdam. Hij begon in 1956 met de studie aan de LH te Wageningen. Van 1957 tot 1959 werd het eerste deel van de militaire dienstplicht vervuld. In 1965 werd het ingenieursdiploma behaald in de richting tuinbouwplantenteelt, met als specialisaties erfelijkheidsleer en planteziektenkunde. Sindsdien is de auteur werkzaam bij het Instituut voor Veredeling van Tuinbouwgewassen te Wageningen, momenteel als hoofd van de hoofdafdeling groentegewassen. Hij verricht veredelingsonderzoek op het gebied van resistentie tegen ziekten en plagen, incompatibiliteit en soortkruisingsproblemen en de ontwikkeling van nieuwe planttypen.

De publikaties waaruit dit proefschrift is samengesteld, zijn een resultaat van een studie van kruisingsbarrières in *Lycopersicon* en van mogelijkheden om deze te doorbreken.

Introduction

Intimate relationships between two organisms occur in different forms in the plant and animal kingdom. A thoroughly studied relationship is the one between pistil and pollen of flowering plants. Non-functioning of this sexual partner relationship has generally been associated with only one mechanism, incompatibility, an outbreeding system widespread in plants and based on *S*-genes. The *S*-gene is supposed to play a role not only in intra- but also in interpopulational matings (Townsend, 1971; Hogenboom, 1973). To maintain the hypothesis that non-functioning of interpopulational pistil-pollen relationships is governed by the incompatibility system, more and more complicated hypotheses on the nature and structure of the *S*-gene were necessary and different forms of additional control had to be suggested. Even then not all results could be explained. This gave rise to doubt about the role of the *S*-gene in interpopulational matings and about the validity of current hypotheses on the genetics of non-functioning of interpopulational pistil-pollen relationships (Martin, 1963; Grun & Aubertin, 1966).

In a recent evolutionary study on pistil-pollen relationships in the genus *Lycopersicon* Mill. the self-incompatibility in *L. peruvianum* (L.) Mill. and subsequently also the unilateral incompatibility between this species and *L. esculentum* Mill. were broken down. This new situation gave the opportunity for a genetic study within a species of the two barriers and their possible interrelation. This study and related research demonstrated the existence of a separate mechanism for the common phenomenon of non-functioning of pistil-pollen relationships in interpopulational matings. Incompatibility and this second mechanism for non-functioning were shown to be distinct and independent (Hogenboom, 1972a, b, c, d).

Two different mechanisms for non-functioning: incompatibility and incongruity

The nature of the second mechanism for non-functioning of an intimate partner relationship can be easily understood on the basis of the following genetic model. (Here the model is applied to the pistil-pollen relationship, but it can be applied to any intimate partner relationship). To achieve fertilization pollen germination, pollen tube growth in a certain direction, penetration of different pistil tissues and fusion of nuclei has to take place. This complex is based on a chain of processes in both partners of which those in the pollen must closely interact with those in the pistil (Linskens, 1968; Stanley, 1971; Rosen, 1971). The pistil functions as a complex of barriers and promoters, the pollen grain as an organism carrying all genetic information necessary to penetrate all barriers and react to all promoters. Structure and physiology of the pistil are governed by a number of genes or gene-complexes, each of which rules a barrier or a promotion process. In the pollen a number of genes or gene-complexes

govern the structure and physiology of the pollen and pollen tube. For a normal functioning of the pistil-pollen relationship, as a counterpart of each barrier and promotion process in the pistil, the potential for the corresponding penetration and reaction process must be present in the pollen and become operative at the right moment. Thus, corresponding to each barrier gene or gene-complex active in the pistil there is a penetration gene or gene-complex active in the pollen. Corresponding to each promotion gene or gene-complex active in the pistil there is a reaction gene or gene-complex active in the pollen. Each of these couples govern a part of the chain of processes and interactions necessary for a good progress of progame phase and fertilization. As promoters may be seen as negative barriers, and penetration and reaction processes are not essentially different, we can for brevity speak of pistils with a certain barrier capacity (i.e. the total of characters of the pistil relevant to fertilization) and of pollen with a corresponding penetration capacity (i.e. the total of genetic information in the pollen grain relevant to fertilization). These two capacities are the components of the intimate relationship between pistil and pollen, based on matching genic systems.

This model allows at least two mechanisms for non-functioning of an intimate partner relationship. One is incompatibility, generally governed by multiple alleles at one or two loci. The *inhibiting action* of identical incompatibility genes (Lewis, 1965; Linskens, 1968) renders the partner relationship non-functional, though the potential for functioning and coordination of both partners is complete. Another cause of non-functioning is a *lack of genetic information* in one partner about the other. If, for example, in the pollen some essential penetration gene or gene-complex corresponding to a certain barrier gene or gene-complex in the pistil is lacking, the pollen tube will not be able to penetrate a barrier and tube growth will stop at some moment between pollination and fertilization. For this phenomenon of non-functioning of a partner relationship, resulting from a lack of genetic information in one partner about the other, I propose the term incongruity. From the above it will be evident that incongruity is genetically quite different from incompatibility. Incompatibility is non-functioning as a consequence of similarity of partners for *S*-alleles. Incongruity is non-functioning as a consequence of non-matching of partners for the genetic information regulating interaction and coordination. Incongruity may include a number of different processes, based on independent genes. As incongruity is considered a by-product of evolutionary divergence, its origin too is quite different from that of incompatibility.

Reinterpretation of earlier results

Basic elements of the present model for the relationship between pistil and pollen are the following: 1) each population, isolated from others through impeded fertilization, has its own pistil-pollen relationship; 2) the barrier capacity is mainly based on dominant genes; 3) in a pistil with a certain barrier capacity only pollen with all matching penetration genes can function. Earlier research in interpopulational crosses in flowering plants yielded much information on non-functioning pistil-pollen relationships. The results of these studies have been used to test the validity of the present model. This test showed the general applicability of the model and at the same time

led to some radical changes in the interpretation of those results (Hogenboom, 1973). The most interesting points arising from the test are the following. The so-called bilateral compatibility, unilateral incompatibility and bilateral incompatibility in wide crosses generally result from matching or non-matching barrier and penetration capacities of the populations concerned. The three situations should therefore be termed bilateral congruity, unilateral incongruity and bilateral incongruity, respectively, and they occur irrespective of presence or absence of any incompatibility system.

The distinction between the behaviour in interspecific crosses of self-compatible species of recent origin and those of ancient origin has generally been explained on the basis of the $SI \rightarrow Sc \rightarrow Sc^1 \rightarrow SC$ sequence of *S*-gene mutations (Lewis & Crowe, 1958). The present model allows a reinterpretation of these steps in the evolution of self-compatible species. It is based on the partial heterozygosity of the barrier capacity in self-incompatible species and on the high degree of homozygosity of the penetration capacity (pollen grains with incomplete penetration capacity are unsuccessful). Inbreeding will result in segregation of barrier genes. Development of lower barrier capacities is therefore very likely. Thus, evolution of self-compatible populations is mostly at the same time evolution of populations with a lower barrier capacity. Consequently, gradual erosion of the penetration capacity may result from the lack of positive selection pressure for certain penetration genes. Two of the steps have been brought about artificially in *Lycopersicon peruvianum* (L.) Mill. (Hogenboom, 1972a, b, c, d).

Numerous results of genetical studies on hybrids and later generations from crosses between populations conform to the present model and so can be reinterpreted. They show that in crosses between populations two mechanisms for non-functioning may occur: incongruity as the most important and, if present, incompatibility as the secondary mechanism. *S*-gene polymorphism in interpopulational crosses (Pandey, 1969a) is reinterpreted by supposing linkage between certain *S*-alleles and certain barrier genes for which the corresponding penetration genes in the pollen parent are lacking.

Complex patterns of crossability, revealing stepwise unilateral relations between populations, as found in some genera, require complex hypotheses if they are to be explained on the basis of incompatibility genes (Pandey, 1968, 1969b). With the present model, however, these patterns can easily be interpreted. They result from crosses between populations of which each has its own pistil-pollen relationship and so its own barrier and penetration capacities. The present model reveals that the *S*-gene is not a supergene. Different properties ascribed to it, which do not agree with the *S*-gene model (Lewis, 1965; Ascher, 1966; Linskens, 1968), are in fact based on other genes and other mechanisms.

Very interesting is the finding that sexual partner relationships and host-parasite relationships are not essentially different in their genetic basis. This was inferred from a comparison of the nature and evolution of host-parasite relationships with that of sexual partner relationships. A remarkable similarity was found between crossability patterns between populations of plants and the scheme for the gene-for-gene relationship between populations of host and parasite (Flor, 1956; Person, 1959). From a genetic point of view the latter is the most simple form of an intimate partner relationship: one gene in one partner corresponds to one gene in the other through

matching genic systems. The present model suggests that genes-for-gene, gene-for-genes and genes-for-genes relationships will also be found. According to this interpretation resistance and susceptibility are a consequence of incongruity and congruity, respectively. An interesting scientific cross-fertilization may be expected from communication between the field of research on sexual partner relationships and that on host-parasite relationships.

Avoidance and exploitation of naturally occurring incongruity

Interesting examples are known of lowering the barrier capacity (Gardella, 1950) and of making up a shortage in penetration capacity (Knox et al., 1972). Better understanding of the nature and occurrence of incongruity and clear distinction between incompatibility and incongruity may lead to a better use of these systems and to a more efficient search for new possibilities to avoid or to obtain non-functioning of pistil-pollen or host-parasite relationships.

Exploitation of naturally occurring incongruity opens up interesting prospects. One might use it as a new means of preventing self-fertilization, which is interesting to plant breeders for hybrid seed production. An extra barrier gene may be transferred from a related population to the female line, the corresponding penetration gene(s) to the male. Another possibility is to use incongruity for eradication of species. In host-parasite relationships one may transfer a barrier to the host which cannot be overcome by the parasite. In unisexuals, incongruent gametes may be brought together in cases when incongruity is expressed just before or during fertilization. Experiments in the field of insect control (Laven, 1967) prove that this interesting technique is feasible.

Conclusions

At least two independent mechanisms for non-functioning of intimate partner relationships exist: incongruity and incompatibility. In interpopulational relationships incongruity plays a major or even the only role. While in general incompatibility between partners is an exception and compatibility the rule, incongruity is the rule and congruity an exception. The *S*-gene is not a supergene; many properties ascribed to it are in fact governed by other genes. Sexual partner relationships are not essentially different from host-parasite relationships as regards their genetic basis. Exploitation of incongruity can be of interest in plant breeding and in disease or pest control.

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Inleiding en probleemstelling (Euphytica 21 (1972): 221-227)

De exploitatie van wilde soorten voor de veredeling van groentegewassen is gebrekkig. Ze wordt belemmerd door vele kruisingsbarrières en door een gebrek aan kennis over deze verschijnselen. Dit geldt ook voor het geslacht *Lycopersicon*. De behoefte aan een betere exploitatie van wilde *Lycopersicon*-soorten voor de veredeling van tomaat (*Lycopersicon esculentum*) leidde tot een onderzoek naar kruisingsbarrières in dit geslacht en naar mogelijkheden om deze te doorbreken. Een dergelijk onderzoek kan enerzijds leiden tot resultaten welke direct toepasbaar zijn bij de exploitatie van wilde *Lycopersicon*-soorten en anderzijds tot een beter inzicht in de achtergronden van en de relaties tussen kruisingsbarrières. Vooral de oplossing van de vraag welke relatie bestaat tussen zelfincompatibiliteit en interspecifieke incompatibiliteit is van algemeen belang.

Lycopersicon, een klein geslacht met nauwverwante soorten, vertoont verschillende kruisingsbarrières en is zeer geschikt voor een studie van deze verschijnselen. In de publikatie wordt een overzicht gegeven van het geslacht en van de daarin voorkomende kruisingsbarrières. Het meest interessant is *L. peruvianum*, tevens de soort met de grootste waarde voor de veredeling van tomaat. *L. peruvianum* is strikt zelfincompatibel. De kruising *L. peruvianum* × *L. esculentum* mislukt door belemmering van pollenbuisgroei, de reciproke kruising door embryoabortie.

In dit onderzoek is een antwoord gezocht op de volgende vragen:

1. Is het mogelijk de kruisingsbarrières binnen *L. peruvianum* en die tussen deze soort en *L. esculentum* te doorbreken?
2. Zo ja, waarop berust deze doorbreking?
3. Welke relatie bestaat er tussen zelfincompatibiliteit en interspecifieke incompatibiliteit?

Het onderzoek werd verricht aan twee verschillende herkomsten van *L. peruvianum*.

Het doorbreken van de zelfincompatibiliteit in L. peruvianum (Euphytica 21 (1972): 228-243)

De zelfincompatibiliteit van *L. peruvianum*, homomorph gametofytisch en gebaseerd op multipele allelen op één locus, neemt in het kader van pogingen om kruisingsbarrières te doorbreken een sleutelpositie in. Doorbreking van zelfincompatibiliteit vergemakkelijkt de evaluatie en exploitatie van genetisch materiaal van de wilde soort en maakt het vooral mogelijk de genetische variatie van deze kruisbevruchter snel en gemakkelijk door zelfbevruchting bloot te leggen. In het ingeteelde materiaal kunnen zich nieuwe eigenschappen voordoen, ook in relatie met andere kruisings-

barrières. Daarom werd begonnen met een poging de zelfincompatibiliteit van *L. peruvianum* te doorbreken.

Door zelfbestuiving op grote schaal van klonen met pollen dat in verschillende ontwikkelingsstadia mutageen behandeld was, werden onder meer pollenkorrels met niet-actieve *S*-allelen geselecteerd. Uit meer dan 22000 zelfbestuivingen op vijf klonen werden 1527 zaden verkregen. De zaadzetting was zeer onregelmatig, waarschijnlijk als gevolg van het ontstaan van nieuwe *S*-allelen of door genotype-milieu-interacties. Uit de zaden werden 1036 planten verkregen welke op zelfcompatibiliteit konden worden getoetst.

Van 823 planten, nakomelingen van vier klonen, waren zeven min of meer zelfcompatibel. Deze zelfcompatibiliteit is waarschijnlijk spontaan ontstaan en niet als gevolg van de mutagene behandeling. In de nakomelingschap van de vijfde kloon werden planten gevonden die zelfcompatibel waren bij ongeveer 40°C en zelfincompatibel bij een lagere temperatuur. Waarschijnlijk berust deze gevoeligheid van de incompatibiliteitsreactie voor hoge temperatuur op een recessief gen, dat in dit materiaal uitsplitste.

Ook in andere inteeltlijnen werd naar zelfcompatibiliteit gezocht. Er werden planten gevonden met een stabiele vorm van zelfcompatibiliteit en ook planten met een voor hoge temperatuur gevoelige incompatibiliteitsreactie.

Het onderzoek werd beïnvloed door inteelteffecten en toonde het belang van microscopische waarnemingen omtrent pollenkieming en pollenbuisgroei. Het verschijnen van zeer uiteenlopende eigenschappen in ingeteeld materiaal bewees de grote waarde van *L. peruvianum* als bron van variatie.

De erfelijkheid van zelfcompatibiliteit in L. peruvianum (Euphytica 21 (1972): 244-256)

Zelfcompatibiliteit in soorten met een één-locus gametofytisch incompatibiliteitssysteem kan op heel uiteenlopende genetische grondslagen berusten. Hiervan wordt in de publikatie een overzicht gegeven. De erfelijkheid van de zelfcompatibiliteit welke gevonden werd in *L. peruvianum*, werd bestudeerd enerzijds om deze eigenschap efficiënt te kunnen gebruiken bij onderzoek binnen de wilde soort, anderzijds vanwege het belang van deze kennis bij een studie van de relatie tussen zelfincompatibiliteit en de interspecifieke incompatibiliteit.

Uit de resultaten van verschillende series toetskruisingen en zelfbestuivingen en uit cytologisch onderzoek kon worden geconcludeerd dat zelfcompatibiliteit in *L. peruvianum* – naast die welke alleen optreedt bij hoge temperatuur en waarschijnlijk gebaseerd is op één recessief gen – kan berusten op verschillende typen *S*-allelmutaties (één of meer van de cistronen), op additie van een chromosoomfragment met een *S*-allel en interactie tussen verschillende *S*-allelen in het pollen en tenslotte op genen welke de expressiviteit van *S*-allelen modificeren.

Er werden aanwijzingen gevonden, dat het ontstaan van nieuwe *S*-allelen in ingeteeld materiaal van *L. peruvianum* een veel voorkomend verschijnsel is en dat pollen met een gemuteerd *S*-allel of een extra chromosoomfragment minder vitaal is dan normaal pollen.

Bij het onderzoek werd met succes gebruik gemaakt van een nieuwe korte notatie voor incompatibiliteitsgenotypen, gebaseerd op het drie-cistronen-model voor *S*-allelen.

Het doorbreken van de unilaterale incompatibiliteit tussen L. peruvianum en L. esculentum (Euphytica 21 (1972): 397-404)

Belemmering van de groei van soortvreemde pollenbuizen is een zeer veel voorkomend verschijnsel. In een aantal gevallen, vooral tussen relatief nauw verwante soorten zoals *L. esculentum* en *L. peruvianum*, treedt de belemmering slechts eenzijdig op. Men spreekt dan van unilaterale incompatibiliteit. Er wordt in deze publikatie een overzicht gegeven van het voorkomen van dit verschijnsel.

Nadat in *L. peruvianum* zelfcompatibiliteit was verkregen werd getracht de unilaterale incompatibiliteit met *L. esculentum* te doorbreken. Deze doorbreking is enerzijds van belang omdat ze – samen met de doorbreking van de zelfincompatibiliteit in *L. peruvianum* – de mogelijkheid opent de relatie tussen zelfincompatibiliteit en de interspecifieke incompatibiliteit nader te bestuderen, anderzijds omdat ze de soortkruisingsmogelijkheden verruimt en een mogelijkheid geeft om de embryoabortie te omzeilen en eventuele nieuwe plasma-genoom interacties te realiseren.

Daarom werd *L. peruvianum* materiaal met gebruik van de zelfcompatibiliteit ingeteeld, werd met behulp van UV-microscopie een studie gemaakt van de groei van *L. esculentum* pollenbuizen in de *L. peruvianum* stijl, en werd in ingeteeld *L. peruvianum* materiaal gezocht naar planten waarin het *L. esculentum* pollen normaal doorgroeide tot in de ovula.

Uit dit onderzoek bleek dat de belemmering van *L. esculentum* pollenbuizen in de *L. peruvianum* stijl verschilt van die welke het gevolg is van zelfincompatibiliteit in *L. peruvianum* en dat ze is opgebouwd uit een complex van processen. In het ovarium werd nog minstens één barrière gevonden.

Enkele planten konden worden verkregen welke nauwelijks enige belemmering vertoonden voor *L. esculentum* pollen. In het algemeen was de mate van embryoabortie, welke in zulke planten bij de kruising *L. peruvianum* × *L. esculentum* optrad, veel lager dan in de reciproke kruising. Het bleek dus mogelijk door selectie en kruising in het ingeteelde materiaal planten te ontwikkelen waarop *L. peruvianum* × *L. esculentum*-hybriden, die een geheel nieuw idiotype geven, kunnen worden geproduceerd.

Het is waarschijnlijk de eerste keer, dat interspecifieke incompatibiliteit door inteelt en kunstmatige selectie werd doorbroken.

De erfelijkheid van de unilaterale incompatibiliteit tussen L. peruvianum en L. esculentum en de genetica van de doorbreking (Euphytica 21 (1972): 405-414)

Kennis van de erfelijkheid van de interspecifieke incompatibiliteit en van de genetica van de doorbreking van deze barrière is van belang enerzijds om een eventuele relatie tussen zelfincompatibiliteit en deze barrière vast te stellen, anderzijds om deze eigenschap en de doorbreking in de plantenveredeling efficiënt te kunnen toepassen.

Over de genetische achtergronden van de barrière voor soortvreemd pollen bestaan verschillende hypothesen. Vrij algemeen wordt aangenomen dat deze barrière is gebaseerd op, of minstens nauw geassocieerd met, het incompatibiliteitssysteem. Een literatuurstudie wees uit dat, naarmate het onderzoek op dit gebied vorderde, steeds gecompliceerder hypothesen nodig waren om dit idee te handhaven. De juistheid van deze hypothesen wordt aangevochten.

Een belangrijke tekortkoming van veel vroeger onderzoek naar de erfelijkheid van de barrière voor soortvreemd pollen is dat het vrijwel steeds werd uitgevoerd aan interspecifiek hybride materiaal, met alle complicerende consequenties van dien. Door het doorbreken van de unilaterale incompatibiliteit tussen *L. esculentum* en *L. peruvianum* kon hier een betere benadering worden toegepast, namelijk door binnen de soort *L. peruvianum* het genetisch verschil vast te stellen tussen planten die de barrière voor *L. esculentum* pollen vertoonden enerzijds en planten welke deze barrière niet bezaten anderzijds.

De resultaten toonden aan dat de unilaterale incompatibiliteit tussen *L. esculentum* en *L. peruvianum* is opgebouwd uit verschillende processen, gebaseerd op onafhankelijke dominante genen. Het belemmerende karakter van de processen is een gevolg van evolutionaire divergentie, welke leidt tot incongruentie tussen partners van verschillende soorten. Het doorbreken van de incongruentie tussen *L. peruvianum* en *L. esculentum* is een gevolg van inteelt in de heterozygote soort *L. peruvianum* en uitsplitsing van planten die homozygoot recessief zijn voor de betrokken genen.

