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Evaluasie van die genetiese variasie in genaturaliseerde populasies van *Medicago polymorpha* in die Winterreënstreek van Suid-Afrika en die moontlikheid om 'n teelprogram vir die verbetering van die spesie as weidingsgewas te ontwikkel

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ABSTRACT

Medicago polymorpha is an annual, self pollinating leguminous plant which is used in short term rotation with cereal crops in the Winter Rainfall Region. Both the vegetative growth and pods provide a protein rich fodder, whereas its nitrogen fixing abilities can add to increased grain yields and quality. Although an exotic species, *M.polymorpha* has successfully adapted to local environmental conditions and naturalised populations are widespread within the region. This source of germplasm should be protected and utilised to satisfy the need for locally developed medic cultivars. Research was therefore undertaken to determine the natural diversity within these populations, as well as to ascertain the potential for generating additional diversity through cross-breeding. In doing so, problems specific to this crop could be identified and taken into consideration when planning a breeding programme.

During November/December 1994, 516 *M.polymorpha* accessions were collected from 73 different locations throughout the Winter Rainfall Region. Following seed multiplication, 165 accessions were subjected to Phase I evaluation in the 1995 season and the remaining 212 accessions in the subsequent year (1996). Single rows served as experimental unit and the commercial cultivar, 'Santiago', was included as control. During Phase II evaluation a replicated design was used to compare 16 smooth pod genotypes with Santiago in plot-sized units. In the spring of 1995, Santiago and four promising accessions, selected for vegetative potential only, were crossed in accordance to a complete diallel design. Both the F_1 and F_2 progenies, created by either self pollination or crossing of the five parental lines, were evaluated for several agronomically important characteristics. The choice of parental lines also provided an opportunity to test Simon's hypothesis (Simon, 1964) on the inheritance of the smooth pod genotype. An evaluation of the feasibility of resistance testing was conducted by subjecting 36 accessions to high population densities of the blue-green aphid (*Acyrthosiphon kondoi*) under controlled environmental conditions.

From Phase I evaluation it became evident that the collected germplasm is characterised by a considerable amount of variation for vegetative production and flowering date. Due to the labour involved in the threshing of pods, only vegetative production was used as criterium for initial selection. Hence a total of 75 germplasm lines were selected over a period of two seasons. These selected lines, though small in number, still displayed a considerable amount of variation. The most suitable selection method for future execution of Phase I evaluation seems to be a combination of index selection and independent culling levels.

The Phase II evaluation trial was characterised by low repeatability of entry means. This was probably due to ineffective blocking, high levels of genetic variation within genotypes and incorrect sampling methods. In an attempt to create an actual basis for selection, it was decided to make use of shortfall testing. A summary of the lines that fell short of the "best" or could be considered as marginal cases, is provided for each of the characteristics measured and index values could serve as further basis for selection. Recommendations for future execution of Phase II evaluation are made.

Griffing's Method 4, model I (Griffing, 1956) was used to perform an analysis of the diallel cross with respect to general and specific combining abilities. In an attempt to determine the available genetic variation, the parental lines were subjected to a separate analysis. For both the parental and progeny groupings no definite conclusions could be made due to considerable variation in the results obtained across generations. Therefore only a summary of the lines, exhibiting the best general and specific combining abilities in both the F_1 and F_2 generations for a number of agronomically important characteristics, is given. Chi-square testing provided evidence in support of Simon's hypothesis (Simon, 1964), although not irrefutable.

Resistance testing was performed on the basis of two criteria, viz. the number of normal leaves per plant and the extent to which the leaves were dwarfed and internodes shortened. Use of neither of these criteria did however allow for making a reliable distinction among lines. Resistance breeding is seemingly not the most cost-effective solution to the problem. Chemical control or gene introgression via wide hybridisation could be considered as alternative strategies. If it is decided to continue with resistance testing, adaptations to the current method will be necessary.

All relevant information obtained from the study was eventually considered in planning a breeding programme for the genetic improvement of *M.polymorpha*. Three different selection methods, based on the principles of single seed descent and bulk-population selection, are proposed and differ mainly in the management of the early, segregating generations. The economic implications of each of the proposed breeding strategies must be taken into account when deciding which method to apply.

OPSOMMING

Medicago polymorpha is 'n selfbestuiwende, eenjarige peulgewas wat in die Winterreënstreek van Suid-Afrika in korttermyn rotasie met kleingrane benut word. Die plantmateriaal en peule verskaf hoë gehalte proteïenvoer, terwyl stikstofbinding bydra tot die verhoging van graanopbrengste en die verbetering van graankwaliteit. Hoewel uitheems, het *M.polymorpha* suksesvol by plaaslike omgewingstoestande aangepas en word dit wydverspreid aangetref. Hierdie kiemplasmabron moet egter bewaar en benut word, ten einde in die behoefte aan 'n plaaslik ontwikkelde medickultivar te voldoen. 'n Ondersoek is dus geloods om 'n aanduiding te verkry van die natuurlike variasie binne genaturaliseerde populasies van *M.polymorpha* in die Winterreënstreek, asook die potensiaal om nuwe variasie deur kruisings daar te stel. Terselfdertyd sou spesifieke probleme wat deur die betrokke gewas gelewer word, uitgewys en in die beplanning van 'n praktykgerigte teelprogram oorweeg kan word.

