Range of susceptibility within and between diploid and tetraploid strains of *Chloris gayana* (Rhodes grass) to *Meloidogyne javanica*

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SUMMARY

Host status of 22 accessions of *Chloris gayana* (Rhodes grass) to *Meloidogyne javanica* (root-knot nematode) was assessed. Eleven diploid and eleven tetraploid types were screened. Tetraploids showed a wider range of susceptibility and were generally more susceptible than diploids. The maximum egg-masses recorded on a diploid plant was 213 and on a tetraploid 412. Not all diploids were sufficiently resistant to suppress nematode population increase. Number of egg-masses was a reliable discriminant to identify susceptible plants and select for resistance. Host status is reported with reference to a breeding programme to produce an improved root-knot resistant Rhodes grass to control *M. javanica* in tobacco rotations. Results are discussed in relation to source and history of material and the nature of resistance.

RÉSUMÉ

Échelle de sensibilité à Meloidogyne javanica *chez les souches diploïdes et tetraploïdes de* Chloris gayana

La qualité de plante-hôte de 22 accessions de *Chloris gayana* vis-à-vis de *Meloidogyne javanica* est définie. Onze souches diploïdes et onze tetraploïdes ont été testées. Les souches tetraploïdes montrent une grande variabilité dans leur sensibilité et sont généralement plus sensibles que les diploïdes. Le nombre maximum de masses d'œufs observées sur une plante diploïde est de 213 contre 412 pour une plante tetraploïde. Aucune souche diploïde n'est suffisamment résistante pour empêcher une augmentation de la population du nématode. Le nombre de masses d'œufs est un caractère discriminant pour identifier les plantes sensibles et sélectionner en vue de la résistance. La qualité d'hôte est étudiée dans l'optique d'un programme de sélection visant la création de souches de *C. gayana* résistantes à *M. javanica* utilisables en rotation avec le tabac. Les résultats sont discutés en relation avec l'origine et l'histoire du matériel végétal, et la nature de la résistance.

A programme has been initiated in Zimbabwe to combine resistance to *Meloidogyne javanica* (root-knot) with improved yield and quality of herbage in *Chloris gayana* (Rhodes grass). Initial results showed that resistance to root-knot could be improved by selection within tetraploid lines of Rhodes grass (York, in press, *a*). The diploid cultivar Katambora has been used to control root-knot in tobacco rotations for some years. It produces less herbage than tetraploid strains such as " Giant ", grown in Zimbabwe. It is probably more variable in host status than assumed, allowing more reproduction of *M. javanica* than desired (York, 1989).

Results with tetraploid Rhodes grass have shown these to be generally more susceptible to *M. javanica* than Katambora both in field tests (Shepherd, 1968) and as determined by nematode reproduction in controlled inoculation tests (Way, pers. comm.; York, in press, *a*). The experiments reported were conducted to provide wider information on the degree of susceptibility and distribution of resistance in diploid and tetraploid strains and cultivars of Rhodes grass.

Materials and methods

Details of the various accessions of Rhodes grass assessed are given in Table 1. Rhind (1977) asserted that all South African Rhodes grasses examined were diploid. From habit and general appearance the collections from South Africa appeared diploid. Cg29 material was grown as tillers from ten plants in a spaced plant nursery. Other material was derived from seed germinated on moistened filter pads in Petri dishes at 25 °C. Plants were grown in black polythene pouches (145 × 45 × 25 mm) containing roughly 200 cm^3 of methyl bromide treated granitic sand soil. Soil was treated with methyl bromide at 50 g/m^2 in a bed about 20 cm deep under polythene, and stored for two weeks before use. Inoculum was obtained from roots of tomato plants maintaining a culture of *M. javanica*. Roots were chopped and mixed with commercial bleach diluted one to eight volumes with tap water for five minutes (Hussey & Barker, 1973), and released eggs collected on a 38 µm mesh sieve. Plants were inoculated when those grown

Table 1

Origin and ploidy of 22 accessions of Rhodes grass *(Chloris gayana)* assessed for host status to root-knot nematode *(Meloidogyne javanica)*