Op basis van dit reëvolutionaire onderzoek in *Lycopersicon* en van een overzicht van eerder gevonden verschillen tussen zelfincompatibiliteit en interspecifieke incompatibiliteit is de conclusie gerechtvaardigd dat deze twee verschijnselen verschillend zijn.

De relatie tussen zelfincompatibiliteit en interspecifieke incompatibiliteit: incompatibiliteit en incongruentie (Euphytica 22 (1973): 219-233)

De ontoereikendheid van de hypothesen dat het niet functioneren van een interspecifieke stamper-pollen relatie gebaseerd is op incompatibiliteit werd in de vorige publikaties aangetoond. Het bestaan van een apart mechanisme voor het niet functioneren van stamper-pollen relaties, dat een belangrijke of zelfs de enige rol speelt bij soortkruisingen, werd gedemonstreerd.

Ter vervanging van eerdere hypothesen werd een model voor de stamper-pollen relatie geformuleerd op basis van passende gensystemen in stamper en pollen. De stamper-pollen relatie omvat een keten van processen in beide partners waarvan die in het pollen nauwkeurig moeten zijn afgestemd op die in de stamper. De stamper is hierbij een complex, dat resulteert uit de activiteit van 'barrièregenen', hij heeft een bepaalde 'barrièrecapaciteit'. De pollenkorrel draagt de corresponderende 'penetratiegenen', hij heeft een bepaalde 'penetratiecapaciteit'. De stamper-pollen relatie functioneert alleen dan normaal als de genetische informatie in het pollen alle genen omvat die corresponderen met de relevante processen in de stamper.

Dit model biedt ruimte voor tenminste twee mechanismen voor het niet functioneren van de relatie. Het ene is de belemmerende actie van incompatibiliteitsgenen, welke de relatie niet-functioneel maakt hoewel het vermogen tot functioneren volledig is. Een tweede mechanisme is een gebrek aan genetische informatie in het pollen omtrent een relevant proces in de stamper. Voor deze oorzaak van niet-functioneren van de stamper-pollen relatie wordt de term incongruentie voorgesteld. Deze incongruentie is een bijprodukt van evolutionaire divergentie en ontstaat los van incompatibiliteit. De twee mechanismen zijn genetisch verschillend en onafhankelijk.

De resultaten van vele studies aan interspecifieke stamper-pollen relaties, welke

werden gebruikt voor een toets van het model, bleken in het model te passen en moesten worden gereïnterpreteerd. Deze reïnterpretatie liet zien dat de zogenoemde interspecifieke incompatibiliteit in feite vrijwel steeds een gevolg is van niet passende barrière- en penetratiecapaciteiten en dus incongruentie moet worden genoemd. Terwijl incompatibiliteit uitzondering is en compatibiliteit regel, is incongruentie regel en congruentie uitzondering. De evolutie van zelfcompatibele soorten kan niet worden verklaard met een serie van opeenvolgende *S*-gen-mutaties, maar met de evolutie van de barrière- en penetratiecapaciteiten. (Een deel hiervan werd kunstmatig gerealiseerd in *Lycopersicon*). Andere herinterpretaties betreffen resultaten van erfelijkheidsstudies met soorthybriden en latere generaties, het *S*-gen polymorphisme en het ontstaan van complexe kruisbaarheidspatronen tussen populaties. Al deze verschijnselen bleken steeds op basis van hetzelfde principe van passende gensystemen in stamper en pollen te verklaren. Het *S*-gen blijkt geen supergen. Allerlei eigenschappen welke eraan werden toegeschreven, zijn in feite op andere genen en principes gebaseerd.

Het model lijkt algemeen toepasbaar voor alle intieme partner relaties. Zo werd een treffende overeenkomst gevonden tussen de genetische achtergrond van sexuele partner relaties en die van de waard-parasiet relatie. Het verbinden van de onderzoekgebieden van deze twee relaties zal voor beide gebieden zeer stimulerend zijn.

Er worden, naast mogelijkheden om incongruentie te doorbreken, enkele praktische toepassingen van incongruentie behandeld op het gebied van de gereguleerde bevruchting – als een nieuwe methode toe te passen bij hybridezaadproduktie – en op het gebied van de bestrijding van ziekten en plagen.

Euphytica 21 (1972): 221–227

BREAKING BREEDING BARRIERS IN *LYCOPERSICON*. 1. THE GENUS *LYCOPERSICON*, ITS BREEDING BARRIERS AND THE IMPORTANCE OF BREAKING THESE BARRIERS

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SUMMARY

A brief survey is given of the genus *Lycopersicon* and of the breeding barriers within this genus. The importance of breaking these barriers is discussed in connection with the lack of knowledge of breeding barriers in general and the meagre exploitation of wild species of *Lycopersicon* in tomato breeding.

INTRODUCTION

The exploitation of wild species for vegetable breeding is meagre (RICK, 1967), it being hampered by the many breeding barriers and a lack of knowledge of these barriers. This is also true of the exploitation of wild species of *Lycopersicon* for tomato breeding (*L. esculentum* MILL.).

The need for a better exploitation of wild species of *Lycopersicon* has led to a study of incompatibility and isolating mechanisms in this genus and of the possibilities of breaking them.

THE GENUS LYCOPERSICON

Lycopersicon is a small genus in the Solanaceae and derives its economic importance mainly from one species: *L. esculentum*.

The gene-centre of *Lycopersicon* is along the west coast of South America and in the Galápagos Islands (LUCKWILL, 1943; RICK, 1961). *L. esculentum* and, to a lesser extent, *L. pimpinellifolium* MILL. have been cultivated for a long time and consequently have spread into tropical and subtropical parts of the world and recently also into more temperate regions.

Lycopersicon is closely related to the genus *Solanum*, particularly to the section Tuberarium, series Juglandifolia (LUCKWILL, 1943; RICK, 1951, 1960, 1969; WANN and JOHNSON, 1963). This relationship is so close that it is doubted whether *Lycopersicon* can be rightfully considered as a separate genus (RICK, 1960).

The confusion in the taxonomy of *Lycopersicon* was reduced by the monographs of MULLER (1940) and LUCKWILL (1943), but was not entirely removed thereby. After the studies of RICK (1953, 1956, 1961, 1963), RICK and LAMM (1955), CHMIELEWSKI and

RICK (1962) and CHMIELEWSKI (1968a) the following tentative classification can be made (within the scope of this research):
genus: *Lycopersicon* MILL.;
species in subgenus Eulycopersicon C. H. Mull.: *L. esculentum* MILL. and *L. pimpinellifolium* MILL.;
species in subgenus Eriopersicon C. H. Mull.: *L. peruvianum* (L.) MILL. (= all botanical varieties of the *L. peruvianum* complex including *L. glandulosum* C. H. MULL.), the closely related *L. chilense* DUN. (= *L. peruvianum* var. *dentatum* DUN.) and *L. hirsutum* HUMB. and BONPL.;
species which cannot be placed in one of these subgenera: *L. minutum* (CHMIELEWSKI and RICK, 1962).

The genus shows a multiplicity of forms. The most conspicuous in this respect is the *L. peruvianum* complex. It is described as extremely polymorphic, existing in a wide range of subspecific and varietal forms, showing tremendous variability and large intra-populational variability (see particularly RICK, 1963).

BREEDING BARRIERS IN LYCOPERSICON

Between the species of *Lycopersicon* – all with 12 pairs of chromosomes – there is a close relationship, as appears i.a. from cytological research (MACARTHUR and CHIASSON, 1947; GOTTSCHALK, 1954; RICK and BUTLER, 1956). Yet in many cases crosses are made difficult or impracticable by both intra- and interspecific breeding barriers (SMITH, 1944; LAMM, 1950; BOHN, 1951; DE ZERPA, 1952; RICK, 1953, 1961, 1963; MCGUIRE and RICK, 1954; RICK and LAMM, 1955; RICK and BUTLER, 1956; LEWIS and CROWE, 1958; MARTIN, 1961a, b, 1966; CHMIELEWSKI, 1962, 1966, 1968a, b). A survey of these barriers is given in Table 1.

This scheme shows that *Lycopersicon* presents widely different breeding barriers within a limited amount of material and consequently is highly suitable for a study of such barriers. Sometimes a barrier between species only occurs in part of the material, providing a good basis for genetical research.

No barriers exist between *L. pimpinellifolium* and *L. esculentum*. Although *L. minutum* and *L. hirsutum* show unilateral incompatibility with *L. esculentum*, and in the cross *L. minutum* × *L. esculentum* embryo abortion has been observed, *L. minutum* and *L. hirsutum* are not isolated from *L. esculentum*.

Table 1. Survey of intra- and interspecific breeding barriers in *Lycopersicon*.

♀ \ ♂	*L. esc.*	*L. pim.*	*L. min.*	*L. hir.*	*L. chi.*	*L. per.*
L. esc.	+	+	+	+	EA	EA
L. pim.	+	+	+	+	EA	EA
L. min.	+, UI, EA	+, UI	+	EA	EA	EA
L. hir.	+, UI	+, UI	+, UI	+, SI, UI	?	EA
L. chi.	UI	UI	UI	?	SI	EA
L. per.	UI	UI	UI	UI	EA	SI

+ = no serious barrier; SI = self-incompatibility; UI = unilateral incompatibility; EA = embryo abortion; ? = no research results known

Almost entirely isolated from *L. esculentum* are the *L. peruvianum* complex and *L. chilense*. In this group a very strict form of self-incompatibility occurs. Further, this group shows unilateral incompatibility with *L. esculentum* while in crosses in the compatible combination the embryos abort. This embryo abortion is a very serious barrier, which may in some measure be avoided with the aid of embryo culture (SMITH, 1944), but there are many instances on record where, in spite of this possibility, interspecific hybrids were hardly obtained. In view of our own experience this is probably mainly due to a lack of culturable embryos, besides being attributable to the low percentages of success. Embryo abortion is less complete when using tetraploid *L. esculentum* as the female in the species cross (BOHN, 1948; SZTEYN, unpublished), although the problem is then only shifted to the next generation and replaced by other difficulties.

Besides it being difficult to produce an F_1 between *L. peruvianum* or *L. chilense* on the one hand and *L. esculentum* on the other, the inheritance of the combination of barriers will result in a repetition of the problems in the first generations after the species cross (MCGUIRE and RICK, 1954; MARTIN, 1961b).

IMPORTANCE OF BREAKING THE BREEDING BARRIERS

The wild species of *Lycopersicon* have so far derived their importance mainly from the fact that they are a rich source of disease resistances. Of the greatest importance in this connection are the many botanical varieties of the *L. peruvianum* complex (ALEXANDER et al., 1942; DOOLITTLE, 1954; ALEXANDER and HOOVER, 1955; HOOVER et al., 1955; ALEXANDER, 1959; HOLMES, 1960; SKRDLA et al., 1968; and many others). In these, by far the most resistances were found and in many cases also the highest level of resistance.

Besides being interesting sources of disease resistances the wild species also appear to have unexpectedly great importance as sources of variation, e.g. in the form of 'novel variation' (RICK and SMITH, 1953; RICK, 1967). This may concern morphological, physiological, floral biological and other characters ('The genetics of species hybrids thus furnishes a good example of serendipity in research...', RICK, 1967). Also as a source of variation in forms and adaptability, particularly the *L. peruvianum* complex is of importance (RICK, 1963).

Comparison with the previous section shows that exactly the most promising material for tomato breeding is the most strongly isolated from *L. esculentum*. This has led to a meagre exploitation of this material. Only in a few cases and with great difficulty was a successful hybridization realized – e.g. TMV-resistance (ALEXANDER, 1963), nematode resistance (see RICK, 1967), *Cladosporium* resistance (KERR and BAILY, 1964) and corky root resistance (SZTEYN, 1962) – in which mostly only one resistance gene was transferred to tomato.

Therefore an investigation into the possibilities of breaking breeding barriers in *Lycopersicon*, besides being scientifically interesting, is of great practical importance in terms of direct application of positive results, which may lead to a better exploitation of wild species. Of scientific interest is, among other things, a better insight into interspecific differences, the action, genetics and evolution of the isolating mechanisms, into the interrelation of these two, and extending the possibilities of genetical research on *Lycopersicon*.

Breaking the self-incompatibility in the wild species would enable the application of self-fertilization, which may have important consequences. Thus there is the possibility of easily revealing the genetic variability which is typical for a cross-fertilizing species. In this new situation there is a good chance of finding new characters, among which we might also find the bilateral compatibility with *L. esculentum* and the capacity for normal endosperm and embryo development after interspecific hybridization.

Generally it will be possible to select against unfavourable recessives and for favourable recessives. The latter is of particular importance with characters that are governed by more than one gene. Homozygotising and accumulation of valuable genes becomes possible. Isolation and identification of different resistance genes and avoidance of resistance losses are greatly facilitated.

Self-fertilization also offers the possibility of rapid genetical analysis of selected characters within the wild species before hybridization with the cultivated species and thereby without the complicating interspecific interactions. This enables a more efficient use of these characters; the evaluation of the genetic potential of the wild species for breeding is facilitated and unfavourable linkages can be determined and broken or avoided.

Breaking the unilateral incompatibility between the cultivated tomato and wild species extends the possibilities of interspecific hybridization. Besides it becomes possible to determine any interaction between the plasm of the wild species and the genome of *L. esculentum* and attempts may then also be made to avoid embryo abortion after the species cross by using the wild species as the female parent. It is in this case that the physiological situation in the developing seed will very likely be different from that which exists when *L. esculentum* is used as the female parent.

Bypassing the embryo abortion barrier in this way or breaking it is the only possibility of inducing a gene flow to *L. esculentum* by crossing with more plants of the wild species and increasing the number of offspring per cross. Thus the chance of transferring favourable gene combinations (e.g. resistance genes) is increased.

Finally, if breaking the self-incompatibility and consequent inbreeding should lead to breaking the unilateral incompatibility, it will be feasible to study the possible relationship of these barriers in this new situation. This is of vital importance as this matter is not yet clear (LEWIS and CROWE, 1958; GRUN and RADLOW, 1961; MARTIN, 1963a, 1963b, 1967; GRUN and AUBERTIN, 1966; PANDEY, 1968, 1969; ABDALLA, 1970). The importance of results from such a research extends further than the genus *Lycopersicon* and possibly even further than the family Solanaceae. Particularly a better insight into the possible relationship of self-incompatibility and unilateral incompatibility is generally useful.

SCOPE OF THIS RESEARCH

In this study, started in 1966 as part of our tomato breeding work, an answer was sought to the following questions: Is it possible to break the breeding barriers in the *L. peruvianum* complex and between this and *L. esculentum*? If so, on what factors is this breaking based (phases, genetics)? What is the relationship between self-incompatibility and unilateral incompatibility?

The material used in this research is the *L. peruvianum* complex because it shows the

complete series of breeding barriers and particularly because it contains the most promising material for tomato breeding. Furthermore *L. peruvianum* is nearly ideal material for a study of breeding barriers on account of the following (LEWIS, 1948; MCGUIRE and RICK, 1954; RICK, 1963):

- the incompatibility system is known;
- the incompatibility is strict, compatible and incompatible plants are clearly distinct;
- the incompatibility reaction can easily be observed by means of research on pollen tube growth in the style;
- the stigma is large and so is the number of pollen grains per flower; large numbers of pollen tubes grow down the style;
- the number of ovules per ovary is large and so is the number of seeds per fruit (30–100);
- the cultivation is easy;
- the plant flowers profusely nearly throughout the year;
- the vegetative maintenance is easy;
- the trusses and flowers can be easily handled.

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BREAKING BREEDING BARRIERS IN *LYCOPERSICON*. 2. BREAKDOWN OF SELF-INCOMPATIBILITY IN *L. PERUVIANUM* (L.) MILL.

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SUMMARY

Attempts were made to break the self-incompatibility in *L. peruvianum* by selection of mutated *S*-alleles through large-scale self-pollination on clones with pollen which was mutagenically treated in different stages of development. Besides self-compatibility was searched for in inbred lines.

The self-incompatibility in *L. peruvianum* was found to be very strict indeed. From more than 22,000 self-pollinations on 5 clones 1527 seeds were obtained, seed set being very erratic. The possible causes of this seed set are discussed. From 1527 seeds 1036 plants were raised and tested for self-compatibility.

In the progenies of 4 clones from 823 plants tested, 7 were more or less self-compatible. It is concluded that this self-compatibility was spontaneous and not the result of the mutagenic treatment.

In the progeny of the fifth clone plants were found which reacted as self-compatible at a temperature of about 40 °C and as self-incompatible at lower temperatures. It is suggested that this character – a high temperature sensitive incompatibility reaction – is governed by one recessive gene.

In inbred lines plants were found with a stable form of self-compatibility and also plants with a high temperature sensitive incompatibility reaction.

The problems of an incompatibility research on inbred material are discussed. A brief survey is given of some of the characters found in inbred *L. peruvianum*.

INTRODUCTION

There are various incompatibility systems (LEWIS, 1954; ARASU, 1968). In *L. peruvianum*, as in the rest of the subgenus Eriopersicon, self-incompatibility is based on a homomorphic gametophytic system controlled by multiple alleles on 1 locus, according to the scheme of PRELL (1921) and EAST and MANGELSDORF (1925) (LAMM, 1950, 1953; MCGUIRE and RICK, 1954; MARTIN, 1961, 1963; GÜNTHER et al., 1968). The self-incompatibility in *L. peruvianum* is very strict and of general occurrence (LAMM, 1950; MCGUIRE and RICK, 1954; RICK and LAMM, 1955; RICK, 1963). Only GÜNTHER et al. (1968) report a slight variation in the degree of self-incompatibility within a population.

Within the scope of attempts to break breeding barriers between this wild and the cultivated tomato the self-incompatibility in *L. peruvianum* holds a key position. Once self-compatibility has been obtained it will be possible to reveal the genetic variation simply by selfing. In the inbred material new characters may develop, including those which relate to other breeding barriers.

In some cases self-compatibility can be obtained temporarily by certain treatments of the mother plant preventing or disturbing the self-incompatibility reaction (for a survey of the many possibilities see LINSKENS and KROH, 1967). In our case, however, it is more attractive to break self-incompatibility by mutation of incompatibility genes, as has been previously applied in *Oenothera* (LEWIS, 1949, 1951), *Prunus* (LEWIS and CROWE, 1954), *Trifolium* (PANDEY, 1956), *Petunia* (BREWBAKER and SHAPIRO, 1959; BREWBAKER and NATARAJAN, 1960) and *Nicotiana* (PANDEY, 1965, 1967).

Earlier attempts to break the self-incompatibility in *L. peruvianum* by mutations were not successful (DAVIES and WALL, 1961). In our research further attempts were made to break the self-incompatibility in *L. peruvianum* by inducing and isolating mutations of *S*-alleles by means of selfing with pollen which was mutagenically treated in different stages of development; in addition a search was made for spontaneous self-compatibility in inbred lines, obtained with great difficulty after large-scale self-pollination. For a survey of effects of radiation and inbreeding on the one-locus gametophytic incompatibility system reference is made to DE NETTANCOURT (1969) and DE NETTANCOURT et al. (1971).

MATERIAL AND METHODS

Plant material

A seed sample, described as 'an inbred selection of *L. peruvianum* No P.I. 128650-6Y-IV-1-12-22' was received from Dr L. J. Alexander, Wooster, Ohio, USA (our No 65503). The degree of inbreeding was not known but was probably slight. The material was identified as *L. peruvianum* (L.) MILL. (VAREKAMP, 1970 pers. comm.). From three plants of this material we made the *clones* 65503-4, 65503-9 and 65503-11.

Another seed sample was received from Dr K. Verkerk, Wageningen (our No 56137). This material was described as coming nearest to *L. peruvianum* ssp. *commutatum* Walp. (VAREKAMP, pers. comm.). From 3 plants of progenies obtained after self-pollination we made the *clones* 60080–3, 60080–8 and 65462–14. The interrelationship of these plants was: 56137 ⊗ = 60080; 60080 ⊗ = 61211; 61211–3 ⊗ = 65462.

The clones each of 40–50 plants were made from healthy free-flowering plants of good fertility. They were grown 30 m or further apart, in isolation from one another in various glasshouses, which were not insect-proof and in use for tomato crops.

The *inbred lines* of the Verkerk sample were obtained by large-scale selfing for a few generations, in continuation of the work of K. Szteyn. The seed yields of these self-pollinations were nearly always very slight or nil. The interrelationship between these inbred lines and the other material of this sample was: 56137 ⊗ = 60080; 60080 ⊗ = 61211; 61211–3 ⊗ = 65462; 65462–8 ⊗ = 66371; 66371– 4 ⊗ = 661571
66371– 5 ⊗ = 661572
66371– 7 ⊗ = 661573
66371–11 ⊗ = 661576

66371–11 ⊗ = 661577
66371–16 ⊗ = 661578

In 1967 for the first time some selected plants of these lines were tested for self-compatibility.

Cultivation of the material was as for a normal Dutch tomato crop under glass; in winter, spring and autumn the glasshouse was heated.

Mutagenic treatments were briefly as follows. Irradiation: acute irradiation was applied to some potted plants of each clone, at all stages of flower development, that is, inter alia all stages of meiosis, with X-ray doses of 200 R and 400 R, about 50 R/min, distance about 200 cm; acute irradiation of ripe pollen occurred with X-ray doses of 3000 R and 5000 R, about 200 R/min, distance about 50 cm; the moistness of the pollen was as after harvesting in the glasshouse in plastic containers of about 3 cm^3. All irradiations, executed at the ITAL, Wageningen, by Dr D. de Nettancourt and Dr R. Ecochard, took place at room temperature.

EMS-treatment of trusses with buds in all stages of development was done on the plant by the 'Anschnitt'-method (OEHLKERS, 1946). After a tentative investigation a treatment was chosen, on the basis of the response of the trusses, which just did not cause visible damage: 0.4% solution for 23 h at 14°C.