Gedurende November/Desember 1994 is 516 *M.polymorpha* aanwinste in 73 verskillende lokaliteite in die Winterreënstreek versamel. Na saadvermeerdering is 'n groep van 165 aanwinste in die 1995 groeiseisoen, en 'n verdere 212 aanwinste in die daaropvolgende jaar (1996) aan Fase I evaluasie onderwerp. Enkelrye het as eksperimentele eenheid gedien en die kommersiële kultivar, 'Santiago', is as kontrole ingesluit. Om ekonomiese redes is slegs kiemplasmalyne wat oor gladde peule beskik vir Fase II evaluasie in oorweging geneem. Toetsing het aan die hand van 'n gerepliseerde proefontwerp geskied en 16 genotipes is op perseelbasis met Santiago vergelyk. Gedurende die lente van 1995 is vier belowende aanwinste op grond van die beskikbare velddata geselekteer en tesame met Santiago as kruisingsouers in 'n volledige dialleel benut. Beide die selfbestuifde en kruisingsnageslagte is oor die verloop van twee generasies (F_1 en F_2) ten opsigte van verskeie agronomies belangrike eienskappe geëvalueer. Die keuse van ouerlyne het terselfdertyd 'n geleentheid gebied om ondersoek in te stel na Simon (1964) se hipotese aangaande die oorerwingswyse van die gladde peul kenmerk. Die moontlikheid van weerstandsevaluasie is ook oorweeg deur 36 aanwinste onder gekontroleerde toestande aan hoë populasievlakke van die blougroenplantluis (*Acyrthosiphon kondoi*) bloot te stel.

Tydens Fase I evaluasie het dit duidelik geblyk dat die versamelde kiemplasma deur 'n geweldige hoeveelheid variasie in vegetatiewe materiaalproduksie, asook die aantal dae tot blom gekenmerk word. Weens die tydsfaktor verbonde aan die uitdors van groot peulmonsters, het vegetatiewe produksie as enigste kriterium vir die eerste vlak van seleksie gedien en is 'n totaal van 75 kiemplasmalyne oor die verloop van twee seisoene geselekteer. Hierdie klein groepie van geselekteerde lyne het egter steeds 'n

aansienlike mate van variasie in peulmassa, saadmassa en saadpersentasie getoon. Die mees geskikte seleksiemetode vir toekomstige uitvoering van Fase I evaluasie skyn 'n kombinasie van indeksseleksie en onafhanklik bepaalde afsnydingsvlakke te wees.

Die Fase II evaluasieproef is gekenmerk deur swak herhaalbaarheid van die inskrywingsgemiddeldes. Die vernaamste redes hiervoor was waarskynlik oneffektiewe blokking, hoë vlakke van variasie binne genotipes en foutiewe monsternemingstegnieke. In 'n poging om wel 'n basis vir seleksie daar te stel, is van kortskiettoetsing gebruik gemaak. 'n Opsomming van die lyne wat met betrekking tot elkeen van die gemete eienskappe betekenisvol swakker as die "beste" gevaar het of as grensgevalle beskou kan word, is verskaf en verdere seleksie moet aan die hand van indekswaardes toegepas word. Verskeie aanbevelings is met die oog op die toekomstige uitvoering van Fase II evaluasie gemaak.

Griffing (1956) se Metode 4, model I is benut om 'n analise van die dialleelkruising in terme van algemene en spesifieke kombineervermoëns uit te voer. Die ouerlyne is afsonderlik ontleed ten einde 'n aanduiding van die beskikbare genetiese variasie te verkry. Vir beide die ouerlike en nageslagsgroeperings kon geen definitiewe gevolgtrekkings egter gemaak word nie, aangesien die verkreë resultate heelwat variasie oor generasies getoon het. Slegs 'n opsomming van die beste algemene en spesifieke kombineerders, soos uitgewys in die F_1 - en F_2 -generasies vir 'n aantal agronomies belangrike eienskappe, word dus verskaf. Chi-kwadraattoetsing het wel getuienis ter ondersteuning van Simon (1964) se hipotese gelewer, maar kan weens die gepaardgaande teenstrydighede nie as onomstootlik beskou word nie.

Weerstandsevaluasie het aan die hand van twee kriteria geskied, naamlik die aantal normale blare per plant en die mate waartoe die blare verdwerg en die internodes verkort is. Geeneen van hierdie kriteria kon egter gebruik word om 'n betekenisvolle onderskeid tussen lyne tref nie. Dit mag dus wees dat weerstandsteling nie die mees kostedoeltreffende benadering tot die probleem is nie. Chemiese beheer of interspesie kruisings kan dan as moontlike alternatiewe oorweeg word. Sou wel besluit word om met weerstandsteling voort te gaan, moet 'n aantal wysigings aan die huidige metode aangebring word.

Alle toepaslike inligting is uiteindelik oorweeg in die beplanning van 'n teelprogram vir die genetiese verbetering van *Medicago polymorpha*. Drie verskillende seleksiemetodes, gebaseer op enkelsaadnageslag- en bulkpopulasieseleksie, is voorgestel en verskil hoofsaaklik in hantering van die vroeë, segregerende generasies. Die ekonomiese uitvoerbaarheid van elkeen van die voorgestelde seleksiemetodes sal bepaal watter telingsmetodiek gevolg gaan word.