Accession	**Ploidy*	*Cv/Strain*	*Source*	*Comments*
Cg 2	2n	unnamed	CSIRO	Originally from Henderson Res. Stn. Zimbabwe
Cg12	2n	Katambora	CSIRO	Early stock of this cv. sent to Australia
Cg21	2n	Nzoia (improved)	CSIRO	Once popular Kenyan cv.
Cg29	2n	Nzoia	CSIRO	Once popular Kenyan cv.
Cg34	4n	Chepararia	CSIRO	Early Malawian strain
Cg38	4n	Rongai	CSIRO	Kenyan strain
Cg40	4n	Giant	CSIRO	Zimbabwean strain or range of strains
Cg52	2n	Pioneer	CSIRO	Early Australian cv. formerly " Commercial "
Cg53	(2n)	West Transvaal	SA	Wild material assumed 2n (Rhind, 1977)
Cg54	(2n)	Kruger Park	SA	
Cg55	(2n)	East Transvaal	SA	
Cg56	(2n)	Natal	SA	
Cg57	(2n)	Soutpan	SA	
Cg71	4n	Giant	Zimbabwe farmer	An old srain handed down father to son
Cg74	2n	Katambora	Zimbabwe farmer	Considered authentic by DR & SS Seed Services
	4n	Mbarara	Commercial stocks ex isolated seed increase plots at Grasslands Res. Stn.	Older more variable East African strains
	4n	Masaba		
	4n	Elmba		Selected from the above (Boonman, 1978)
	4n	Boma		
	4n	Callide		Australian cvv. originally from East African material (Barnard, 1972)
	4n	Samford		
	4n	Mt Makulu 56	SI plots at GRS	A productive strain selected in Zambia (van Rensburg, 1968)

*Ploidy determined by Hutton (1963), Pritchard and Gould (1964), or in the case of named cvv., Cg71 and Cg74 established on other stocks.

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SA = Department of Agricultural Economics and Marketing, Directorate of Plant and Seed Control, Pvt Bag XI79, Pretoria, South Africa.

DR & SS = Department of Research and Specialist Services, P.O.Box 8108, Causeway, Harare, Zimbabwe.

from germinated seed were six weeks old. Poor germination and plant mortality resulted in different numbers of plants being assessed for each accession (Tab. 2). Inoculum applied in 1 cm^3 suspension on three fortnightly occasions amounted to an accumulated 3 700 eggs per plant. Uniformity of application was insured by frequent sample dose monitoring.

Initially plants were arranged in variety rows inoculated across rows. After final inoculation plants were labelled individually and fully randomised. Plants were further randomised prior to washing out. Sampling was destructive. Bulk root weights were assessed for varieties as earlier tests had shown no relationship between root weight and susceptibility for individual plants (York, in press, *a*). Nine weeks after the final inoculation plant roots were washed free of soil. Plant roots wrapped in butter muslin were stained in 0.15 % aqueous Phloxine B (Hollbrook, Knauft & Dickson, 1983) and examined

Table 2

Host status of diploid and tetraploid *C. gayana* accessions to *M. javanica* based upon numbers of egg-masses per plant

Accession	*n*	*% plants with*				*mean egg-masses per plant*	*log 10 egg-masses + 2*	
		0	*⩽ 5*	*⩽ 10*	*> 10*		*mean*	*SD*
		egg-masses						
Cg 2	24	8	29	13	50	36.0	1.212	.608
Cg12	25	28	32	8	32	14.1	0.856	.532
Cg21	19	47	32	0	21	7.4	0.636	.484
Cg29	20	60	20	5	15	3.3	0.548	363
Cg52	26	26	35	4	35	16.0	0.846	.545
Cg53	28	21	36	4	39	20.0	0.937	.573
Cg54	24	33	17	8	42	19.6	0.912	.591
Cg55	23	22	17	13	48	10.0	1.039	.555
Cg56	24	17	33	4	46	16.8	1.005	.516
Cg57	10	20	20	20	40	15.3	0.989	.523
Cg74	25	48	40	8	4	2.2	0.527	.272
Diploid	248	29	29	7	34	15.9	0.864	.215
Tetraploid	245	9	15	9	67	45.8	1.329	.179
Cg34	23	9	17	4	70	32.9	1.293	.516
Cg38	23	13	26	22	39	17.1	1.041	.473
Cg40	26	12	19	0	69	43.0	1.302	.645
Cg71	7	0	14	0	86	86.4	1.555	.675
Mbarara	27	0	11	11	78	46.3	1.496	.436
Masaba	23	9	4	26	61	42.9	1.397	.557
Elmba	23	4	4	4	88	89.9	1.734	.530
Boma	23	9	17	0	74	55.0	1.440	.663
Callide	25	8	12	0	80	48.8	1.455	.563
Samford	22	23	27	14	36	27.2	1.001	.650
Mt. Makulu 56	23	9	9	13	69	41.3	1.404	.546

immediately to pick off infection sites and egg-masses. These were stored in 4 % formalin and counted under stereomicroscope. Egg-masses were then bulked by variety extracted as for inoculum and numbers estimated by series dilution and subsampling.