Pollinations

By vibration of flowers pollen was collected in a tea-spoon. For pollination the stigma was dipped in an excess of pollen.

The *clones* were self-pollinated with mutagenically treated pollen or with pollen of mutagenically treated plants. As controls self-pollinations with untreated pollen were executed. In that way one makes use of the incompatibility-sieve technique (LEWIS, 1948) as an efficient method of selecting mutants.

The flowers were not emasculated, pollinated trusses were not bagged. To prevent abscission of ovaries with a very low number of developing seeds, the trusses of the clones were sprayed with a 25 ppm NAA solution every 7–10 days (LEWIS, 1948).

The self-pollinations on the clones were carried out in the period April 25–September 14, 1966. Pollen irradiated with 3000 R and 5000 R, and kept in the containers, was used on the day of irradiation. Pollen of plants irradiated with 200 R and 400 R was harvested several times between 1 and 30 days after irradiation and used on the day of harvesting. Pollen of EMS-treated trusses was harvested between 2 and 15 days after treatment and used on the day of harvesting. The number of dates on which the pollinations were carried out was 5–7 for 3000 R and 5000 R pollen, and about 15 for the other treatments.

Self-pollinations on plants of *inbred lines* were always made on bagged trusses and exclusively with untreated pollen.

Test for self-compatibility

Per plant two or more trusses were enclosed in paper bags before flowering. When these trusses flowered the bags were removed and pollen of one of the bagged flowers was used to pollinate the other bagged flowers. Buds were removed from the trusses and the trusses were again bagged. Per plant in general 15 or more flowers were thus

selfed. Two months later the setting of fruit and the number of seeds were recorded. The degree of self-compatibility was expressed in the average number of seeds obtained per selfed flower.

Results of testing groups of plants for self-compatibility are generally given in distributions of the plants over the following self-compatibility classes (in seeds per flower): 0; > 0 and $\leqslant 5$; > 5 and $\leqslant 10$, etc., the last class being > 30.

Observations on pollen tube growth using fluorescence microscopy can greatly refine the determination of the degree of self-compatibility. Since 1968, when most of the experiments dealt with in this article had already been executed, observations of this kind had been possible. The method employed has been derived from that of LINSKENS and ESSER (1957), and as described by KHO and BAËR (1968), with one modification: after the NaOH-treatment the NaOH is exhausted and the aniline solution immediately added to the material. In this solution the material can be stored for a couple of days. The treatment was carried out two days after pollination. As a measure of the degree of self-compatibility we took the number of pollen tubes that reached the base of the style, after self-pollination on flowers bagged in bud-stage or on fully grown buds just before flowering, provided that pollen germination was normal. The number of styles on which observations were made was mostly 10 or more per plant, and at least 4.

RESULTS AND DISCUSSION

Experiments with clones

In discussing the results it should be borne in mind that this investigation was designed to break the self-incompatibility and not to study quantitative effects of mutagenic treatments on self-incompatibility. Hence the pollinations were always made when time was available for this purpose; as a consequence the dates of self-pollination were often different for the various treatments. As these experiments were not made under controlled conditions, comparisons of overall results of the different treatments are not justified.

Clone 65462–14 appeared to be self-compatible (HOGENBOOM, 1968) and was not further treated for the induction of *S*-gene mutations.

The results of self-pollinations on the other clones are summarized in Table 1. All these clones were highly self-incompatible. In two clones only a few seeds were obtained. In all cases where more than a few seeds were obtained very striking irregularities occurred in seed production, that is, seed production was concentrated on only one or a few dates and only a small part of the total number of pollinations. Table 2 gives data on these concentrations in seed production.

The results cannot be accounted for by effects of the mutagenic treatments. For the 200 R-, 400 R- and EMS-treatments the irregularities in seed production could perhaps be based on varying effects of the treatment resulting from differences in sensitivity between the developmental stages of the pollen (LEWIS, 1949). However, no relationship could be established between seed production and the number of days between the treatment of the father plant and the harvest of the pollen for the pollinations. Moreover, only after few of the treatments were seeds produced.

The results of pollinations with pollen which was mutagenically treated as ripe pol-

Table 1. Results of selfings with untreated pollen and with mutagenically treated pollen on 5 clones of *L. peruvianum* in 1966.

Clone No		Fruit and seed setting						
		no treatment	plant 200 R	plant 400 R	pollen 3000 R	pollen 5000 R	truss EMS	total
60080–3	pollinated flowers	557	978	1247	475	748	1080	5085
	fruits with seed	2	1	2	1	1	4	11
	seeds	7	1	2	1	1	4	16
60080–8	pollinated flowers	520	1335	1296	718	828	553	5250
	fruits with seed	1	44	4	0	0	0	49
	seeds	1	201	61	0	0	0	263
65503–4	pollinated flowers	528	1519	1542	490	500	969	5548
	fruits with seed	31	96	83	6	6	8	230
	seeds	209	361	385	15	20	48	1038
65503–9	pollinated flowers	202	472	408	636	647	203	2568
	fruits with seed	0	1	2	3	40	0	46
	seeds	0	1	2	3	171	0	177
65503–11	pollinated flowers	660	828	732	406	398	536	3560
	fruits with seed	0	1	2	4	1	?	8+?
	seeds	0	2	2	8	2	19	33
Total	pollinated flowers	2467	5132	5225	2725	3121	3341	22011
	fruits with seed	34	143	93	14	48	12+?	344+?
	seeds	217	566	452	27	194	71	1527

len might indicate that the suggestion of LEWIS (1949) that ripe pollen does not respond to mutagenic treatment because the 'mating reaction has been laid down before this stage' also holds for *Lycopersicon*.

As to be expected for a species with a strong incompatibility reaction the results cannot be accounted for by a self-compatibility-inducing action of NAA, such as was found by EYSTER (1941) in i.a. *Petunia* (cf. MCGUIRE and RICK, 1954; DE NETTANCOURT et al., 1971).

It is practically impossible to attribute the concentrations in seed production to contamination, the clones growing in isolation from one another and insect activity being slight. However, the trusses were not bagged and the glasshouses were not insect-proof, so some contamination might have taken place, e.g. by insects (MCGUIRE and RICK, 1954). In this connection it should be noted, however, that in most clones fruits were encountered which were unlabelled and, so, had been produced without hand pollination. The frequency of this phenomenon (28 fruits on all clones together, on average less than 5 seeds per fruit) was so low, in proportion to the about 30,000 flowers present, besides those which were used for hand pollination, that it may be concluded that practically no contamination has occurred.

It seems that – also in the light of results of the progenies of the clones, which will be discussed later – only two of the many causes of the so-called pseudo-self-compatibility (PANDEY, 1959; DENWARD, 1963; LINSKENS and KROH, 1967) remain as a possible explanation, viz: 1. an effect of extraneous environmental factors on the action of certain

S-alleles, or their products, directly or via interaction with their genetic background; 2. the generation of new *S*-alleles.

1. In Table 2 attention is paid to the temperature on and around the dates of self-pollination. The results do not show a temperature effect.

The data in Table 2 do not clearly point to a relationship between the date of pollination and the degree of seed setting. Almost every indication in this direction is discounted by negative results, particularly if the results of all clones are taken together. For the date 23 May only negative results are to be found in other clones than 65503–4 (in all 1 seed from 425 pollinations), just as for 29 August (no seeds from 83 pollinations). For 2 May nearly exclusively negative results were found outside 60080–8 (a total of 26 seeds from 623 pollinations). The experiments were not made under controlled conditions. We must conclude, therefore, that part of the results may possibly be ascribed to a slightly reduced self-incompatibility at certain dates by genotype-environment interactions.

Table 2. Data on striking concentrations in seed production after selfing of clones in 1966, and some of the negative results.

Clone, Treatment	Concentrations in seed production*				
	date of treatment	date of pollination, particulars about this date	number of pollinations	number of fruits	number of seeds
65503–4, no		18 May, previous day 29 °C**	56	17	135
		23 May, previous day 28 °C	10	3	18
		29 Aug, cool day	40	4	49
65503–4, 200 R (plant)	16 May	18 May, previous day 29 °C	54	9	32
	16 May	23 May, previous day 28 °C	182	49	257
	21 June	4 July, previous day 26 °C	50	0	0
65503–4, 400 R (plant)	16 May	18 May, previous day 29 °C	30	0	0
	16 May	23 May, previous day 28 °C	60	25	189
	21 June	4 July, previous day 26 °C	50	18	127
65503–4, EMS (truss)	18 Aug	29 Aug, cool day	65	4	39
65503–9, 3000 R (pollen)	31 May	31 May, cool day	67	0	0
65503–9, 5000 R (pollen)	31 May	31 May, cool day	65	33	156
60080–8, 200 R (plant)	25 April	2 May, warm day (27 °C)	119	12	50
	25 April	10 May, cool day	370	23	99
	25 April	16 May, next day 29 °C	146	5	39
	12 July	21 July, cool day	30	0	0
60080–8, 400 R (plant)	12 July	21 July, cool day	30	3	60
	25 April	9 May, cool day	348	1	1
60080–8, no		9 May, cool day	84	1	1
60080–8, 3000/5000 R (pollen)	17 May	17 May, warm day (29 °C)	504	0	0

* sometimes rather high maximum numbers of seeds per fruit, up to 30, occurred.
** the maximum outside temperature is given, the temperature in the glasshouse is about 10 °C higher; the periods in which the given dates of pollination fell, were generally cool, with few or no warm days.

2. Part of the results can be explained by assuming that generation of new *S*-alleles has occurred here as a result of recombination through inbreeding (DENWARD, 1963; PANDEY, 1970a), as was also found by DE NETTANCOURT and ECOCHARD (1969) and DE NETTANCOURT et al. (1971) in progenies obtained after self-pollination of their *L. peruvianum* clone 006 (emanating from IVT and our code No 60080–6). The sudden and unreproducible seed productions after selfing in their material closely agree with our results, although the numbers of seeds per pollinated flower in our material were generally smaller. Later more indications were found that indeed generation of new *S*-alleles occurs in this material (HOGENBOOM, 1972).

After mutagenic treatment of seed of an S_1S_3-population of *L. peruvianum*, HOFFMANN (1969) found a varying degree of self-compatibility in about 40 M_1-plants (out of nearly 4000). This was possibly inherent in the material or partly caused by physiological effects of the mutagenic treatment, as appears from part of the responses in M_2 (cf. PANDEY, 1968). In the M_2 varying percentages of plants occurred which set some seeds after selfing. Here, too, part of the results may perhaps be accounted for by newly generated *S*-alleles in this inbred material.

Experiments with progenies of the clones

In 1967 the plants, grown from the seeds obtained in 1966 after selfing of the clones, were tested for self-compatibility. The number of selfed flowers per plant averaged 20. In nearly all cases the test was done on 2 or more trusses per plant, usually on more than one date in the period 30 March to 18 September, 1967. The results are summarized in Table 3.

The difference between the number of plants tested in 1967 and the number of seeds in 1966 is attributable to, sometimes low, germination percentages and segregation of various types of plants not to be tested. Particularly in 65503–4 ⊗ many lethal and sub-lethal plants were found, plants that flowered little if at all or plants without pollen. This may be regarded as an inbreeding effect and indicates that the plants were produced by selfing. It also means that in interpreting the results obtained with this material account should be taken of inbreeding effects.

Apart from the progeny of 60080–8 to be discussed later, 823 plants were tested. Of these, 555 plants gave 0 seeds per selfed flower, 230 plants gave less than one seed per selfed flower, and few plants gave more than two seeds per flower. So the great majority of the plants responded as self-incompatible. In such cases experimenters sometimes speak of a result of revertible mutations of *S*-alleles (PANDEY, 1956, 1965; DAVIES and WALL, 1961), as suggested by LEWIS (1951, 1954; LEWIS and CROWE, 1953). A more likely explanation seems to be that it is a result of incomplete self-incompatibility which under certain conditions is fairly common (in our case we should also reckon with inbreeding effects on the genetic background), or of generation of new *S*-alleles (see earlier comments).

Plants clearly deviating in regard of seed setting numbered only 7. Data on these plants are given in Table 4. On 2–4 dates 3–6 trusses of each plant were tested. The seed set after selfing of some of these plants proved to be equal to that of a compatible cross.

In research of DAVIES and WALL (1961) and DE NETTANCOURT and ECOCHARD (1968) mutagenic treatment of *L. peruvianum* did not lead to stable mutations of *S*-genes. The

Table 3. Results of self-compatibility tests made in 1967 with progenies of 5 clones of *L. peruvianum*.

Clone		Progenies obtained after selfing of the clones							
No	treatment of father plant or pollen	number of plants tested	distribution of plants over self-compatibility classes (in seeds per pollinated flower)						
			0	5	10	15	20	25	30
60080–3	no	6	4	2					
	200 R (plant)	1	1						
	400 R (plant)	1	1						
	3000 R (pollen)	1	1						
	5000 R (pollen)	1	1						
	EMS (truss)	1	1						
60080–8									
	no	1	1						
	200 R (plant)*	186	98	60	13	9	0	3	1	2
	400 R (plant)	26	26						
65503–4									
	no	129	76	51	0	2			
	200 R (plant)	204	157	47					
	400 R (plant)	250	185	61	2	1	0	0	0	1
	3000 R (pollen)	8	3	5					
	5000 R (pollen)	15	11	4					
	EMS (truss)	45	35	10					
65503–9									
	3000 R (pollen)	2	1	1					
	5000 R (pollen)	133	62	70	0	0	0	1	
65503–11									
	3000 R (pollen)	5	4	1					
	5000 R (pollen)	2	0	2					
	EMS (truss)	19	12	7					

* This distribution, corrected for high temperature effect (that is, results of tests at very high temperature, about 40 °C, have been omitted): 109; 59; 9; 6; 0; 2; 0; 1, respectively.

Table 4. Survey of plants of deviating seed setting habit in the progenies of clones 65503–4 and 65503–9.

Clone No	Treatment of father plant or pollen	Number and degree of self-compatibility of seven plants	
		plant number	number of seeds per selfed flower
65503–4	no	219– 1	10.3
		217–17	13.1
	400 R (plant)	165–10	6.4
		142–3	9.8
		143–7	14.9
		144–6	32.4
65503–9	5000 R (pollen)	257–11	21.1

fact that in our research after selfing with mutagenically treated pollen relatively no more self-compatible plants arose than after selfing with untreated pollen (Table 1 and 3) suggests that here, too, an effect of the treatments is lacking and that the self-compatibility pointed out here is not due to the mutagenic treatments but may be based on a fairly rare gene combination or, more probably, on a spontaneous mutation at the *S*-locus. The unequal distribution of this through the gamete sample used here still remains a problem.

The probability that the self-compatibility found is attributable to other causes than the mutagenic treatment also holds for the self-compatible plant found after selfing of 65503–9 with pollen that had been treated with 5000 R. Hence this cannot serve as a valuable argument in the discussion on any relationship between *S*-mutability in post-meiotic haploid cells and the time of *S*-gene action (LEWIS, 1949; BREWBAKER and SHAPIRO, 1959; PANDEY, 1970b).

If the self-compatibility found here should indeed be due to a spontaneous mutation, this would have occurred in 65503–4 in about 1 to 10^6 pollen grains (on the basis of about 1000 pollen tubes per style), in 65503–9 in about 1 to 2.5×10^6 pollen grains. Apparently such mutations did not occur in 60080–3 and 65503–11. Nor did they occur in clone Lp 2/76 used by DAVIES and WALL (1961), or in clone 60080–6 used by DE NETTANCOURT and ECOCHARD (1968). The differences found in mutation frequency between the clones might point to differences in mutability of the *S*-alleles concerned or combinations of *S*-alleles, as was also found in *Oenothera* (LEWIS, 1951) and *Trifolium* (PANDEY, 1956).

It should be noted that the frequencies calculated here must be regarded as a rough approximation and consequently be used with much reserve. Important deviations may arise from variation in the number of pollen grains per stigma, in germination of the pollen, in seed setting and germination of the seeds, among other things as influenced by mutagenic treatment (BREWBAKER and EMERY, 1962; SHAPIRO, 1966).

A greatly deviating behaviour was shown by the progeny of clone 60080–8, obtained after selfing with pollen of 200 R-treated plants (see Table 3). Here 28 plants gave more than 5 seeds per self-pollinated flower, which is far more than in the other progenies. A striking feature about the results was the following. Of the plants tested, 138 were tested in the period 13–18 July 1967. The results of these plants, recorded in Table 5, show that especially on 13 and 18 July, by comparison, very many plants gave more

Table 5. Results of testing plants of 60080–8 ⊗ for self-compatibility, inter alia during the period 13–18 July 1967.

Testing date	Number of plants tested	Distribution of plants over the self-compatibility classes (in seeds per flower)							
		0	5	10	15	20	25	30	
July 13	47	21	7	5	3	6	2	0	3
July 14	42	27	12	1	2				
July 17	31	21	6	0	2	1	0	1	
July 18	18	9	2	2	3	1	0	0	1
July 20 and later	48	38	6	3	0	0	0	0	1

than 5 seeds per flower. When after a month these plants were re-tested the results were entirely different. Of the 19 plants tested on 13 July (more than 5 seeds per flower), 11 gave 0 or practically 0 seeds per flower at a later date (strongest decrease: 24 seeds per flower on 13 July, 0 at a later date), 3 plants gave only 2–3 seeds per flower and only 5 plants produced more than 5 seeds per flower at a later date too, although mostly much less than on 13 July (strongest decrease: 75 seeds per flower on 13 July, 7 seeds per flower at a later date). Similar shifts occurred in the results of 18 July.

There seems to be only one explanation for this phenomenon: high temperature prevents the incompatibility reaction in some plants. On 13 and 18 July the maximum outside temperature was 31.3 and 32.0 °C respectively, that is, the maximum temperature in the glasshouse was above 40 °C. It was in this respect only that these days clearly differed from the other days on which tests were made.

These results closely agree with those of HOFFMANN (1966, cit. GÜNTHER et al., 1968), who demonstrated by hot water treatment of the style that the substances in the styles of some genotypes of *L. peruvianum* that are responsible for the incompatibility reaction are sensitive to temperature above 40 °C, and also tally with effects of high temperatures on the self-incompatibility of some genotypes in *Trifolium pratense* (LEFFEL, 1963; KENDALL and TAYLOR, 1969), *T. hybridum* (TOWNSEND, 1966, 1968), *Oenothera* spp. (LINSKENS and KROH, 1967) and others.

In the tests made on 13 and 18 July, 39 plants were clearly self-incompatible, nearly all gave 0–1 seeds per flower; 18 plants showed more or less self-compatibility (between 5 and 25 seeds per flower) and in the later test nearly all gave 0–1 seeds per flower. These numbers point to a segregation in the progeny of 60080–8 of a recessive gene which governs the high temperature sensitivity of the *S*-allele action or its products in the style. Up to now such a situation has only been found in *Trifolium hybridum* (TOWNSEND, 1966, 1970). From the behaviour of parent plant 60080–8 and the segregation found it appears that the supposed sensitivity to high temperature is not inherent in one of the *S*-alleles. An investigation under controlled conditions is necessary to determine the background of these results with certainty (see TOWNSEND, 1966, 1968, 1970).

The footnote under Table 3 shows the distribution of the plants of 60080–8 (200 R) ⊗ over the self-compatibility classes, omitting the results of selfings on days with very high temperatures. Even after this correction a good many plants occur in the classes of more than 5 seeds per flower. It is unlikely that all these should have resulted from *S*-allele mutations, rather the correction for high-temperature effect may be incomplete. A gene for high-temperature-sensitivity may also show a certain expressivity – be it slight – at temperatures slightly below the extremely high ones (cf. TOWNSEND, 1970). In addition to other factors – inbreeding gives segregation of genes influencing the processes of fruit and seed setting, among them *S*-allele-modifiers or pseudo-compatibility genes (ATWOOD, 1942; LUNDQVIST, 1961; DENWARD, 1963; DE NETTANCOURT, 1969; NASRALLAH and WALLACE, 1968) and genes influencing *S*-allele recombinations (PANDEY, 1970a) – this may account for the established distribution of the plants over the self-compatibility classes. This, too, has to be investigated further.

The fact that of 26 plants of 65503–4 (200 R) ⊗, tested during the period 13–18 July 1967, the great majority gave less than 1 seed per flower and none gave more than 2.6 seeds per flower suggests that in this progeny no genes for temperature sensitivity se-

gregated. The other progenies were not tested enough on warm days to be able to draw safe conclusions.

Some progenies obtained after selfing of plants shown in Table 3 were tested in 1968 for self-compatibility. The results are given in Table 6. The offspring generally showed a large variation in the degree of self-compatibility. Besides the degree of self-compatibility seems to have decreased considerably, compared with the parent plant. This was also found in *Trifolium* (PANDEY, 1956), cf. also MELTON (1970).

In connection with this it should be noted that fluorescence-microscopic observations on selfed pistils of plants of some of the progenies in Table 6 revealed that in 257–11 ⊗, 9 of the 42 plants showed practically no pollen germination or a very deviating pollen tube growth which was rapidly arrested. In 53–5 ⊗, 42–4 ⊗ and 47–6 ⊗ also 20–30 % of the plants showed no pollen germination. Besides in these lines more or less serious, macroscopically observable deviations occurred, sometimes in a large number of plants. In addition to these striking results of inbreeding, no doubt many non-perceptible segregations will take place, e.g. of genes which have a negative effect on the results of pollinations or render them erratic, owing to such factors as the occurrence of a greater sensitivity to environment (see also DE NETTANCOURT et al., 1971).

It is clear that testing for self-compatibility in inbred material presents special difficulties, that disturbances resulting from inbreeding make an interpretation of the results difficult, that the results in terms of seeds per pollinated flower may give a wrong impression of the degree of self-compatibility in a line, and that incompatibility research without microscopic observations is unreliable unless the material is fully known.

Experiments with inbred lines

In the inbred lines many plants with deviating habit occurred. In 1967 a number of non-deviating plants were selected for a self-compatibility test, of which the results are

Table 6. Result of a test for self-compatibility of progenies obtained after selfing of a few plants which in 1967 produced more than 5 seeds per selfed flower.

Parent plant			Progeny						
No	origin	number of seeds per selfed flower	distribution of plants over the self-compatibility classes (in seeds per flower)						
			0	5	10	15	20	25	30
42– 4	60080–8 ⊗	41.1	4	26	4	4	2		
47– 6		11.9	13	28	3	3	1		
51– 4		8.9	15	33	2				
53– 5		22.5	18	13	4	3	3		
143–7	65503–4 ⊗	14.9	5	9	5	1	0	0	1
144–6		32.4	27	12	2	1	1		
217–17		13.1	10	3	0	0	1	0	0 1
257–11	65503–9 ⊗	21.1	4	7	3	2	3	0	5 24

Table 7. Result of a test for self-compatibility with selected plants of inbred lines (I_5) in 1967.

No of line	Number of plants tested	Distribution of plants over the self-compatibility classes (in seeds per flower)						
		0	5	10	15	20	25	30
661571	8		5	2	0	0	0	1
661572	8	3	5					
661573	6	2	1	0	2	1		
661576	3		1	0	0	0	1	0	1
661577	6	3	1	1	0	0	0	0	1
661578	6	1	2	0	1	1	0	0	1

shown in Table 7. Some of the plants were found to have a high degree of self-compatibility.

In 1968, 8 inbred lines, obtained by selfing of plants shown in Table 7, were tested for self-compatibility (1–3 dates). The results for 6 of these lines are given in Table 8. Two of the lines, 671019 and 671039, are conspicuous for their high degree of self-compatibility. The relationship between the degree of self-compatibility in the parent plant and that in the line is generally not clear. Probably inbreeding effects will have played a role (see earlier comments). Observed deviations were 'flowering with closed flower' in 671036 and 'little or no pollen' and 'no flowering' in line 671016, characters which point to segregation of genes with effects on floral biology.

The temperature cannot have had an effect in this case, all lines and their parents having been tested at a glasshouse temperature below 30 °C.

A different result was obtained with the other two inbred lines, 671038 (= 661576–5 ⊗, degree of self-compatibility in parent plant: more than 30 seeds per flower) and 671045 (= 661578–5 ⊗, degree of self-compatibility in parent plant: between 15 and 20 seeds per flower), which were tested in summer, mainly between 11 July and 6 August 1968. In both lines plants occurred which after selfing on 29, 30 or 31 July gave much more seeds than after selfing on other dates (see Table 9). The only explanation for this phenomenon seems to be a prevention or disturbance of the incompatibility reaction as a result of high temperature. The maximum outside temperature on 31 July was

Table 8. Result of a test for self-compatibility in inbred lines (I_6) in 1968.

No of line	No of parent plant	Number of seeds per selfed flower in parent	Distribution of plants over self-compatibility classes (in seeds per flower)							
			0	5	10	15	20	25	30	
671016	661571–1	8.1	14	10	3	0	1	0	3	1
671019	661571–4	39.6		12	4	7	0	2	2	22
671033	661573–7	14.6	19	22	1	1	0	0	1	
671036	661576–2	22.5	22	8	4	1	4	2	0	4
671039	661577–1	38.5		2	1	1	6	4	5	30
671042	661578–1	31.9	16	22	5	2	2	1		

Table 9. Reaction pattern of plants of lines 671038 and 671045 (I_6) after selfing on 29 and 30 July 1968 (671038) or 29, 30 and 31 July 1968 (671045) and after selfing on other dates.

No of line	Number of plants with the different reaction patterns		
	on 29, 30, 31 July SC[1] otherwise SI	on 29, 30, 31 July SI otherwise SI	on 29, 30, 31 July SC[2] otherwise SC
671038	8[3]	18[5]	11
671045	28[4]	8	5

[1] For technical reasons a line has been drawn between SI (self-incompatible) and SC (self-compatible) at 5 seeds per flower. The results would not be essentially different if this line should be situated a little higher.

[2] In this group, too, plants occurred which gave more seeds on 29, 30, and 31 July than on other dates; greatest shift from more than 30 to between 5 and 10 seeds per flower (both lines 1 plant)

[3] Slightest shift from between 15 and 20 to between 0 and 5 seeds per flower (1 plant)

[4] Slightest shift from between 5 and 10 to between 0 and 5 (3 plants)

[5] 16 plants always 0 seeds per flower, 2 plants 0 or between 0 and 5 seeds per flower

28 °C. So in the glasshouse the temperature ranged between 35 and 40 °C or was slightly higher than 40 °C. The outside temperature on the other days of the test period was considerably lower.

Besides a stable form of self-compatibility which segregates in these lines, in 671038 a recessive gene for high temperature sensitivity of the *S*-allele action or its products seems to segregate.

In line 671045 the differences between the results of the various dates were mostly less marked and the distribution of the plants over the various reaction patterns differed from that in line 671038. Apparently the results cannot be accounted for by a segregation of a recessive gene for high temperature sensitivity. However, flowering in this line was irregular and 20 % of the plants did not flower at all at a later date. Therefore it is quite possible that the 8 plants reacting as self-incompatible at all dates actually showed floral-biological disturbances and that all other plants of the line gave a self-compatible reaction after selfing at high temperature, or in other words that the line was pure for high temperature sensitivity.

That such a response to high temperature was found in the inbred lines accords with the suggestion made earlier in this paper that a similar response occurs in the progeny obtained after selfing of clone 60080–8, which is allied to these lines.

Some characters observed in inbred material of L. peruvianum

Besides segregation of self-compatible plants and plants with the suggested sensitivity to high temperature, a large number of very dissimilar deviating types were found in the inbred material. Some of these types appeared to result from segregation of 1 or 2 recessive genes; some were also found after the species cross *L. esculentum* × *L. peruvianum* (Rick and Smith, 1953). A number of them may be of interest for cultivation or breeding.

Some of the deviations are the following: different forms of male sterility (no germinative pollen), deviations in different parts of the flower, absence of flowers, flowering with closed corolla, fruits drop easily when fully grown, deviating leaf shape and

leaf colour, contorted stem, leaves all turned to one side, large number of ramifications in the truss (compound inflorescence), delayed sideshoot formation, different forms of (sub)lethality and necrosis.

Several of these characters should be studied further.

CONCLUSIONS

Self-incompatibility in *L. peruvianum* appears to have been broken. Probably in *L. peruvianum* at the *S*-locus spontaneous mutations for self-compatibility occur in low frequency. In the material involved in the present research between 0 and 1 per 10^6 gametes of a plant would carry this mutation. The breaking of the self-incompatibility could also be based on a combination of genes outside the *S*-locus that are already present, plants becoming homozygous as a result of selfing.

Also, a high temperature sensitivity of the incompatibility reaction was found in *L. peruvianum*, after inbreeding. The character that high temperature (about 40°C) prevents the incompatibility reaction seems to be governed by 1 recessive gene.

It cannot be assumed that the mutagenic treatments applied in this research are responsible for the seed yield found after selfing of the self-incompatible clones, or for breaking the self-incompatibility, or for the high temperature sensitivity of the incompatibility reaction.

With some genotypes account should be taken of the occurrence of genotype-environment interactions, which slightly weaken the self-incompatibility, and of the generation of new *S*-alleles.

Incompatibility research on inbred material is hampered by inbreeding effects. These are not only effects of segregation of *S*-allele modifiers, but particularly also effects of segregation of genes which influence fruit and seed setting generally. Except when the experimental material is very well known, an incompatibility research without observations on pollen germination and pollen tube growth is unreliable.

L. peruvianum harbours very much variability. In inbreeding, characters may be produced which are of importance for cultivation and breeding.

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BREAKING BREEDING BARRIERS IN *LYCOPERSICON*. 3. INHERITANCE OF SELF-COMPATIBILITY IN *L. PERUVIANUM* (L.) MILL.

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SUMMARY

A brief survey is given of the genetics of self-compatibility in species with a one-locus gametophytic system of incompatibility.

A study has been made of the genetics of the self-compatibility found in *L. peruvianum*.

From the results of various test crosses and selfings and of cytological research it is concluded that self-compatibility in *L. peruvianum* can be based on different types of *S*-allele mutations, on addition of an *S*-allele bearing chromosome fragment, or on genes modifying the *S*-allele expressivity.

The results further indicate that generation of new *S*-alleles is a frequently occurring phenomenon in inbred material of *L. peruvianum* and that pollen with an *S*-allele mutation or an extra chromosome fragment is less vital than normal pollen.

A short notation for incompatibility genotypes is given.

INTRODUCTION

The homomorphic gametophytic incompatibility system in *L. peruvianum* is controlled by multiple alleles (*S*-alleles) at one locus (LAMM, 1950; MCGUIRE and RICK, 1954; GÜNTHER et al., 1968). It is assumed that each *S*-allele in this system consists of 3 cistrons. There is one cistron controlling the specificity of the protein which acts in the incompatibility reaction – in pollen and style the same, but different for each *S*-allele – and there are two regulatory cistrons: one controlling the activity of the specificity cistron in the pollen, the other controlling that in the style. The latter two are the same for all *S*-alleles. The parts of the *S*-alleles are mutationally independent and functionally integrated (LEWIS, 1947a, 1949, 1951, 1958, 1960; LEWIS and CROWE, 1954; PANDEY, 1956a; LUNDQVIST, 1965).

The action of *S*-alleles is schematically represented in the gene-action models of LEWIS (1965), ASCHER (1966), PANDEY (1967a) and LINSKENS (1968). When the product of *S*-allele activity in the pollen tube is identical with one of those products in the style, a repressor is formed which inhibits further pollen tube growth.

Self-compatibility occurs when in pollen tube and style no or insufficient identical *S*-allele products (glycoproteins) are present. This may be due to the absence of an incompatibility mechanism as well as to various mutations.

Mutation of the pollen-regulatory cistron leads to loss of *S*-allele activity in the pollen, as was found in *Oenothera organensis* (LEWIS, 1951, 1961), *Prunus avium* (LEWIS and CROWE, 1954), *Trifolium pratense* and *T. repens* (PANDEY, 1956a), *Nicotiana alata* (PANDEY, 1965, 1967b, 1970a) and *N. bonariensis* (PANDEY, 1969).

Mutation of the stylar regulatory cistron leads to loss of *S*-allele activity in the style, as was established in *Prunus avium* (LEWIS and CROWE, 1954), *Trifolium pratense* (PANDEY, 1956a) and *Nicotiana alata* (PANDEY, 1967b, 1970a).

Loss of *S*-allele activity in pollen and style by mutation of both regulatory cistrons (or of the specificity cistron) was found in *Trifolium repens* (PANDEY, 1956a).

It appeared that mutations of regulatory cistrons could be stable or revertible (LEWIS, 1951; LEWIS and CROWE, 1953).

The literature contains a number of references to stable mutations of (not further analysed parts of) *S*-alleles, in various species, designated as 'self-compatibility factor *Sf*' or '*Sf*-alleles'.

Mutations of the specificity controlling cistron, which lead to a change in specificity, cause self-compatibility in the generation in which the mutation occurs, provided this is in the pollen or in the style. The phenomenon has been found in inbred material of *Trifolium pratense* (DENWARD, 1963), *Lycopersicon peruvianum* (DE NETTANCOURT and ECOCHARD, 1969; DE NETTANCOURT et al., 1971) and *Nicotiana bonariensis* (PANDEY, 1970b).

In some species and for certain *S*-allele-combinations tetraploidization leads to self-compatibility, through the supposed competitive interaction between different *S*-alleles in the pollen (LEWIS, 1943, 1947b; ATWOOD and BREWBAKER, 1950, 1953; BREWBAKER, 1953, 1954; PANDEY, 1956b, 1962, 1968; TOWNSEND, 1965).

Addition of an *S*-allele-bearing centric chromosome fragment to the normal genome leads – likewise through interaction between *S*-alleles in the pollen – to self-compatibility in *Petunia inflata* (BREWBAKER and NATARAJAN, 1960) and *Nicotiana alata* (PANDEY, 1965, 1967b, 1970a). In *N. alata* the fragment is said to have at the same time the function of complementing a mutant allele.

Besides by mutations the incompatibility reaction can also partially or entirely fail by the effect of major or minor genes outside the *S*-locus, as was established in *Trifolium* (ATWOOD, 1942; PANDEY, 1956; DENWARD, 1963; LEFFEL, 1963; TOWNSEND, 1965, 1966, 1969), *Petunia* (MATHER, 1943; MOSIG, 1960; BIANCHI and DIJKHUIZEN, 1961), *Secale cereale* (LUNDQVIST, 1968; WRICKE, 1969), *Nicotiana* (EAST, 1932) and *Abutilon* (PANDEY, 1960).

Finally the incompatibility reaction can be influenced more or less by internal and external environmental factors (see survey in LINSKENS and KROH, 1967).

In this paper an attempt will be made to determine the genetics of the self-compatibility which was found in *Lycopersicon peruvianum* (HOGENBOOM, 1968, 1972). This study is necessary to come to an efficient use of the self-compatibility in pre-breeding of *L. peruvianum*.

MATERIAL AND METHODS

This inheritance research, parts of which were executed in the years 1968–70, was made with material of both samples of *L. peruvianum* used in this study.

The origin of the parent plants used is as follows: plants 143–7, 144–6 and 160–5 were obtained after selfing of clone 65503–4 with pollen from 400 R-treated plants; plants 257–1 and 257–11 after selfing of clone 65503–9 with 5000 R-treated pollen.

The degree of self- or cross- compatibility was determined on the basis of 'the number of seeds per pollinated flower' (seed) and also, or exclusively, from observations on pollen tube growth. These observations were made on flowers that had been pollinated in the glasshouse (UV-house) and also, or exclusively, on flowers on detached trusses which, after pollination, had been placed in water at 20 °C in the phytotron for 2 days (UV–20 °). As much as possible these observations were made on 10 or more styles per plant, the minimum being 4 styles showing good pollen germination. In cases of doubt the test was repeated, if necessary several times. Trusses were bagged before flowering. Pollinations for determination of cross-compatibility were made on emasculated mature buds.

A few plants were subjected to a preliminary cytological investigation into the occurrence of extra chromosome fragments. Root tips were fixed in alcohol-acetic acid 3:1, chromosome counts were made on squash preparations after treatment with orcein and fast green (ZEILINGA, 1956).

The notation of an *S*-allele is as follows: the specificity cistron is indicated by a figure: 1, 2, etc., in order of discussion and, if mutated, by – (loss mutation) or x(mutation to other specificity); the pollen-regulatory cistron by *p* and, if mutated, by –; the stylar regulatory cistron by *s* and, if mutated, by –. Thus the incompatibility genotype of a plant is e.g. 1*ps*/2*ps*, and if mutation of a pollen-regulatory cistron has taken place e.g. 1–*s*/2*ps*.

The abbreviations SI and SC mean self-incompatible and self-compatible, respectively. The line between SI and SC is drawn, based on experience, near 5 seeds per flower or pollen tubes per stylar base. Segregations were tested with the aid of binomial probability paper. Material and methods not described here were described earlier (HOGENBOOM, 1972).

RESULTS AND DISCUSSION

On considering the results it should be borne in mind that this study is based on a very recent development arisen from inbreeding in a cross-fertilizing species. As a consequence the chance of genetically unbalanced material is great.

The self-compatibility of plant 257–11

257–11 was a vigorous plant, yielding 21 seeds per flower after selfing. The number of pollen tubes at the base of the style was 50 or more. Plants of the progeny obtained after selfing of 257–11, which did not show floral biological deviations, were all SC. The inheritance of the self-compatibility of 257–11 was studied on the F_1's obtained from crossing this plant with a strictly SI sister plant 257–1. (Strictly SI means: in the upper part of the style a bundle of up to many hundreds of pollen tubes; the bundle becomes thinner towards the base, mostly gradually; only very few, if any, tubes reach the stylar base. After selfing no seed is obtained.)

Plants 257–11 and 257–1 were bilaterally compatible. Both F_1's from crosses of 257–11 with 257–1 segregated into two clearly distinct groups of plants: SI and SC

(see Tables 1 and 2). Because of the similarity between the results of 'UV-house' and 'UV-20°', these have been taken together; subsequently, for the same reason and because of the advantage of the controlled environment, we mainly used 'UV-20°'.

Since 1969 results are more reliable than earlier results (this also applies to the F_1's which are discussed later), because since then work has been carried out under controlled conditions after pollination (UV-20°), observations have been made on more flowers per plant and in cases of doubt have been repeated several times. This appeared to be necessary because for a number of plants the reaction pattern could only be established after repeated tests as a result of sometimes insufficient pollen germination or a deviating growth pattern of the pollen tubes. In all cases the number of tubes at the stylar base has been used for the determination of compatibility, in such particular cases, however, also the number of tubes at the base in relation to the number of normal tubes in the upper part of the style. Owing to the occurrence of plants with sometimes deviating pollen germination and/or pollen tube growth, a test on 'seed', which cannot be repeated several times, will frequently result in too many plants being classed in the SI group.

The F_1 plants of 1969 (Table 2) were maintained vegetatively and tested again in 1970, this time also for their reaction after crossing with various genotypes. The results

Table 1. Distribution of plants of F_1's from crosses of 257–11 with 257–1 over the self-compatibility classes.

Pollinating date	F_1	Distribution of plants over the self-compatibility classes (in seeds per flower)								Total number of plants tested
		0	5	10	15	20	25	30		
April 22-May 9, 1968	257–11 × 257–1	16	6	1	1	0	1	1	11	37
Sept. 5-Sept. 24, 1968	257–1 × 257–11	23	5	3	0	2	2	2	10	47

Table 2. Distribution of plants of the F_1's from crosses of 257–11 with 257–1 over the self-compatibility classes.

Pollinating date	F_1	Distribution of plants over the self-compatibility classes (in pollen tubes per stylar base)								Total number of plants tested
		0	5	10	15	20	25	30		
Aug. 12, 1968	257–11 × 257–1[1]	6	10	0	2	1	0	2	8	29
Oct. 14, 1968	257–1 × 257–11	22	9	1	1	1	2	1	11	48
June 17–23, 1969	257–11 × 257–1[2]	5	15	0	0	0	2	1	13	36
June 16–17, 1969	257–1 × 257–11[3]	11	12	1	1[4]	0	1	1	11	38

[1] 4 plants showed (practically) no pollen germination; this material is the same as in Table 1

[2] 4 plants not to be classified owing to deviating pattern of pollen tube development: many very short and abnormally developed tubes in the stigma and often little pollen germination; 3 of these 4 plants were probably SC

[3] 2 plants not be classified, as in [2]; both probably SC

[4] to be classified as SC only after repeated tests; in this case the number of pollen tubes at the stylar base has to be considered in relation to the number of normal tubes in the upper part of the style

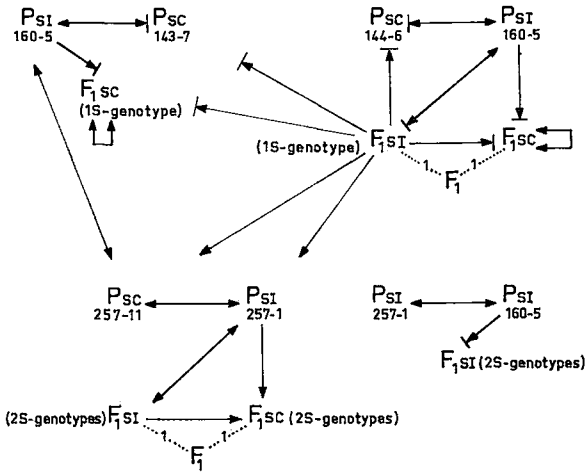

Fig. 1. Incompatibility relations between some of the SI (257–1 and 160–5) and SC (143–7, 144–6 and 257–11) plants and their F_1's. → and →| mean: the ♀ to which the arrow points is compatible and incompatible, respectively, for the pollen of the ♂ from which the arrow comes.

are shown in Fig. 1. The segregation in the F_1's into SI and SC plants proved to be in the ratio of 1:1 (21 SI, 19 SC in the F_1 of 257–11 × 257–1 and 23 SI, 16 SC in the F_1 of the reciprocal cross). The SI F_1-plants appeared to belong to two intra-incompatible, inter-compatible groups. SI plants of both genotypes were compatible as male with the SC plants.

Using the SI parent 257–1 as female, crosses were made with three randomly chosen plants of each of the incompatibility groups of each of the F_1's; in all cases the combination was compatible. When used as female the F_1-plants were compatible with the SI parent.

To ascertain how many genotypes occurred in the group SC F_1-plants full sib F_2's, obtained by crossing in each of the two F_1's one SC plant as male with each of the other SC plants, were tested in 1970. Table 3 shows the results of selfing in the sib-F_2's ('UV-20°'), of which at least 12 plants were tested.

In about half of the sib-F_2's a clear segregation into SC and SI plants occurred (ratio total 3:1, in one group a shortage of SC plants); in the remaining F_2's all plants were SC. The SI plants which segregated in the full sib F_2's were crossed as females with 257–1. All proved incompatible, except in one F_2 (No 162). In all 40 plants of this

Table 3. Results of selfing in full sib F_2's of the SC plants in the F_1's from crosses of 257–11 with 257–1.