In a contemporaneous but separate test larger numbers of Mbarara, Elmba and Callide plants were treated in a similar way to the above. These plants were assessed at ten weeks by simple visual assessment. Immediately after staining they were scored for egg-mass numbers. Five categories were used : 0 : no egg-masses detected; 1:⩽ 5; 2:⩽ 10; 3:11-100; 4:> 100 egg-masses.

Results

Results for individual accessions are given in Table 2, with means for ploidy types. Plants are grouped as zero, five or less, ten or less and more than ten egg-masses. Values are for percentage of each group in each accession. Number of plants, overall mean egg-masses, and mean log 10 (2+ egg-masses) and its standard deviation are given. The assumed diploid wild collection (Cg53-Cg57) had an overall variation around equal proportions of resistant and susceptible plants. The Cg74 Katambora stock was most resistant in terms of proportion of resistant plants (under five egg-masses/plant), lower proportion of plants with over 10 egg-masses and mean egg-masses/plant. The accessions Cg21 and Cg29 (Nzoia) also had high proportions of resistant plants, but a larger percentage of plants with more than 10 egg-masses than Katambora. The poorest diploid Cg2 had more susceptible than resistant plants, and a high mean egg-masses/plant, greater than that of some tetraploids. Cg12 (Katambora) had a reasonable proportion of resistant plants but more plants with over ten egg-masses than Cg74.

For tetraploids, overall mean egg-masses/plant was three times that of diploids, the lowest value 17.1 higher than the mean for diploids. In this test, Cg71 and Mbarara had no egg-mass free plants. The largest number of egg-masses (412) were found on an Elmba plant, and this cv. had the highest mean egg-masses/

plant (89.9), and lowest proportion of resisters of all accessions. The egg count for this accession was unexpectedly low. Of tetraploids Samford had the most egg-mass free plants and greatest proportion of resistant plants whilst Cg38 had the lowest mean egg-masses/plant.

Figure 1 shows the distribution of plants with various numbers of egg-masses plotted for diploid (and assumed diploid) and tetraploid types. As overall numbers of plants (2n, 248 and 4n, 245) were similar, actual plant values are given not percentage distribution.

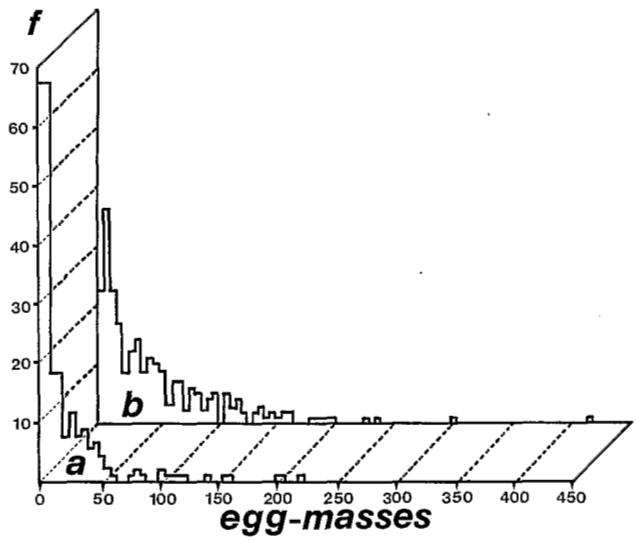

Fig. 1. Host status of *C. gayana* to *M. javanica* - distribution of egg-masses/plant for (a) diploid (n = 248), and (b) tetraploid (n = 245) accessions.