Origin full sib F_2's	Number of F_2's			In segregating F_2's			
	tested	segregating into SC and SI	with only SC plants	total number of plants tested	total number of SC plants	total number of SI plants	P 3:1
of F_1 257–11 × 257–1	14	9	5	133	98	35	$0.70<P<0.75$
of F_1 257–1 × 257–11	15	9	6	130	84	46	$0.005<P<0.010$

F_2 were tested; 20 were SI. Of these, 17 were crossed as females with 257–1, 7 proved compatible.

In the progeny obtained after selfing of 257–11 two types of plants occurred: in about half of the plants very few pollen tubes in the style stopped growing after selfing, nearly all tubes reached the base of the style. In the other plants after selfing many tubes in the style stopped growing, particularly in the upper part of the style. (This technique of distinguishing complete compatibility from 1-allele-compatibility can only be used under optimal conditions).

All results point to the following situation in relation to the incompatibility genotypes: 257–1 is heterozygous: 1*ps*/2*ps*; 257–11 has one completely intact *S*-allele which is different from the two in 257–1. The other allele in 257–11 has mutated; from the results it is not directly clear which part has mutated. It is, however, unlikely that we should be concerned here with a mutation of only the pollen-regulatory cistron or the stylar regulatory cistron. This can be concluded from the results summarized above and also from the fact that crosses of diverse material with 257–11 as male and as female were always compatible. Self-compatibility in 257–11 only on the basis of addition of an extra *S*-allele-bearing fragment cannot be reconciled with the results. Therefore it is assumed that the incompatibility genotype of 257–11 is 1– –/3*ps* or 3– –/3*ps* or –ps/3*ps* or contains a combination of mutations (of regulatory cistron(s) and specificity cistron). HOFFMANN (1971) also mentioned the occurrence of mutations of 'pollen- and stylar part' in *L. peruvianum*, besides 'pollen part' and 'stylar part' mutations, but in his material physiological effects of the mutagenic treatments (on seeds) seem to play an important role.

Plant 257–11 was bilaterally compatible with its parent 65503–9. This also suggests mutation of both the pollen – and the stylar regulatory cistron or loss mutation of the specificity cistron. However, 257–1, too, was bilaterally compatible with 65503–9 and so contained at least one *S*-allele that dit not occur in the parent. Therefore it is not impossible that the compatibility between 257–11 and its parent was also a result of the presence of an *S*-allele in 257–11 that did not occur in the parent.

The anomalous results of the sib-F_2 No 162 after crossing with 257–1 indicate that in the Alexander-sample of *L. peruvianum* generation of a new specificity occurs, as was already established for the Verkerk-sample (DE NETTANCOURT et al., 1971). Earlier results of our research also suggested the occurrence of this phenomenon in our material (HOGENBOOM, 1972). Apparently the phenomenon is frequent in inbred material of *L. peruvianum*.

The shortage of SC plants after the crosses SI × 1-allele-SC (Tables 1–2, text) and 1-allele-SC × 1–allele-SC (Table 3) indicates that pollen with the *S*-allele-mutation is less vital than that with the intact *S*-allele (cf. ATWOOD, 1945).

With a plant of the progeny obtained after selfing of 257–11 the same series of experiments was carried out as with 257–11. The results with this plant (6–45) were identical with those of 257–11; however in none of the sib-F_2's were SI plants encountered that were compatible as females with 257–1.

The self-compatibility of plant 144–6

Plant 144–6 was highly SC: 32 seeds per selfed flower, 50 or more pollen tubes at the base of the style. The few plants of the progeny obtained after selfing of 144–6 that did

not show floral biological deviations, were SC. The compatibility between 144–6 and its parent 65503–4 could not be tested because the parent was lost when the plants were vegetatively maintained. The inheritance of the self-compatibility in 144–6 was investigated on the F_1 of the cross with plant 160–5, a strictly SI sister plant.

Crosses and observations on pollen tubes showed that 160–5 was only compatible with 144–6 when used as female. From the cross 144–6 × 160–5 in one case two plants were obtained (from more than 35 pollinations), one being SI, the other SC.

The F_1 160–5 × 144–6 proved to consist of two distinct groups, one of SI and one of SC plants (Tables 4 and 5). The F_1 plants of 1969 supplemented by others of the same F_1, were vegetatively maintained and more fully tested in 1970. The results are given in Fig. 1.

Segregation into SI and SC plants was in the ratio of 1:1, again with a shortage of SC plants: 16 SC, 20 SI. The SI plants all belonged to one intra-incompatible group; as males they were (also) incompatible with all SC F_1-plants. SC F_1-plants were compatible with each other. When used as females all F_1-plants were incompatible with the SI parent 160–5. When used as males two SI F_1 representatives were compatible with 160–5, incompatible with 144–6 and compatible with 257–11 and 257–1.

In both F_1's from crosses between 257–1 and 160–5, 20 plants were tested. All were found to be SI. In both F_1's only two incompatibility genotypes occurred. These appeared to be the same in the two F_1's and were both incompatible as female with 160–5. From this it appeared that 160–5 is an *S*-homozygote. Plant 160–5 was bilaterally compatible with 257–11.

A preliminary cytological investigation showed that plant 144–6 contained, besides the normal number of chromosomes (24), an extra chromosome fragment.

The results point to the following situation: the incompatibility genotype of plant 160–5 is 4*ps*/4*ps*; that of 144–6 is 4*ps*/5*ps* + fragment 4*ps* or 5*ps*/5*ps* + fragment 4*ps*, or, combined with a mutation of the pollen- and/or the stylar regulatory cistron, 4–*s*/5*ps* + fragment 4*ps*, 4*p*–/5*ps* + fragment 4*ps* or 4– –/5*ps* + fragment 4*ps*, the pollen with mutated pollen-regulatory cistron and without fragment being lethal (cf. PANDEY, 1965). Invariably competitive interaction occurs between different *S*-alleles in the pollen only (cf. BREWBAKER and NATARAJAN, 1960; PANDEY, 1967b). The F_1 – only that with 160–5 as female can be made – is in all cases: 4*ps*/5*ps* and 4*ps*/5*ps* + fragment 4*ps*. The shortage of SC plants in F_1 (Table 4–5, and text) indicates a smaller vitality of pollen with the extra chromosome fragment.

The self-compatibility of plant 143–7

After selfing plant 143–7 gave 15 seeds per flower; 20 or more pollen tubes reached

Table 4. Distribution of the plants of F_1 160–5 × 144–6 over the self-compatibility classes.

Pollinating date	Distribution of plants over the self-compatibility classes (in seeds per flower)							Total number of plants tested
	0	5	10	15	20	25	30	
Sept. 1–13, 1968	18	11	1	2	3	4	1	8	48
Sept. 18–Oct. 16, 1969	12	1	1	0	2	2	1	3	22

Table 5. Distribution of the plants of F_1 160–5 × 144–6 over the self-compatibility classes.

Pollinating date	Distribution of plants over the self-compatibility classes (in pollen tubes per stylar base)							Total number of plants tested
	0	5	10	15	20	25	30	
Oct. 14, 1968[1]	17	5	3	0	3	2	2	6	38
Sept. 10-Oct. 28, 1969[2]	1	7	0	0	2	0	1	6	17
July 7, 1969		3	1	0	0	0	0	4	8

[1] the same material as in Table 4, 1968; 4 plants were found which showed (practically) no pollen germination

[2] the same material as in Table 4, 1969

the base of the style. Plants without deviations in the progeny obtained after selfing of 143–7 were SC. As with 144–6, the inheritance of the self-compatibility of 143–7 was studied on the F_1 of a cross with sister plant 160–5.

With 143–7 also, 160–5 was only compatible as female (143–7 × 160–5: 0 seeds from 42 pollinations), which was confirmed by observations on the pollen tube growth. The F_1 160–5 × 143–7 consisted of SC plants only (see Tables 6 and 7).

In 1970 17 F_1 plants of 1969 were investigated more extensively. The results are shown in Fig. 1. All F_1-plants (all SC) and plant 143–7 were incompatible as females with an SI representative of F_1 160–5 × 144–6 and with the SI parent.

By crossing six F_1-plants (as female) with one sister plant 6 full sib F_2's were obtained. Of each of these F_2's 16 plants or more were tested for self-compatibility. All plants were SC, suggesting that all F_1-plants had the same *S*-alleles.

For the present the results permit more than one interpretation. The incompatibility genotype of 143–7 may be *4ps/5–s*. Another possibility is that in 143–7 an extra *S*-alle-

Table 6. Distribution of the plants of F_1 160–5 × 143–7 over the self-compatibility classes

Pollinating date	Distribution of plants over the self-compatibility classes (in seeds per flower)							Total number of plants tested
	0	5	10	15	20	25	30	
Sept. 5-20, 1968			1	1	3	5	3	18	31
Sept. 19-Oct. 28, 1969	1[1]	1[1]	0	1	3	2	0	7	15

[1] this plant was selfed at the end of October

Table 7. Distribution of the plants of F_1 160–5 × 143–7 over the self-compatibility classes.

Pollinating date	Distribution of plants over the self-compatibility classes (in pollen tubes per stylar base)							Total number of plants tested
	0	5	10	15	20	25	30	
Oct. 14, 1968	2[1]	1[1]	0	0	2	0	0	23	28
Sept. 10-Oct. 3, 1969					1	0	0	14	15

[1] 1 plant showed very little pollen germination

le-bearing chromosome fragment occurs and that the incompatibility genotype is *4ps*/*4ps* + fragment *5ps*. Cytological research will show whether such a fragment actually occurs. If so, it may bear an *S*-allele *4ps* or *5ps* and occur side by side with the mutated allele *5–s*.

In the cases discussed above mention was made of clearly distinguishable reactions: compatible or incompatible, and of clearly distinct plants: SC or SI. Other patterns were also found, e.g. in the offspring of plant 65462–14.

The self-compatibility of plant 65462–14

Plant 65462–14 of the Verkerk sample, obtained after 3 generations of selfing (see HOGENBOOM, 1972, material and methods), was moderately SC (HOGENBOOM, 1968). The investigation into the inheritance of self-compatibility in 65462–14 was made in crosses with some strictly SI plants (part of results in Table 8).

Plant 65462–14 was bilaterally compatible with these SI plants. The progeny obtained after selfing of 65462–14 was generally slightly to moderately SC and varied over a large range of degrees of self-compatibility. This variation was made still larger by the variation in pollen germination which occurred in this progeny. The distributions were continuous.

The F_1's from the crosses of 65462–14 with SI plants showed no segregation into SI and SC plants. The mean degree of self-compatibility was close to that of the SI parent. The same was found in F_1's of 65462–14 with SI plants 65503–9, 60080–8 and 65503–4, both in those made with 65462–14 as mother plant and in those made with this plant as the father.

Some F_2's obtained after selfing of F_1-plants with different degrees of seed setting

Table 8. Distribution of the plants in progenies of 65462–14 over the self-compatibility classes

Pollinating date	Progeny	Distribution of plants over the self-compatibility classes (in pollen tubes per stylar base)							Average number of pollen tubes
		0	5	10	15	20	25	30	
Aug. 5, 1968	65462–14⊗[1]	1	3	2	6	2	3	3	5	19
June 8–9, 1970	65462–14⊗[2]		4	6	6	9	4	1	3	16
June 10, 1970	65462–14 × 65503–11	3	39	2	1	1				3
June 16, 1970	(65462–14 × 65503–11)⊗[3]			1	5	3	2	3	8	26
June 18, 1970	(65462–14 × 65503–11)⊗[4]		7	6	1	3	2	1	1	11
June 18, 1970	(65462–14 × 65503–11)⊗[5]	1	21	3	4	0	1	2		6

[1] 12 plants showed (practically) no pollen germination (of a total of 37)

[2] 16 plants showed (practically) no pollen germination (of a total of 49)

[3] progeny obtained after selfing of F_1-plant giving more than 30 seeds per flower after selfing; 31 plants gave (practically) no pollen or pollen germination

[4] progeny obtained after selfing of F_1-plant giving 14 seeds per flower after selfing; 35 plants gave (practically) no pollen or pollen germination

[5] progeny obtained after selfing of F_1-plant giving 6 seeds per flower after selfing; 3 plants gave (practically) no pollen or pollen germination

after selfing (see foot notes Table 8) were tested for degree of self-compatibility. The results mentioned in Table 8 show a clear relationship between the degree of seed setting after selfing in the F_1-plant and the mean degree of self-compatibility in its progeny obtained after selfing. The F_2's varied over a larger range than the F_1.

The same tendency was observed after determination of the self-compatibility on the basis of seed setting. However the distributions were then more skewed towards 0; they were less reliable because of segregating sterility genes in this material ((practically) no pollen germination, (practically) no pollen).

Data on B_1's only relate to seed setting after selfing. They agree with the data previously mentioned: continuous distributions; crossing an F_1-plant with the SC parent gave a progeny with a higher average seed set than crossing the same plant with the SI parent (see Table 9). The differences were larger than is reflected in the means shown in this table, because the backcross to the SI parent gave, comparatively, many more B_1-plants of the class between 0 and 5, which yielded less than a mean of 1 seed per selfed flower, than the backcross to the SC parent. Moreover it was found that from the backcross with 65462–14 more plants with non-germinative pollen were obtained than from other crosses.

Two full sib F_2's, obtained by crossing F_1-plants with a very low level of self-compatibility or with no self-compatibility at all, gave practically no seed after selfing; however another sib-F_2 (of two F_1-plants which both gave 0 seeds per selfed flower) was found of which some plants were moderately, and two plants were sufficiently SC (more than 10 seeds per flower).

In connection with these results the following should be noted. In SI species there is a balance between incompatibility genes and the genetic background to the incompatibility mechanism (LEWIS, 1947b). In inbred material of such species account should be taken of (i) segregation of genes weakening the expressivity of *S*-alleles (ATWOOD, 1942; PANDEY, 1959; DENWARD, 1963) and (ii) segregation of genes weakening the effect of any *S*-allele mutation or the competitive interaction of *S*-alleles in the case of an addition of an *S*-allele-bearing fragment (cf. TOWNSEND, 1965).

Table 9. Distribution of the B_1-plants over the self-compatibility classes.

Pollinating date	B_1	Distribution of plants over the self-compatibility classes (in seeds per flower)								Mean number of seeds per flower
		0	5	10	15	20	25	30		
July 1–15, 1968	661503–2[1] × 65462–14	16	28	3	1	1	0	1		3
July 1–15, 1968	661529–25[2] × 65462–14	18	18	9	2	0	0	0	1	4
July 4–31, 1968	661535–29[3] × 65462–14	23	22	2						2
July 1–19, 1968	661503–2 × 60080–8	34	15	1						1
July 4–16, 1968	661529–25 × 65503–4	27	22							1
July 5–19, 1968	661535–29 × 65503–11	29	19							1

[1] plant 661503–2 (0.1 seeds per selfed flower) = 60080–8 × 65462–14
[2] plant 661529–25 (0 seeds per selfed flower) = 65503–4 × 65462–14
[3] plant 661535–29 (0 seeds per selfed flower) = 65503–11 × 65462–14

The results with 65462–14 indicate that we are concerned here with (i) and that self-compatibility in this plant is based on a number of genes weakening the self-incompatibility reaction. These genes are mainly recessive; 65462–14 is heterozygous for part of these genes.

Whether there is also the possibility of interaction between genes from the genetic backgrounds in the two different *L. peruvianum* samples is not clear.

In the Verkerk sample of *L. peruvianum* another form of self-compatibility was found (plant 53–5, derived from clone 60080–8 after selfing with pollen of a 200 R–treated plant), of which an inheritance study has not yet led to a solution. The results of the F_1's from crosses with a strictly SI sister plant differed greatly from season to season. No segregation into groups occurred, the distributions were always continuous. No intra-incompatible groups could be established. Also because of the origin of 53–5 it is probable that, besides *S*-allele-modifyers, a high temperature sensitivity plays a part, as was found in the progeny obtained after selfing of parent plant 60080–8 (HOGENBOOM, 1972). An investigation of such material should be carried out under controlled conditions.

CONCLUSIONS

Self-compatibility in *L. peruvianum* may have various genetical bases. Besides the self-compatibility which only occurs at high temperature and which is supposed to be governed by a recessive gene for high temperature sensitivity of the incompatibility reaction (HOGENBOOM, 1972), three other possibilities have been established.

Self-compatibility can also be based on 1. mutations of *S*-alleles, that is, of the pollen- and stylar regulatory cistron or the specificity cistron, or of the pollen-regulatory cistron only; 2. the addition of an *S*-allele-bearing chromosome fragment and competitive interaction between different *S*-alleles in the pollen; 3. genes weakening the self-incompatibility reaction.

From the fact that in the limited material chosen randomly from the highly polymorphic species *L. peruvianum* so many different possibilities are present it appears that *L. peruvianum*, besides being technically suitable for incompatibility research, also offers many possibilities from a genetical viewpoint.

The self-compatibility based on *S*-allele mutations, as well as that caused by the addition of an *S*-allele-bearing chromosome fragment, and that based on high temperature sensitivity are highly valuable in the exploitation of *L. peruvianum*.

Pollen with an *S*-allele mutation or an extra chromosome fragment is probably less vital than normal pollen.

Generation of new specificities in inbred material seems a frequent phenomenon in *L. peruvianum*.

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BREAKING BREEDING BARRIERS IN *LYCOPERSICON*. 4. BREAKDOWN OF UNILATERAL INCOMPATIBILITY BETWEEN *L. PERUVIANUM* (L.) MILL. AND *L. ESCULENTUM* MILL.

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SUMMARY

A brief survey is given of the occurrence of unilateral incompatibility between populations. It is reported how the unilateral incompatibility between *L. peruvianum* and *L. esculentum* has been broken stepwise by inbreeding with the aid of self-compatibility in *L. peruvianum* and selection in this self-compatible *L. peruvianum* material for absence of *L. esculentum* pollen tube inhibition, and how *L. peruvianum* material has been developed on which large-scale *L. peruvianum* × *L. esculentum* hybrid production is possible. It is concluded that the unilateral incompatibility between *L. esculentum* and *L. peruvianum* consists of a complex of separate processes.

INTRODUCTION

The phenomenon that the pollen of a population is inhibited to function on the plants of another population, while in the reciprocal cross no inhibition occurs, has been named unilateral incompatibility. The term does not include failures of crossing by other causes than inhibition of normal functioning of intact pollen.

The phenomenon has been demonstrated to occur between species of *Antirrhinum* (HARRISON and DARBY, 1955), *Brassica* (RÖBBELEN, 1960), *Darwinia* (BRIGGS, 1963), *Datura* (BUCHHOLZ et al., 1935), *Lathyrus* (DAVIES, 1957), *Lilium* (ASCHER and PELOQUIN, 1968), *Linum* (STRASBURGER, 1886; TAMMES 1928), *Lycopersicon* (MACARTHUR and CHIASSON, 1947; BOHN, 1951; DE ZERPA, 1952; MCGUIRE and RICK, 1954; LEWIS and CROWE, 1958; MARTIN, 1961a; CHMIELEWSKI, 1962, 1968), *Mirabilis* (STRASBURGER, 1886; JOST, 1907; THOMPSON, 1930), *Nicotiana* (ANDERSON and DE WINTON, 1931; SWAMINATHAN and MURTY, 1957; PANDEY, 1967, 1968), *Orchis* (STRASBURGER, 1886), *Petunia* (BATEMAN, 1943; MATHER, 1943), *Solanum* (PUSHKARNATH, 1953; LEWIS and CROWE, 1958; GARDÉ, 1959; DIONNE, 1961; GRUN, 1961; PANDEY, 1962) and *Trifolium* (MÜLLER, 1960). Besides the phenomenon has been found to occur between species of different genera (STRASBURGER, 1886; KARPECHENKO, 1924; BELLARTZ, 1956; LEWIS and CROWE, 1958; RICK, 1960; HARDON, 1962, 1967; SAMPSON, 1962; CHMIELEWSKI, 1968) as well as between different populations within a species (MARTIN, 1961b, 1963; HARDON, 1967).

The phenomenon occurs between populations of self-incompatible plants (STOUT, 1952; PUSHKARNATH, 1953; LEWIS and CROWE, 1958; GARDÉ, 1959; DIONNE, 1961; MARTIN, 1961b; PANDEY, 1962, 1968; SAMPSON, 1962; ASCHER and PELOQUIN, 1968), between populations of self-compatible plants (BUCHHOLZ et al., 1935; HARRISON and DARBY, 1955; SWAMINATHAN and MURTY, 1957; LEWIS and CROWE, 1958; MARTIN, 1961b; CHMIELEWSKI, 1962, 1968; PANDEY, 1962, 1968; SAMPSON, 1962; HARDON, 1967) and between populations of self-incompatible and populations of self-compatible plants, both in the cross self-incompatible × self-compatible (BATEMAN, 1943; MATHER, 1943; MACARTHUR and CHIASSON, 1947; BOHN, 1951; DE ZERPA, 1952; MCGUIRE and RICK, 1954; HARRISON and DARBY, 1955; LEWIS and CROWE, 1958; GARDÉ, 1959; MARTIN, 1961a, 1961b; PANDEY, 1962, 1968; SAMPSON, 1962; HARDON, 1967; ASCHER and PELOQUIN, 1968) and in the cross self-compatible × self-incompatible (HARDON, 1967; PANDEY, 1968, 1969).

In *Lycopersicon* unilateral incompatibility generally occurs between the species of the subgenus Eulycopersicon and those of Eriopersicon, only the cross Eulycopersicon × Eriopersicon being successful (MACARTHUR and CHIASSON, 1947; BOHN, 1951, DE ZERPA, 1952; MCGUIRE and RICK, 1954; LEWIS and CROWE, 1958; MARTIN, 1961a), although exceptions have been found in *L. hirsutum* var. *glabratum* (CHMIELEWSKI, 1966; MARTIN, 1966). *L. hirsutum* is also of special interest because of the unilateral incompatibility between its different populations (MARTIN, 1961b, 1963). *L. minutum*, a species not to be classified in one of the subgenera of *Lycopersicon*, holds an intermediate position (CHMIELEWSKI, 1962, 1968).

After self-compatibility had been obtained in *L. peruvianum* (HOGENBOOM, 1972a), attempts have been made to break the unilateral incompatibility between *L. esculentum* and *L. peruvianum* for various purposes. These concern the broadening of possibilities for species crossing, for novel variation by plasmatic interaction (RICK, 1967) using the plasmon differentiation in *Lycopersicon* (ANDERSEN, 1964) and for attempts to avoid embryo abortion, all leading to a better evaluation and exploitation of the wild species. It would also broaden possibilities for studying the relationship between self-incompatibility and unilateral incompatibility (see also HOGENBOOM, 1972b).

Within the scope of these attempts with inbred material of *L. peruvianum*, obtained with the aid of self-compatibility, a study was made of the growth of *L. esculentum* pollen tubes in the pistil of *L. peruvianum* and *L. peruvianum* plants were searched for which show uninhibited growth of *L. esculentum* pollen tubes and penetration into the ovules.

MATERIAL AND METHODS

Inbred material obtained by selfing of two seed stocks of *L. peruvianum* was used. This material, and the interrelationships of the material have been described earlier (HOGENBOOM, 1972a). The plants were grown under normal conditions in the glasshouse, unless otherwise stated. The great majority of the plants were found to be self-compatible, the reaction of part of the plants after self-pollination was unknown or insufficiently tested.

Pollen of *L. esculentum* came from cv. Moneymaker. It was collected and used on the same day.

The test for the interspecific pistil-pollen interaction was made as follows: trusses were cut from the plants of *L. peruvianum* and open flowers and immature buds were removed, leaving only the mature buds on the truss. The trusses were put in water and the mature buds emasculated and pollinated with *L. esculentum* pollen. After two days at 20 °C under natural light conditions in a phytotron glasshouse – contamination by other pollen was prevented – the styles or the complete pistils were collected and prepared for observations on pollen tube growth with a fluorescence microscope. With each *L. peruvianum* plant tested observations were made on at least four and mostly about ten pistils. In some of the earlier experiments the trusses with mature emasculated buds were left on the plant for two days after pollination instead of being placed in water at 20 °C. As the results with these two techniques did not differ significantly, they have been taken together.

Material of the Verkerk seed stock was tested mainly in the summer of 1968, the other material mainly in the summer of 1970. Material and methods not described here were described in earlier papers of this series.

RESULTS AND DISCUSSION

L. esculentum pollen tubes in the L. peruvianum stigma and style

More than 1,300 plants of different generations of inbreeding of *L. peruvianum* were tested for the interaction between their stigmas and styles and *L. esculentum* pollen. A summary of the results is given in Table 1.

The majority of the plants showed the picture which is normally found in *L. peruvianum* (class 0(I)): the *L. esculentum* pollen germinates very well, hundreds of pollen tubes grow into the stylar tissues, but stop growing after having penetrated to about one third of the style length or earlier. This stop of growth is in general very uniform

Table 1. Results of tests for the interaction between the stigma and style of *L. peruvianum* plants and the pollen of *L. esculentum*. Alexander I_3 was grown in the open, other material in the glasshouse.

Seed stock of *L. peruvianum* plants	Generation of inbreeding	Number of lines	Distribution of plants over interaction classes (in mean number of pollen tubes at the stylar base)				
			0(I)	0(II)	10	30	
Verkerk	I_1	1	1				
	I_2	1	21				
	I_3	5	149	4			
	I_4	1	17	1			
	I_5	3	42	49			
	I_6	8	220	20	2	1	1*
Alexander	I_0	1	2				
	I_1	2	10				
	I_2	4	70	11	2		
	I_3	8	234	298	160	37	13
* Progeny obtained after selfing of this plant:					1	3	36

for all tubes. The picture is thus quite different from that caused by the self-incompatibility reaction within *L. peruvianum* (cf. McGUIRE and RICK, 1954). A similar difference between intra- and interspecific crosses has been found in other genera (BATEMAN, 1943; LEWIS and CROWE, 1958; ASCHER and PELOQUIN, 1968; PANDEY, 1968).

In most of the inbred lines of *L. peruvianum* other interaction patterns were also found: the pollen tubes did not all stop growing at the same level but the bundle of tubes gradually became thinner. The general result was that no tubes reached the base of the style (class 0(II)), or very few. Only few plants were found which showed this same pattern of a thinning bundle and in which from ten to thirty pollen tubes reached the base of the style.

Plants of which the stigma and style showed little or no inhibition to *L. esculentum* pollen tube growth (little or no thinning of the bundle of tubes, more than thirty tubes and often far more reaching the stylar base) were also found, although very few in number and – probably as a consequence of the different numbers of plants tested – only in the further inbred material. Tests with progenies obtained after selfing of such plants showed that this absence of pollen tube inhibition is a heritable character (Table 1, note).

The above results indicate that in *L. peruvianum* the stylar barrier to *L. esculentum* pollen tube growth is built up of different steps or processes.

L. esculentum pollen tubes in the L. peruvianum ovary

L. peruvianum plants that showed uninhibited *L. esculentum* pollen tube growth in their styles were studied for growth of *L. esculentum* pollen tubes in their ovary. It was found that these plants mostly showed a complete or nearly complete stop of pollen tube growth just below the stylar base after the entry into the ovary, the ends of many tubes being distorted and swollen. Only few, if any, tubes distributed over the ovules, no ovules or very few were penetrated. These observations indicate that in *L. peruvianum* another barrier to growth of *L. esculentum* pollen tubes is present in the ovary. This separate ovarial barrier has also been found in interspecific crosses in *Solanum* (GRUN and AUBERTIN, 1966).

The progenies obtained after selfing of such plants with an ovarial barrier were again tested. Part of the results are summarized in Table 2. They show that in this

Table 2. Growth of *L. esculentum* pollen tubes in the ovary of *L. peruvianum* plants of a progeny obtained after selfing of plant 671019–10, which showed no inhibition in its style but a strong inhibition in the ovary. All results relate to plants with more than thirty *L. esculentum* pollen tubes at the stylar base.

Year	Number of plants tested	Distribution of plants over the interaction classes in									
		mean number of tubes spreading over the ovules					mean number of ovules penetrated				
		0	10	20	30		0	10	20	30	
1969	69	3	30	23	11	2	7	56	5	1	
1970	56	19	16	11	4	6	24	28	3	1	

material there was much variation in ovarial inhibition. Therefore the inbred material was selfed and intercrossed and progenies screened for plants not showing *L. esculentum* pollen tube inhibition in the ovary. In this way some *L. peruvianum* plants have been obtained – until now only in part of the material – showing fairly good distribution of *L. esculentum* pollen tubes over the ovules and with a rather high number of penetrated ovules. The highest number of penetrated ovules found in one ovary until now has been 75, the highest mean number per plant being 33.

The results indicate that also within the ovary the barrier for *L. esculentum* pollen tube growth and ovule penetration is built up of separate parts or processes.

As far as the author is aware, this is the first case of a direct stepwise breakdown of unilateral interspecific incompatibility by inbreeding and artificial selection. In this connection the important role of self-compatibility in a previously strictly cross-fertilizing species is clear. In literature unilateral incompatibility is generally treated as one process. The stepwise breakdown found here indicates that the phenomenon is – at least in certain cases – built up of a number of separate mechanisms.

The material developed here is self-compatible, does not inhibit growth of pollen of the 'old' self-compatible species *L. esculentum* and its pollen is not inhibited to grow on the self-incompatible plants of the species *L. peruvianum*. It might thus be suggested to represent the so far lacking stage Sc^1 in the model of LEWIS and CROWE (1958) on SI–>SC evolution. However, my explanation of what has occurred in this material is quite different and will be dealt with in further papers.

Seed set in the cross L. peruvianum × L. esculentum

Plants of *L. peruvianum* which showed no inhibition of *L. esculentum* pollen tube growth in stigma and style and little or no inhibition in the ovary, so that at least some ovules were entered, were tested for seed-set after pollination with *L. esculentum* pollen. Part of the results are summarized in Table 3.

The fruit-set indicates that, as expected, inhibition of pollen tube growth was not the cause of the low seed-set.

In some cases two types of seeds were obtained: normal fully developed seeds and

Table 3. Results of crosses *L. peruvianum* ♀ × *L. esculentum* ♂ with plants selected for absence of barriers to *L. esculentum* pollen tube growth (all plants had shown more than thirty tubes at the stylar base and various numbers of penetrated ovules in a UV test).

Plant No and parentage No	Mean number of penetrated ovules	Number of pollinations	Number of fruits	Number of normal seeds	Number of hybrid plants
200–9, 671019–10 ⊗	24	26	6	0	
205–7, 671019–10 ⊗	10	17	17	4	3
205–11, 671019–10 ⊗	11	21	19	6	3
211–21, cross in 671019–10 ⊗	10	35	29	4	3
211–32, cross in 671019–10 ⊗	18	17	16	1	1
211–38, cross in 671019–10 ⊗	5	26	19	4	2
211–50, cross in 671019–10 ⊗	18	24	13	0	
212–8, 671019–10 ⊗ × 671019–6 ⊗	10	16	13	3	3

very tiny but filled seeds. Some of these tiny seeds were able to germinate but only developed a root. In other cases only tiny or shrivelled seeds or ovules occurred. These results indicate that in these crosses embryo abortion occurred, that it occurred in different developmental stages and that it may be incomplete.

The plants differed in the degree of embryo abortion after the interspecific cross, and in most cases the degree was very much lower than in the cross *L. esculentum* ♀ × *L. peruvianum* ♂. In the latter there is no inhibition of pollen tubes at all and many tens of ovules are generally penetrated, the normal result being, however, that hardly any hybrids are obtained.

A step forward is that hybrids between *L. esculentum* and *L. peruvianum* can now be produced on a large scale without the need of artificial techniques. The fact that a new idiotype is created may have interesting consequences.

CONCLUSIONS

The unilateral incompatibility between *L. esculentum* and *L. peruvianum* has been broken. By inbreeding and selection in *L. peruvianum* a stepwise breakdown of the barriers to *L. esculentum* pollen tube growth was achieved.

As in other cases, the unilateral incompatibility between *L. peruvianum* and *L. esculentum* results in a different interaction pattern from that caused by self-incompatibility. Also the breakdown of unilateral incompatibility, occurring in a number of steps, is in this case different from that of self-incompatibility, being one step. Apparently the unilateral incompatibility between *L. peruvianum* and *L. esculentum* consists of a number of separate processes.

Large-scale *L. peruvianum* × *L. esculentum* hybrid production is now possible.

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BREAKING BREEDING BARRIERS IN *LYCOPERSICON*. 5. THE INHERITANCE OF THE UNILATERAL INCOMPATIBILITY BETWEEN *L. PERUVIANUM* (L.) MILL. AND *L. ESCULENTUM* MILL. AND THE GENETICS OF ITS BREAKDOWN

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SUMMARY

The validity of the current hypotheses on the inheritance of unilateral incompatibility is disputed and the importance of intrapopulational research on the inheritance of unilateral incompatibility is emphasized.

Such research has been carried out on the unilateral incompatibility between *L. peruvianum* and *L. esculentum* with inbred material of *L. peruvianum*. The results indicate that in this species the inhibition of *L. esculentum* pollen tube growth, built up of distinct processes, is based on independent dominant genes, and that the breakdown of the phenomenon is a consequence of segregation of recessive alleles, the expressivity of at least some of these being influenced by environmental factors.

Research demonstrating differences between self-incompatibility and interspecific pollen tube growth inhibition is reviewed and it is concluded from this and own research that the two phenomena are distinct.

INTRODUCTION

Knowledge of the inheritance of unilateral incompatibility between populations and of the genetics of its breakdown is important for determining any possible relationship between self-incompatibility and unilateral incompatibility and for an efficient use of the character and its breakdown in plant breeding.

Studies on the occurrence of unilateral incompatibility between species and on the inheritance of this phenomenon, the latter often being restricted to the 'SI × SC inhibition', have in most cases led to the conclusion that unilateral incompatibility is based on the self-incompatibility system or is at least strongly associated with it (ANDERSON and DE WINTON, 1931; MATHER, 1943; PUSHKARNATH, 1953; LEWIS, 1954, 1955; MCGUIRE and RICK, 1954; LEWIS and CROWE, 1958; GARDÉ, 1959; RICK, 1960; MARTIN, 1961, 1964, 1967, 1968; HARDON, 1962, 1967; PANDEY, 1962, 1967, 1968, 1969a, 1969b; GÜNTHER und JÜTTERSONKE, 1971).

The results of the earlier studies seemed to be explained satisfactorily by the hypothetic dual role of *S*-alleles (i.e. interacting with identical *S*-alleles and in a special way with '*Sc*-alleles'). Very frequently, especially in inheritance studies with segregating generations, the results showed a more complex basis of unilateral incom-

patibility so that in many cases some additional control, mono- or polygenic, had to be suggested. As research in various genera was extended, more and more complicated hypotheses were necessary to explain unilateral incompatibility as a function of the self-incompatibility system. Some of the hypotheses appear to be rather sophisticated or have a rather weak basis; they are insufficiently borne out by the facts collected and none has general applicability. The hypothesis of ABDALLA (1970) and ABDALLA and HERMSEN (1972) also suggests that the unilateral incompatibility is based on an incompatibility system, be it a special one in which specific '*UI* genes' interact with matching specific '*Sc*-alleles'.

It is very important to note and essential in this type of studies concerning the cessation of pollen tube growth – a phenomenon which may be due to several causes – that these studies, in so far they were more than merely inventories of crossing-relationships, have been carried out almost exclusively on interspecific hybrid material. With this line of approach, mostly followed because bilateral compatibility and unilateral incompatibility are not found within the same species, one introduces complicating interspecific differences and interactions. The consequences of this generally can not be estimated but may be important. One of these consequences might be a type of pollen tube growth inhibition that can not be distinguished from a result of *S*-allele action. These interspecific complications will in many cases have played a role and as a consequence the conclusions drawn may be unreliable.

A better approach to the elucidation of the inheritance of unilateral incompatibility is to determine within a species the genetic difference between plants inhibiting pollen tube growth of another species and plants not showing this inhibition. This line of approach was adopted in *Solanum* where GRUN and RADLOW (1961) found in some species both unilateral incompatibility with another species and absence of this character. GRUN and AUBERTIN (1966) studied within the species *S. chacoense* the genetic difference between plants showing inhibited growth of *S. verrucosum* pollen tubes and other plants of the same species not showing this inhibition. They found that inhibition of *S. verrucosum* pollen tubes in other species was based on two or more independent dominant genes. They concluded from their own research and from a study of literature that unilateral incompatibility is not a function of incompatibility genes (*S*-genes) – as was also suggested by STOUT (1952) and MARTIN (1963) – but of independent genes conditioning what they called acceptance or non-acceptance of pollen tubes of other species.

Through the breakdown of the unilateral incompatibility between *L. peruvianum* and *L. esculentum* by the development of *L. peruvianum* material not showing inhibition of *L. esculentum* pollen tube growth (HOGENBOOM, 1972a) a similar approach was made possible in *Lycopersicon*. The inheritance of the unilateral incompatibility was studied by determining the genetic difference between *L. peruvianum* plants showing inhibition of *L. esculentum* pollen tube growth and other *L. peruvianum* plants not showing such inhibition.

MATERIAL AND METHODS

Reciprocal crosses were made between *L. peruvianum* plants showing in their pistils different interaction patterns with *L. esculentum* pollen tubes (HOGENBOOM, 1972a).

Plants of F_1, F_2 and B_1 were tested for their interaction with *L. esculentum* pollen. This test and further methods as well as the material used were as has been described earlier (HOGENBOOM 1972a and earlier papers in this series), unless otherwise stated.

Results were obtained in the years 1968–1971.

RESULTS AND DISCUSSION

Segregations in inbred lines

Plants of 32 inbred lines of *L. peruvianum* were tested for interaction with *L. esculentum* pollen. Table 1 gives the results of representatives of each of the segregations found.

From these it appears that at least three of the interaction patterns described earlier (HOGENBOOM, 1972a) might be based on different genotypes. These patterns are: 1) uniform stop of pollen tube growth after penetration of about one third of the style length (0(I)); 2) thinning bundle (0(II) to about 25–30 tubes at stylar base); 3) little or no inhibition (more than about 25–30 tubes at the stylar base).

The results can be explained satisfactorily by assuming that a group of dominant genes governs *L. esculentum* pollen tube growth inhibition. In Table 1 suggestions for parental genotypes are given. In the Alexander stock presence of gene *A*, *B*, *C*, or *D*, or possible combinations of *A*, *C* and *D* or of *B*, *C* and *D* will lead to the pattern of a thinning bundle. Genes *A* and *B* appear to interact, presence of the combination *A. B.* giving interaction type 0(I). In the Verkerk stock one of the genes (say *B*) gives interaction type 0(I), otherwise the same situation exists as in the other material. If only recessive alleles are present, little or no inhibition of *L. esculentum* pollen tube growth occurs in the style. These results confirm those of GRUN and AUBERTIN (1966).

Results with F_1, F_2 and B_1

Table 2 surveys the results of tests with progenies from different crosses and selfings. The above suggestions appear to be confirmed and can be extended. Inhibition of *L. esculentum* pollen tube growth is based on a number of dominant genes, each having an action as already suggested. Plasmatic influence seems to be absent. As suggested by the results with plant 671039–3 a rather large number of dominant genes may be present in homozygous condition each giving the 0(I) pattern. Another possible explanation of these results is that one or more of the genes for inhibition are linked with a gene for gamete promotion.

In the Verkerk stock also genes *A* and *C* appear to interact, the genotype *A. C.* giving the 0(I) pattern. In Table 2 suggested parental genotypes are mentioned. The suggested segregations do not necessarily preclude other possibilities. The arbitrary line drawn at 30 tubes at the stylar base may also, and sometimes even better, be drawn at 25 tubes at the stylar base.

The number of genes governing the inhibition of *L. esculentum* pollen tubes in *L. peruvianum* stylar tissues is one or more.

Interaction between ovary of L. peruvianum and L. esculentum pollen tubes

Preliminary results of tests with progenies from crosses between *L. peruvianum* plants showing in the ovary strong inhibition of *L. esculentum* pollen tube growth and plants

Table 1. Results of tests for the interaction between the stigma and style of plants of inbred lines of *L. peruvianum* and the pollen of *L. esculentum*.

Line	Generation of inbreeding	Distribution of plants over interaction classes in mean number of pollen tubes at the stylar base								Number plants tested	Suggested parental genotype	Suggested segregation 0(I); 0(II)–30; > 30			P
		0(I)	0(II)	5	10	15	20	25	30						
V 67604 = 51–4 ⊗	I_3	44								44	..*BB*....				
V 671039 = 661577–1⊗	I_6	18								18	..*BB*....				
V 671038 = 661576–5⊗	I_6	38								38	..*BB*....				
V 671045 = 661578–5⊗	I_6	33	7							40	*AABb cc*..	3:	1		0.20–0.30
V 671033 = 661573–7⊗	I_6	19	2							21	*AABb cc*..	3:	1		0.10–0.20
V 67902 = 661517–7⊗[1]	I_5	16	12							28	*Aabb Cc*..	9:	6:	1	0.50–0.70
V 671019 = 661571–4⊗	I_6	34	10	1	1	0	1	0	0	1	48	*AaBb ccdd*	12:	3:	1	0.10–0.20
A 67801 = 257–11⊗[1]	I_2	17	10	1	1						29	*AaBb CcDd*	144:	111:	1	0.90–0.95
A 681441 = 257–11⊗⊗	I_3	56	6	10	6	5	8	4	2	4	101	*AaBb ccdd*	9:	6:	1	0.50–0.70
A 681443 = 257–11⊗⊗	I_3	53	11	15	11	2	2	0	3	5	102	*AaBb ccdd*	9:	6:	1	0.30–0.50
A 681440 = 257–11⊗⊗	I_3		60	28	9	4	1	1	0	1	104	*aaBb CcDd*		63:	1	0.30–0.50
A 681442 = 257–11⊗⊗	I_3		68	22	7	3	1	0	0	1	102	*aaBb CcDd*		63:	1	0.30–0.50
A 681446 = 257–11⊗⊗	I_3	1	95	11	1						108	*aaBB*				
A 681447 = 257–11⊗⊗	I_3	50	28	11	5						94	*AaBb CC*..	9:	7		0.50–0.70

[1] Interaction pattern of 257–11 and 661517–7:0(I).

V = Verkerk seed stock; A = Alexander steed stock.

Table 2. Results of tests for the interaction between the stigma and style of plants of progenies from crosses in L. peruvianum and the pollen of L. esculentum. The numbers of the lines 671019 and 671039 have been abbreviated to 19 and 39, respectively. The pattern of interaction of 19–13, 39–3 and 257–1 was 0(I), that of 19–10 little or no inhibition. The incompatibility genotype of 257–1 was 1ps/2ps. 257–1 is Alexander seed stock and self-incompatible, other material is Verkerk seed stock and self-compatible.

Material	Distribution of plants over interaction classes in mean number of pollen tubes at the stylar base									Number of plants tested	Suggested parental genotypes	Suggested segregation 0(I); 0(II)–30; >30			P
	0(I)	0(II)	5	10	15	20	25	30							
P⊗: 19–10⊗				1	0	2	1	0	36	40	*aabbccdd*				
19–13⊗	105	10	0	0	1					116	*AABbCcD.*	15:	1		0.10–0.20
F_1: 19–10×19–13	81	25	7	2	2	1	0	0	1	119	*aabbccdd*×*AABbCcD.*	3:	1		0.05–0.10
reciprocal	80	21	10	3	1	1	1	1		118	*AABbCcD.*×*aabbccdd*	3:	1		0.05–0.10
F_2: (19–10×19–13)⊗	64	17	3	0	0	0	1	3	3	91	*AaBbccdd*	12:	3:	1	0.05–0.10
(19–10×19–13)⊗←	35	22	9	3	2	1	0	0	3	75	*AabbCcdd*	9:	6:	1	0.05–0.10
(reciprocal)⊗	74	17	0	0	0	1				92	*AaBbccDd*	48:	15:	1	0.30–0.50
(reciprocal)⊗←	20	18	11	5	2	2	3	2	17	80	?*				
B_1: (19–10×19–13)×19–10←	7	10	9	3	8	4	2	1	32	76	?*				
(19–13×19–10)×19–10←	15	7	12	7	4	5	4	2	21	77	*AabbCcdd* × *aabbccdd*	1:	2:	1	0.50–0.70
F_1: 19–10×257–1	35									35	*aabbccdd* × *AABBCcDd*				
reciprocal	37									37	*AABBCcDd* × *aabbccdd*				
F_2: (19–10×257–1)⊗	48	17	11	1	3	1	3	3	8	95	*AaBbccdd*	9:	6:	1	0.30–0.50
(reciprocal)⊗	92									92	*AaBbCcDd....*				
P⊗ 39–3⊗	47									47	*..BB....*				
F_1: 19–10×39–3	35									35	*aabbccdd* × *..BB....*				
reciprocal	39									39	*..BB....* × *aabbccdd*				
F_2: (19–10×39–3)⊗	101									101	*AaBbCcDd....*				
(reciprocal)⊗	97									97	*AaBbCcDd....*				
B_1: (19–10×39–3)×19–10	49	1								50	*AaBbCcDd....* × *aabbccdd*				
(39–3×19–10)×19–10	49									49	*AaBbCcDd....* × *aabbccdd*				

* No explanation can yet be given for these results. Possible causes will be investigated; probably a mistake has been made. Arrows connect F_2 and B_1 made on the same F_1 plant.

Table 3. Results of tests for the interaction between the ovary of *L. peruvianum* and the pollen tubes of *L. esculentum*. The results relate to plants in which more than thirty tubes reach the stylar base. The parent plants originate from 671019–10⊗.

Material	Distribution of plants over interaction classes in:							
	mean numbers of tubes spreading over ovules				mean number of ovules penetrated			
	0	10	20	30	0	10	20	30
P: 204–7	1							
205–7				1			1	
205–11			1			1		
F1: 205–7 × 204–7		6	11	4	1	7	15	
205–11 × 204–7	1	12	4	1		10	8	

showing little inhibition are given in Table 3. These results indicate that genes for inhibition of pollen tube growth in the ovary are incompletely dominant or that this phase of inhibition is also governed by a group of genes. The precise action of such genes remains to be tested.

In some cases the number of penetrated ovules is highly variable within a plant. In extreme cases values from 0 to 51, or from 1 to 75 have been found, pointing to influences of internal or external environmental factors.

Influence of internal or external environment

Pollen vitality is known to be influenced by environmental factors and so are the processes of interaction between pollen tube and pistil leading to pollen tube growth. The latter may be influenced either directly or indirectly through influences on pistil conditions. It is therefore to be expected that results of studies on pollen tube growth inhibition and on its inheritance will also be influenced by environmental conditions. If, as in this case, such studies are carried out under the changing natural conditions, the results may also be variable. This variation has indeed been found. In Table 4 some examples are given.

The results show that later in the season lower values for pollen tube growth were

Table 4. Results indicating influence of environmental conditions on interspecific *L. peruvianum* × *L. esculentum* pistil – pollen tube interaction.

Line	Date of pollination	Distribution of plants over interaction classes in mean number of pollen tubes at stylar base					
		0(I)	0(II)		10	20	30
671019–10 ⊗	May 1969			1	2	1	36
	Sep 1970			2	5	10	64
681443 = 257–11 ⊗⊗	12 Aug 1970	53	11	26	4	3	5
	26 Aug 1970	48	37	12			
671019–6 ⊗	May 1969	8	7	5	4	3	5
	Aug 1969	8	14	6	5	0	1

Table 5. Results indicating influence of environmental conditions on the interaction between *L. peruvianum* ovary and *L. esculentum* pollen tubes.

Material	Date of pollination	Distribution of plants over interaction classes in:									
		mean number of tubes spreading over ovules					mean number of ovules penetrated				
		0	10	20	30		0	10	20	30	
671019–10⊗ × 671019–6⊗	May 1970		1	3	7	8	1	15	3		
	Sep 1970	1	8	10	1		4	13	3		

sometimes obtained. Lower values were also found in tests very early in the season. The shift towards lower values in later tests was also found for the distribution of tubes over the ovules (Table 5).

These results may be attributable to two causes: early and late in the season the pollen of *L. esculentum* may be less vital, or the physiological condition of the *L. peruvianum* pistil may be less favourable in these periods for *L. esculentum* pollen tube growth. In cases other than those given the values for number of pollen tubes at stylar base, distribution of tubes over the ovules and the number of ovules penetrated were remarkably constant, even so for plants that were vegetatively maintained and tested in different years. These two categories of results indicate that the environment has an effect not only on the pollen of *L. esculentum* but also on physiological conditions of the *L. peruvianum* pistil by some genotype-environment interaction. That the physiology of the pistil is an important source of variability is also indicated by the fact that in tests made at one date and at the same time some plants showed high within-plant variability, probably owing to differences in the physiological condition of the buds used.

The adverse environmental influence appears to be revealed in a slighter expressivity of the recessive genes governing absence of pollen tube growth inhibition, or a severer expressivity of some of the dominant genes governing inhibition, or both. It remains to be tested what environmental factors influence these processes and also whether the age of the plant plays a role.

Environmental influences on unilateral incompatibility were also suggested by MARTIN (1964) and NEWTON et al. (1970) and may account for some of the deviating results of GRUN and AUBERTIN (1966).

It is obvious that *L. esculentum* pollen tube growth in *L. peruvianum* pistils, besides being governed by genetic factors can be influenced by environmental factors and consequently a study on the inheritance of pollen tube inhibition should be carried out under controlled conditions which are optimal for pollen tube growth. Therefore some of the results given here may have to be regarded as preliminary and some of the aspects studied should also be investigated under controlled conditions.

Interspecific pollen tube growth inhibition and self-incompatibility

In the *L. peruvianum* material studied the self-compatibility is based on mutated *S*-alleles or on genes modifying the *S*- allele action (HOGENBOOM, 1972b). In some of the lines used, obtained by selfing of self-compatible plants, one intact *S*-allele may

be present and active; besides mutated *S*-alleles will be present. In other lines the *S*-allele activity is suppressed.

The results obtained in the tests for the interaction between the pistil of *L. peruvianum* and the pollen of *L. esculentum* show a pattern of inheritance of pollen tube growth inhibition which is inconsistent with that of such *S*-alleles.

The results do not suggest a relationship between self-incomptibility and inhibition of *L. esculentum* pollen tubes in *L. peruvianum*. This inhibition can be explained by the presence of a group of genes governing processes which are inhibitive to the growth of pollen tubes from other species as was also found in *Solanum* (GRUN and AUBERTIN, 1966). The inhibitive action of these processes is considered a result of differentiation, leading to incongruity between species.

The results given in this and the previous paper (HOGENBOOM, 1972a) indicate that *L. peruvianum* is at least partly heterozygous for the genes concerned, loss of unilateral incompatibility with *L. esculentum* being explained by segregation of plants homozygous recessive for all genes concerned. The results of many other studies on unilateral incompatibility (see Introduction) as well as those on bilateral incompatibility can be explained on the same basis. This will be dealt with in another paper. The hypotheses on *S*-allele polymorphism suggesting different types of self-incompatibility and self-compatibility alleles, on different specificity systems with complicated relationships and different times of action of *S*-alleles in species of self-incompatible and self-compatible plants and the different types of additional control (cf. PANDEY, 1968, 1969; and others) may thereby be replaced.

Apart from the fact that objections can be raised to the current hypotheses on the common basis of unilateral incompatibility and self-incompatibility and that none have general applicability in all the different cases of unilateral incompatibility (and bilateral incompatibility) found, there are now arguments from different types of studies which indicate that the two phenomena are distinct. These arguments derive from studies on different genera showing differences between self-incompatibility and unilateral incompatibility concerning the morphology (BATEMAN, 1943; MCGUIRE and RICK, 1954; LEWIS and CROWE, 1958; RICK, 1960; ASCHER and PELOQUIN, 1968; PANDEY, 1968, 1969; and others), the physiology (ASCHER and PELOQUIN, 1968; NEWTON et al., 1970; GÜNTHER und JÜTTERSONKE, 1971) and the genetics of pollen tube growth inhibition (GRUN and AUBERTIN, 1966, and this paper). The conclusion that self-incompatibility and unilateral incompatibility are indeed distinct phenomena is therefore justified.

The direct practical use of breaking the unilateral pollen tube growth inhibition between *L. peruvianum* and *L. esculentum* for plant breeding may seem restricted because of the rather large number of genes concerned. However, the distribution over *L. peruvianum* of the genes governing absence of *L. esculentum* pollen tube growth inhibition is possible without complications. Furthermore, the availability of simply inherited self-compatibility in *L. peruvianum* and of a high-temperature-sensitive incompatibility reaction, which can both be spread over the species, permits easy and rapid inbreeding and selection for such genes. Finally, the extended knowledge of this interspecific barrier may enable a more directed search for a simple temporary elimination of the barrier.

A better insight into the inheritance of unilateral incompatibility can be useful for a more efficient transfer of self-incompatibility into tomato for hybrid seed production.

CONCLUSIONS

The inhibition of *L. esculentum* pollen tube growth in the style of *L. peruvianum* is governed by a number of independent dominant genes, each probably governing one of the distinct processes of which the unilateral inhibition of pollen tubes is built up. Some of the genes show interallelic interaction. The interaction between the *L. peruvianum* ovary and *L. esculentum* pollen tubes seems to be governed by one or more other genes.

L. peruvianum appears to be heterozygous for the genes governing inhibition of *L. esculentum* pollen tube growth. Breakdown of the unilateral incompatibility was achieved after selfing *L. peruvianum* by segregation of plants homozygous recessive for all genes concerned.

The expressivity of the recessive genes governing absence of *L. esculentum* pollen tube growth inhibition may be influenced by environmental factors.

Self-incompatibility and interspecific pollen tube growth inhibition are distinct phenomena.

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A MODEL FOR INCONGRUITY IN INTIMATE PARTNER RELATIONSHIPS

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SUMMARY

A model for incongruity in intimate partner relationships is described. The model is tested with results from earlier research on intimate partner relationships in plants and these results are reinterpreted. It is demonstrated that in relationships between partners from different populations incongruity, i.e. non-functioning of a partner relationship resulting from a lack of genetic information in one partner about the other, plays a major role, while that of incompatibility is secondary or absent. Sexual partner relationships and host-parasite relationships are shown to be essentially similar as regards their genetic basis. Some practical implications of incongruity are mentioned.

INTIMATE PARTNER RELATIONSHIPS

Intimate relationships between two organisms, such as between sexual partners and between host and parasite, occur in several forms. In flowering plants such a relationship exists during the progame phase between pistil and pollen. To achieve fertilization the pollen germinates, forms a tube, penetrates the pistil tissues and grows in a certain direction. It finds its way through different types of tissue, enters the ovule and grows into the embryosac where it releases its contents for fusion with the egg. This complex of events includes a chain of processes in both partners, of which those in the pollen (tube) must be accurately coordinated and interact with those in the pistil (LINSKENS, 1967, 1968; STANLEY, 1971; ROSEN, 1971). In this relationship the pistil material functions as a complex of barriers and promoters, the pollen grain as an organism carrying all genetic information necessary to penetrate all barriers and react to all promoters between pollination and fertilization.

The structure and physiology of the pistil are based on a number of genes, each of which governs a barrier process or a promotion process. In the pollen a number of genes govern structure and physiology of the pollen and pollen tube. For a normal functioning of the pollen, as a counterpart of each barrier and promotion process in the pistil, the potential for the corresponding penetration and reaction process must be present in the pollen and become operative at the right moment. In short, corresponding to each barrier gene or gene-complex active in the pistil, there is a penetration gene or gene-complex active in the pollen. Corresponding to each promotion gene or gene-complex active in the pistil, there is a reaction gene or gene-complex active in the pollen. Each of these couples governs a part of the chain of metabolic processes and interactions necessary for a successful progress of progame phase and fertilization.

This description of the partner relationship between pistil and pollen in flowering plants can be applied to any other intimate partner relationship. They are apparently very different but essentially very similar (KAPOOR, 1967; HAUSTEIN, 1967; KÖHLER, 1967; FUCHS, 1971). In each case germination, penetration, growth and/or fusion require accurate coordination of the relevant processes and thus of genetic composition and gene-action of the partners. This system is as old as the existence of any intimate partner relationship between organisms.

INCOMPATIBILITY AND INCONGRUITY

At least two mechanisms can impede the normal functioning of an intimate partner relationship. One is incompatibility, an outbreeding mechanism widespread in plants (ESSER, 1967; ARASU, 1968; definition in RIEGER et al., 1968). In general it is governed by multiple alleles at one or two loci. By the *inhibiting action* of incompatibility genes (SAMPSON, 1960; LEWIS, 1965; ASCHER, 1966; PANDEY, 1967a; LINSKENS, 1968) the partner relationship is non-functional, though the potential for functioning and coordination of both partners is complete.

The other cause of non-functioning is a *lack of genetic information* in one of the partners about structure or physiology of the other partner. If, for example, in the pollen some essential penetration or reaction gene corresponding to a certain barrier or promotion in the pistil is lacking, at some moment between pollination and fertilization the pollen will not be able to penetrate that barrier, or to react adequately to that promotion. Pollen tube growth will then be inhibited. For this phenomenon of non-functioning of a partner relationship resulting from a lack of information in one partner about the other I propose to use the term incongruity. The partners do not fit together.

It follows from the above that incompatibility and incongruity are entirely different phenomena. Only the final result may be similar, e.g. in flowering plants inhibition of pollen tube growth.

Incongruity may be considered an isolating mechanism developed as a by-product of evolutionary divergence (DOBZHANSKY, 1947; STEBBINS, 1950; GRANT, 1971). In a population of freely interbreeding individuals there is no incongruity in the sexual partner relationship. When a sub-population meets a different environment, it may be differentiated physiologically and/or morphologically. The action of natural selection may directly or indirectly (through pleiotropy or linkage) lead to a change in one of the partners of the sexual relationship (e.g. an extra barrier), resulting in incongruity between the partners in this sub-population. This puts corresponding genes in the other sexual partner in this sub-population under selective pressure, which may lead to repaired congruity. Continuation of this process of differentiation may result in the sub-population gradually getting its own sexual partner relationship, different from that in the original population. From the moment at which the relationship between sexual partners in the sub-population is essentially different from that in the original population there is incongruity between sexual partners from these two populations. This incongruity between populations may be slight at first but may increase as the divergence proceeds. The same process may be caused by different ways of adaptation of populations to the same habitat (STEBBINS, 1950).

In the case of a host-parasite relationship the parasite population is dependent on the host population. Consequently a change in the host population (e.g. an extra barrier), resulting in incongruity in the relationship, puts corresponding genes in the parasite population under selective pressure. This may result in repaired congruity. The two populations show co-evolution comparable to that of sexual partners within a population. The interpopulational host-parasite relationship is therefore closely related to the intrapopulational sexual relationship. The situation is different in that the selective advantage of an extra barrier gene in the host is at least partly the result of the action of the other partner, the parasite. The co-evolution limits the degree of incongruity between the partners; mostly there is (near-)congruity.

If the ability of the parasite to adapt itself to a change in the host is lacking, the relationship can be broken. The parasite sub-population that can adapt itself to a change in the host, meets a different environment and may develop its own sexual partner relationship leading to incongruity with the original parasite population (see above). This shows how parasite speciation may accompany host differentiation.

Effects of inbreeding, which may play a role in certain cases of speciation (GRANT, 1971), on incongruity are dealt with below. It follows from the above that incongruity between partners will be the rule, congruity the exception.

A MODEL FOR INCONGRUITY IN INTIMATE PARTNER RELATIONSHIPS

For diploid flowering plants the model for incongruity in the sexual partner relationship may be described as follows. (For other organisms and other relationships the same formulation can, with certain modifications, be applied.)

1. Each population, isolated from other populations through inhibited pollen tube growth or failing fertilization, has its own pistil-pollen relationship for normal pollen germination, tube growth and fertilization after intrapopulational pollination.
2. The pistil part in this relationship is named barrier capacity (b) for short. It is the total of characters of the pistil relevant to pollen germination, tube growth and fertilization. The pollen part is named penetration capacity (p). It is the total of information in the pollen grain for growth and development in the pistil.
3. The barrier capacity is supposed to be mainly based on dominant genes, the corresponding penetration genes may be dominant or recessive. The genes or gene complexes are indicated by letters A to Z. *A* represents the penetration gene or gene complex corresponding to barrier gene or gene complex A. For convenience the same letter is used, but it is evident that gene or gene complex A (e.g. governing cutin production) is quite different from *A* (e.g. governing cutinase production). For the sake of simplicity, barrier and penetration capacities are indicated by the first letters only. Thus [b: AA] should be read as barrier capacity genes or gene complexes AA to ZZ, [b: BB] as barrier capacity genes or gene complexes BB to ZZ (AA lacking), [p: *AACC*] as penetration capacity genes or gene complexes *AA* to *ZZ*, but *BB* lacking, etc. The notation for two plants with a complete functional pistil-pollen relationship without incongruity is, for example: [b: AA, p: *AA*] and [b: AA, p: *AA*].
4. In a pistil with a certain barrier capacity, only pollen with all corresponding penetration genes can function. So an example for two plants showing incongruity is: [b: AA, p: *AA*] and [b: BB, p: *BB*]. The pollen of the second plant with penetration

capacity lacking gene or gene complex *A*, has no information about the pistil character in the first plant governed by gene or gene complex A.

TEST OF THE MODEL AND REINTERPRETATION OF EARLIER RESEARCH

Research on interspecific crosses in flowering plants (references in HOGENBOOM, 1972a) yielded much information on non-functional sexual partner (pistil-pollen) relationships. In the literature two lines of reasoning about their causes can be distinguished. Inhibited growth of pollen tubes in the pistils of an other population has in most cases been supposed to be based on incompatibility or to be strongly associated with it (ANDERSON & DE WINTON, 1931; BATEMAN, 1943; MATHER, 1943; MCGUIRE & RICK, 1954; LEWIS & CROWE, 1958; RICK, 1960; MARTIN, 1961a, 1967, 1968; PANDEY, 1962, 1964, 1967b, 1968, 1969a, b; HARDON, 1967; GÜNTHER & JÜTTERSONKE, 1971; ABDALLA & HERMSEN, 1972). The more research was carried out, the more properties had to be attributed to the incompatibility genes (S-genes) to maintain this hypothesis. Besides, arguments obtained from genetical and physicological research indicated an other system (HOGENBOOM, 1972b). Few authors (STOUT, 1952; BELLARTZ, 1956; SWAMINATHAN & MURTY, 1957; SAMPSON, 1962; MARTIN, 1963; GRUN & AUBERTIN, 1966; HOGENBOOM, 1972) interpreted the results of their studies on interpopulational pollen tube growth inhibition on the basis of other mechanisms than S-gene action.

The present model for incongruity in partner relationships results from my research on pistil-pollen relationships in *Lycopersicon* (HOGENBOOM, 1972a, b). Testing it with results from earlier studies in plants shows that it has general applicability. The most important parts of the test will be discussed and earlier data reinterpreted.

Bilateral congruity, unilateral incongruity and bilateral incongruity

Crosses between plant populations have revealed what was called bilateral compatibility, unilateral incompatibility and bilateral incompatibility. Much attention was given to the unilateral relation between self-incompatible species and self-compatible species (references in HOGENBOOM, 1972a), a phenomenon which is by no means common. To explain this unilateral incompatibility, the S-gene in the pistil was credited with the property of inhibiting the growth of 'SC-pollen' (BATEMAN, 1943; MATHER, 1943; MCGUIRE & RICK, 1954; LEWIS & CROWE, 1958; RICK, 1960; MARTIN, 1961a, 1968; PANDEY, 1962, 1964, 1967b, 1968, 1969a, b; HARDON, 1967; GÜNTHER & JÜTTERSONKE, 1971). This property does not agree with the S-gene model as described by LEWIS (1965), ASCHER (1966) and LINSKENS (1968). This also holds for the different Sc-alleles which interact with matching 'UI-genes' (ABDALLA & HERMSEN, 1972).

The other two situations, bilateral incompatibility and bilateral compatibility, which were also found between self-incompatible species and self-compatible species, did not fit in with the hypothesis on the dual role of S-alleles (cf. references in HOGENBOOM, 1972a).

According to the present model, bilateral compatibility, unilateral incompatibility and bilateral incompatibility in wide crosses generally are a consequence of the barrier and penetration capacities of the species being matching or not. The three situations can be represented by the crosses between [b: A., p: *AA*] and [b: A., p: *AA*],

between [b: A., p: *AA*] and [b: B., p: *BB*], and between [b: A. C., p: *AACC*] and [b: B., p: *BB*] respectively and should therefore be called bilateral congruity, unilateral incongruity and bilateral incongruity respectively. These situations will occur irrespective of the presence or absence of any incompatibility system, as has actually been found. Attribution of properties to S-genes and other hypotheses to explain these phenomena are no longer necessary.

From the model for incongruity and the evolution of incongruity it follows that in general incongruity will be greater as the relatedness between partners becomes more distant. This agrees with what has actually been found (SANZ, 1945; BELLARTZ, 1956; LEWIS & CROWE, 1958; GRUN, 1961; SMITH, 1968). The conclusion must be that most populations are bilaterally incongruous.

Evolution of self-compatible species

Self-compatibility is considered to be a derived condition (STEBBINS, 1950, 1957). Self-compatible species of recent origin were often found to behave differently from those of ancient origin in interspecific crosses. Recent origin is accompanied by bilateral congruity with related self-incompatible species, ancient origin by unilateral incongruity with such species. To explain this distinction the SI $\rightarrow$Sc $\rightarrow$Sc^1 $\rightarrow$SC sequence of mutations of the S-gene has been postulated (LEWIS & CROWE, 1958) and frequently been used since. This hypothesis ascribed characters to intact and mutated S-alleles which have not been borne out by experimental results. With the model for incongruity the three steps in the evolution of self-compatible species can be interpreted as follows. Self-incompatible plants in a certain species will at least partly be heterozygous for barrier capacity by accumulation of recessive mutations. Since pollen grains with incomplete penetration capacity are not successful, the plants will be highly homozygous for penetration capacity. When in this species a mutation for self-compatibility occurs this may result in the development of a sub-population of self-compatible plants. If nothing else happens, this sub-population will be bilaterally congruous with the original self-incompatible species and unilaterally incongruous with ancient self-compatible species (see also below). This is the first step, comparable to SI $\rightarrow$Sc.

When in addition to self-compatibility self-pollination occurs, this entails self-fertilization leading to segregation of barrier genes. Because of heterozygosity of probably many barrier genes, sub-populations with a barrier capacity equal to that of the original species are not likely to develop. Populations with lower barrier capacities, however, will very likely develop. Among these, especially those that lack only unessential barrier genes may be successful. These populations will still be bilaterally congruous with the original self-incompatible species, because the penetration capacity of the sub-populations is still highly complete. At the same time some of these sub-populations with lowered barrier capacity may show bilateral congruity with 'older' self-compatible species (see also below). This is the second step, comparable to Sc $\rightarrow$Sc^1.

Since in the sub-populations with a lower barrier capacity some of the penetration genes will no longer be under selective pressure, these may in the long run disappear from the population. The penetration capacity is then again adapted to the (lower) barrier capacity. In this way these populations become unilaterally incongruous with their ancestral self-incompatible species and also with populations of self-compatible

plants in which this process of lowering barrier/penetration capacity has not (yet) taken place. This is the third step, comparable to $Sc^1 \rightarrow SC$.

The first two steps may in certain cases progress quickly and have been brought about artificially in *Lycopersicon peruvianum:* through inbreeding and artificial selection the barrier capacity of *L. peruvianum* was lowered to the level of the penetration capacity of *L. esculentum* (HOGENBOOM, 1972a, b). The third step will take longer. If the self-incompatibility is temperature sensitive, one may bring about the same process within a population of self-incompatible plants.

The above means that self-compatible species which are bilaterally congruous with self-incompatible species are not intermediates in the evolution of self-compatibility from self-incompatibility (LEWIS & CROWE, 1958) but that they are species with a penetration capacity equal to that of the self-in-compatible species. This situation may be transient, namely for those species that are intermediates in the evolution from a higher to a lower barrier/penetration capacity. In this connection *Lycopersicon hirsutum*, consisting of populations in different stages on the way to being a self-compatible species, each with its own pistil-pollen relationship, is very interesting (MARTIN, 1963, 1964; CHMIELEWSKI, 1966).

The above description of the evolution of unilateral incongruity as a result of inbreeding may also be applied to certain cases of speciation in which inbreeding plays a role (GRANT, 1971).

Species hybrids and later generations

Interspecific crosses reveal complex genetical situations. The available results of studies on hybrids and further generations from crosses between populations (with the same or different barrier and penetration capacities) are very useful for testing the present model. In doing so one should take into account that the behaviour of plants after selfing and crossing and the segregations in the different generations depend on a series of conditions as follows.

1. The presence or absence of functioning S-alleles and, if present, the type of incompatibility system in one or both parents are important. Intact S-alleles lead to incompatibility by interaction only with identical S-alleles in an adequate background.
2. The number of genes in which the penetration capacities of the parents differ, may be one or more. It is one of the factors that determine the percentage of pollen in the F_1 hybrids with the penetration capacity of the parent with the highest barrier capacity. The greater the difference, the smaller the percentage of this pollen.
3. One parent may be heterozygous or homozygous for the barrier gene(s) for which in the second parent the corresponding penetration gene(s) are lacking. Therefore F_1 hybrids may or may not occur with a barrier capacity matching the penetration capacity of the second parent.
4. The degree of heterozygosity of barrier capacities in the parents determines segregation of different barrier capacities in the F_1 hybrids.
5. The number of genes governing barrier and penetration capacities probably being large, linkage of one or more of them with the S-locus is likely to occur. A certain degree of linkage may for example occur in one parent between the S-locus and the barrier gene(s) for which the corresponding penetration gene(s) in the other parent are lacking.

6. In interspecific hybrids recombination is often restricted. For instance crossing-over between the S-locus and the locus (loci) for the penetration gene(s) lacking in one parent may be slight or absent. A possible consequence is that in hybrids of the cross self-compatible × self-incompatible no Sc-pollen develops that has the complete penetration capacity of the hybrids.
7. In interspecific hybrids reduced fertility is often found.
8. In F_2 and later generations the situation in regard to crossing-over etc. may change and become very complicated by segregation of modifyers and disturbed segregations from different causes, such as interaction between genes and cytoplasm.

Taking these conditions into account, the available results of inheritance studies on the behaviour of plants in various interpopulational crosses and in further generations (ANDERSON & DE WINTON, 1931; MATHER, 1943; STOUT, 1952; MCGUIRE & RICK, 1954; PANDEY, 1957, 1962, 1968; GARDÉ, 1959; RICK, 1960; MARTIN, 1961a, 1964, 1967, 1968; CHMIELEWSKI, 1962, 1968; HARDON, 1967; GÜNTHER & JÜTTERSONKE, 1971; HOGENBOOM, 1972a, b) can all be reinterpreted and explained on the same basis with the present model. The results of GRUN & AUBERTIN (1966) also fully agree with it. Their non-acceptor genes in one parent are barrier genes for which in the other parent the corresponding penetration genes are lacking.

The results, found in different genera, are all confirmations of the model. They show that in interspecific crosses two mechanisms are active: incongruity as the most important one and, if present and secondary, incompatibility. In interspecific hybrid plants the two mechanisms may occur together: part of the pollen does not function after self-pollination because of lack of penetration genes (self-incongruity), an other part does not do so because of action of S-alleles (self-incompatibility). The two may be difficult to distinguish.

S-gene polymorphism

S-gene polymorphism in the control of interspecific crosses has been assumed to account for the difference, as found in *Nicotiana*, between self-incompatible plants accepting and such plants rejecting the pollen of an other population (ANDERSON & DE WINTON, 1931; PANDEY, 1964, 1967b, 1968, 1969a, b). The hypothesis distinguishes S-alleles with quite different actions in relation to Sc- and S-alleles from other populations.

As MATHER (1943) stated, 'this is a complicating assumption for which there is no external evidence, and is unlikely to be true because it postulates a sharp difference between two allelomorphs, which otherwise show qualitatively similar behaviour, in their reaction to an allelomorph from a different species'.

The results leading to the hypothesis of S-gene polymorphism can be reinterpreted with the model for incongruity, by supposing that the S-locus and one or more loci for barrier genes are close together (cf. SIMMONDS, 1966), so that linkage occurs between certain S-allele(s) and certain barrier gene(s) for which the corresponding penetration gene(s) in the pollen parent are lacking. E.g. if in the chromosome segment —A—S—B— (A and B barrier genes) no crossing-over occurs, this allows, by mutations in A and/or B, at least four different S-allele/barrier gene combinations. With more genes more possibilities occur. In interpopulational crosses these may each give their own reaction pattern, depending on the penetration capacity of the pollen

parent. This pattern is thus mainly determined by barrier and penetration capacities. As a result of linkage it only seems as if S-alleles play this role. Certain results (PANDEY, 1969a) point to some crossing over between S-locus and some barrier gene.

In this way and taking into account the earlier mentioned conditions in interspecific crosses, all results can be explained. The model for incongruity also explains how this 'polymorphism' may as well occur in populations of self-compatible plants, at least in the first period of self-compatibility. This has been found in *Nicotiana* (PANDEY, 1968, 1969a) and *Lycopersicon* (CHMIELEWSKI, 1966, 1968; HOGENBOOM, 1972a, b). This phenomenon is at the same time an indication of an other mechanism than incompatibility.

Some results obtained with *Nicotiana* (PANDEY 1967b, 1968, 1969) can be used for the test only after a correction. PANDEY considers a cross of ♀ with long style × ♂ with short style compatible if the pollen tubes grow the length of the short style. However, this is just one of the possible expressions of incongruity: by adaptation of the penetration capacity to the barrier capacity, which may in certain cases be adaptation to a short style, the pollen is not informed to grow far enough in long styles of other species. This adaptation to style length is also referred to in the work of SWAMINATHAN & MURTY (1957). It is, however, certainly not a common phenomenon (cf. ANDERSON & DE WINTON, 1931; MATHER, 1943; STOUT, 1952; BELLARTZ, 1956).

Complex patterns of crossability

Stepwise unilateral relations between populations, resulting in complex crossability patterns, have been found in *Lycopersicon* (MCGUIRE & RICK, 1954; CHMIELEWSKI, 1962, 1968; MARTIN, 1961b, 1963), *Nicotiana* (PANDEY, 1968, 1969b) and *Petunia* (STOUT, 1952). These patterns were so complex that they could not be simply explained on the basis of incompatibility genes. The fact that such patterns are found between populations of self-compatible plants and also between populations of self-incompatible plants indicates that we are not concerned with effects of S-gene action.

Yet PANDEY (1968, 1969b) gives an explanation on the basis of incompatibility. He therefore introduces a complex and extremely polymorphic S-gene, consisting of elements for primary specificity, controlling interspecific incompatibility, and for secondary specificity, controlling intraspecific incompatibility. Incompatibility occurs when specificity elements expressed in the pollen are evenly matched by those expressed in the style. To reject pollen of other populations a primary specificity element per population is necessary. These different primary specificity elements are thought to have evolved through duplication and differentiation and spread and be maintained by selection pressure based on the presence of foreign pollen. Isolation of the population would lead to S-gene erosion. To explain complex patterns PANDEY supposes an inverse relationship between stylar and pollen compatibilities, resulting from complex interactions, which are different for pollen and style, between the elements for primary and secondary specificity in the S-gene complex. This hypothesis on the evolution, the structure and the action of the S-gene to explain interspecific relationships, which is an extension of that of LEWIS & CROWE (1958), is a complex of rather speculative and disputable suppositions.

MARTIN (1961b, 1963) supposes different balances of inhibiting substances in the style and of stimulating substances in the pollen. His 'hypothesis of balanced polygen-

Table 1. Explanation of stepwise crossability patterns with the model for incongruity, on the basis of different barrier and penetration capacities. + = congruity, − = incongruity between pistil and pollen.

Barrier capacity ♀		Penetration capacity ♂			
		population 1 *DD*	population 2 *CC*	population 3 *BB*	population 4 *AA*
Population 1	D .	+	+	+	+
Population 2	C .	−	+	+	+
Population 3	B .	−	−	+	+
Population 4	A .	−	−	−	+

ic control of substances affecting pollen tube growth', which assumes that unilateral incompatibility is 'an expression of the basic physiological mechanism controlling pollen tube growth', not based on incompatibility, agrees on some fundamental points with the present model.

With the model for incongruity between populations the complex crossability patterns are easily explained. Each population has its own pistil-pollen relationship, and its own barrier and penetration capacities. In Table 1 a complex pattern is shown and an explanation on the basis of different barrier and penetration capacities is given which agrees with the results obtained. Actually, according to the present model, such complex patterns are likely to be the normal situation in taxa with a certain degree of differentiation. After the discussion on the evolution of populations of self-compatible plants it is clear that self-compatible species are most likely to be found in the top left-hand portion of the table. The distribution of sub-populations of a species over more positions in the table indicates that the species is under development. The distribution results from different barrier and penetration capacities, probably as a result of inbreeding.

Also for this test with the *Nicotiana* results (PANDEY, 1968, 1969b) it should be remembered that if pollen tubes do not grow into the ovules as a consequence of style length, there is incongruity.

The conclusion from this and the previous section is that the S-gene is not a supergene. Different properties ascribed to it, which do not agree with the S-gene model of LEWIS (1965) and the other authors mentioned earlier, are in fact based on other genes and other principles.

Incongruity and contact between populations

In some hypotheses on the evolution of barriers in the pistil of one species to the pollen of an other, contact between populations is a necessary presupposition (GRUN & RADLOW, 1961; PANDEY, 1969b; ABDALLA & HERMSEN, 1972). Reproductive isolation between populations is then suggested to result from a selective advantage of genes which keep foreign pollen out of the population (e.g. keep pollen with a self-compatibility allele out of a population of self-incompatible plants). This means selection for reproductive isolation, a process called the Wallace-effect (GRANT, 1966, 1971). With

regard to incongruity, occurrence of such a process is unlikely for the following reasons.

Incongruity occurs between populations which never were or have long ceased to be in contact (GRUN & RADLOW, 1961; CHMIELEWSKI, 1962, 1968; PANDEY, 1968). Actually, this is the rule.

The hypothesis of a selective advantage of genes which keep pollen with a self-compatibility allele out of a population of self-incompatible plants is based on the supposition that unfavourable effects of inbreeding accompany self-compatibility. In many cases, however, this will not occur, at least not in the beginning, because the plant is still a cross-pollinator. Furthermore, a low and gradually developing degree of selfing is frequently not unfavourable (STEBBINS, 1957). The self-compatibility allele will therefore have spread before a barrier has formed. This may also result from recent populations of self-compatible plants being bilaterally congruous with the original self-incompatible species.

Even in those cases where self-compatibility is unfavourable this is generally not a basis for selective pressure for isolation, because it is unlikely, as a consequence of incongruity, that the self-compatibility, if introduced in a self-incompatible species by crossing, is expressed in the hybrids or in the first generations. The fact that incongruity also occurs between populations of self-incompatible plants and between populations of self-compatible plants and in the cross ♀ self-compatible × ♂ self-incompatible further indicates that it does not result from selection pressure for genes which keep pollen with a self-compatibility allele out of the population (cf. references in HOGENBOOM, 1972a).

From the results of GRUN & AUBERTIN (1966) and HOGENBOOM (1972a, b) it appears that incongruity between populations may be based on independent genes, each of which may cause the barrier. Besides, incongruity is rarely the primary barrier between populations (STEBBINS, 1950); it often occurs in addition to e.g. embryo abortion. These results indicate that the selective advantage of the genes governing incongruity is not the result of any action against pollen of other populations.

In contrast to the above hypotheses the model for incongruity, suggesting that incongruity is a by-product of differentiation and develops irrespective of incompatibility, with and without contact, is in agreement with the experimental results.

Sexual partner relationships and host-parasite relationships

The suggestion that a male gamete entering into female tissues and a parasite entering into its host display the same type of activity is not new (BURGEFF, 1920; EAST, 1929; WEIDEL, 1958). In both systems a mutual coordination of genes and gene activity is necessary for germination, penetration, growth in a certain direction and exchange of substances. This and other facts (see above) indicate that sexual partner relationships and host-parasite relationships may be based on the same genetic principles. If this suggestion is right the present model for incongruity in partner relationships should also be applicable to host-parasite relationships. To test this applicability, special attention was given to some host-parasite relationships, which have been genetically well analysed, such as occur between certain higher plants and fungi.

The patterns of crossability found between populations of plants (a simplification is given in Table 1) and the schemes for the gene-for-gene relationships between populations of host and parasite (FLOR, 1955, 1956; PERSON, 1959) present the same charac-

teristics. In its most simple form there is in fact no essential difference: certain genes in one partner (pistil/host) correspond to certain genes in the other (pollen/parasite) through complementary genic systems; into a partner with a certain barrier capacity only partners with all corresponding penetration genes can enter. Only a translation is needed, e.g. as follows: host = pistil of a population; parasite = pollen of an other population; resistance genes = those barrier genes in the pistil for which the corresponding penetration gene(s) in the pollen are lacking; virulence genes = penetration genes; resistance/susceptibility = a consequence of incongruity/congruity, the partner relationship is incomplete/complete. Also the origin of incongruity in host-parasite relationships has much in common with that in sexual relationships (cf. above and PERSON, 1959).

Thus, the model for incongruity in partner relationships is indeed generally applicable. At the same time this study indicates that FLOR's principle of the gene-for-gene relationship has a much wider applicability. The hypothesis that, from a genetic point of view, the host-parasite relationship is not essentially different from the pistil-pollen or other sexual relationships, is justified. Connection of the two active fields of research, on sexual partner relationships and on host-parasite relationships, may bring about an extremely interesting scientific cross-fertilization. For future research on partner relationships the methods in the two fields may complement each other.

The present model suggests that in intimate partner relationships four different relationships will be found: 1) gene-for-gene; 2) genes-for-gene; 3) gene-for-genes; 4) genes-for-genes. In the case of host-parasite relationships in situations 1 and 2 changes in the host are probably rapidly offset by corresponding changes in the parasite, as has been found in e.g. the *Solanum tuberosum–Phytophthora infestans* relationship. In situation 3 a change in the host is less rapidly offset by a corresponding change in the parasite. It may very well represent stable monogenic or oligogenic resistances such as occur between *Cucumis sativus* and *Cladosporium cucumerinum* or *Phaseolus vulgaris* and *Colletotrichum lindemuthianum*. The history of the latter relationship indicates that a partner relationship can change from a gene-for-gene to a gene-for-genes relationship. Situation 4 may represent the stable polygenic resistance. If this suggestion is borne out it is important that breeders should pay special attention to relationship 3 to obtain long-term resistance easily.

PRACTICAL IMPLICATIONS

To realize interspecific crosses incongruity will have to be neutralized. In certain cases this is probably possible by environmental influence on one or both partners. One may think of inactivation of a certain barrier gene, for which in the pollen of the other species the corresponding penetration gene or gene complex is lacking, e.g. by high temperature or other physical treatment (if the gene inhibits pollen tube growth in the style), or of offsetting the effect of such a barrier gene by application of chemicals (if the gene inhibits germination or forms an other stigmatic barrier). The solution is simple if the incongruity can be solved by removing the barrier, e.g. by removing stylar parts. Other possibilities may lie in the substitution of an absent promotion or in making up a shortage in penetration capacity. In the literature interesting examples can be found of succesful treatments (GARDELLA, 1950; SWAMINATHAN & MURTY,

1959; KNOX et al., 1972).

The discussion on the evolution of self-compatible species implicitly indicates a possibility of breaking incongruity between species. Differences in barrier and penetration capacities may be solved by inbreeding in cross-fertilizers.

An interesting matter is how incongruity might be exploited for practical purposes. Two possibilities are mentioned.

1. In hermaphrodites incongruity may be applied to prevent self-fertilization. This possibility is interesting to plant breeders with a view to hybrid seed production. To prevent self-fertilization, self-incompatibility and male sterility are used. Incongruity is an interesting third mechanism: by creating a shortage of penetration capacity in the female parent, self-fertilization is made impossible. It may be done by introduction from a related species of an extra barrier gene into the female parent. The corresponding penetration gene(s) must be introduced into the male. Another possibility is artificial mutation of an essential penetration gene.
2. Incongruity may also be applied for the eradication of species. In host-parasite relationships one might think of an extra barrier in the host which cannot be overcome by the parasite. For plant breeders this is the ideal resistance, examples of which have been mentioned earlier. In unisexuals one may think of bringing together incongruent gametes in cases where incongruity is only expressed just before or during fertilization. Gametes are thus wasted, without producing individuals. This technique has actually been practised in insect control (LAVEN, 1967). It may perhaps also be a possibility for eradication of other parasites, such as fungi or bacteria. These possibilities for genetic control by application of incongruity require further research.

CONCLUSIONS

The present model for incongruity in intimate partner relationships fully agrees with the results obtained in studies on sexual partner relationships in higher plants and those on host-parasite relationships. It appears to have general applicability.

At least two independent mechanisms occur which can prevent the functioning of an intimate partner relationship, viz incompatibility, leading to an inhibition of the functioning of a complete relationship, and incongruity – i.e. non-functioning resulting from a lack of genetic information in one or both of the partners which is a by-product of evolutionary divergence. In interpopulational relationships incongruity plays a major role, the role of incompatibility is secondary or absent.

Incongruity between partners is the rule, congruity the exception.

The S-gene is not a super-gene. Phenomena have been ascribed to it which are in fact governed by other genes and are based on other principles.

Host-parasite relationships and sexual partner relationships are essentially similar as regards their genetic basis. Cooperation in the two fields of research is important.

Exploitation of incongruity may open up interesting prospects.

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