Table 3 gives the mean eggs/egg-mass, eggs/plant, mean per plant multiplication (mf = eggs per plant/inoculum), and summaries for ploidy. Numbers of eggs/egg-mass per variety range widely, 78 to 382. The mean values for diploids and tetraploids do not differ greatly, but their ranges overlap. The mean mf of 0.1 for Cg74 Katambora was the lowest recorded. Both Nzoia accessions, Cg21 and Cg29, also had low mf values, 0.2, whilst the poorest diploid Cg2 had an mf of 2.0. The overall diploid per plant mean mf value was 0.9 compared with 3.4 for tetraploids, none of which had an mf lower than 1.0. Amongst tetraploids Boma had the highest mf value 5.9, and Cg38 the lowest, 1.0. The low egg count of Elmba was reflected in its mid-range mf, 3.9.

There was no apparent relationship between root weight and degree of susceptibility.

Log 10 total eggs per accession is plotted against log 10 egg-masses in Fig. 2 and is broadly linear. Whilst mean eggs/egg-mass was similar for both ploidy levels,

Table 3

M. javanica egg production data on *C. gayana* accessions.

Accession	*eggs/plant* $\times 10^3$	*eggs/egg-mass*	*mean/plant mf**
Cg 2	7.6	210	2.0
Cg 12	1.8	126	0.5
Cg 21	0.6	85	0.2
Cg 29	0.8	271	0.2
Cg 52	4.0	243	1.1
Cg 53	6.8	338	1.8
Cg 54	4.2	213	1.1
Cg 55	3.7	136	1.0
Cg 56	5.9	341	1.6
Cg 57	1.2	78	0.3
Cg 74	0.5	205	0.1
Diploid	3.4	204	0.9
Tetraploid	12.5	273	3.4
Cg 34	12.4	378	3.4
Cg 38	3.8	221	1.0
Cg 40	11.2	260	3.0
Cg 71	15.6	181	4.2
Mbarara	14.0	303	3.8
Masaba	10.2	238	2.8
Elmba	14.6	156	3.9
Boma	22.0	382	5.9
Callide	15.0	305	4.0
Samford	8.4	310	2.3
Mt Makulu 56	11.2	270	3.0

*mf = eggs per plant/inoculum.

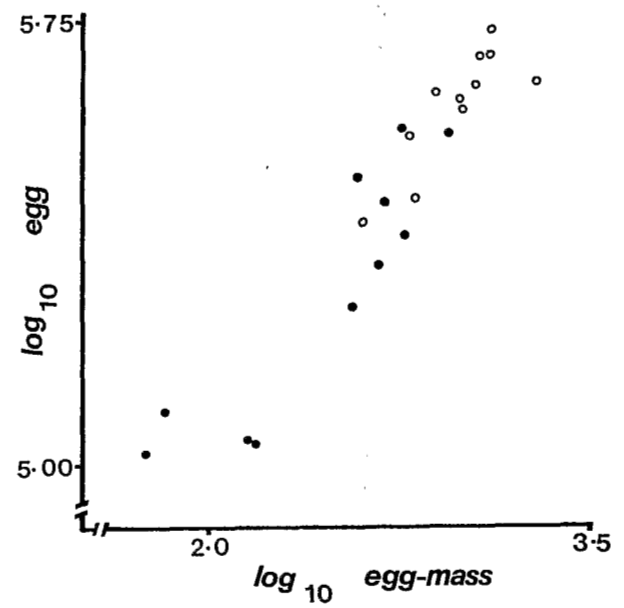

Fig. 2. Reproduction of *M. javanica* on *C. gayana* - log 10 total eggs (y) plotted against log 10 egg-masses (x) for diploid (●) and tetraploid (○).

Table 4

Distribution of classes of *M. javanica* egg-masses/plant of *C. gayana* cvs by simple scoring.

		*Classes**						
Cultivars	*n*	*0*	*1*	*2*	*3*	*4*	*R*	*S*
Mbarara	295	2.4	33.9	23.0	22.4	18.3	36	64
Elmba	250	0	13.6	34.8	28.4	23.2	14	86
Callide	212	3.8	34.0	27.8	27.8	6.6	38	62

*Number of egg-masses per class : 0 = none; 1 = 1-5; 2 = 6-10; 3 = 11-100; 4 > 100 egg-masses; R (resistant) < 5; S (susceptible) > 5.

diploids gravitate to the lower values in this graph and tetraploids tend to the upper values.

Table 4 shows the distribution of classes by the simple visual score of plants in the second test, with proportion of resistant (i.e. with 5 or less egg-masses) and susceptible plants. The results of the simpler scoring system agree only broadly with detailed count results, but are of separate larger samples.

Discussion

The most significant feature of the results from the comparative test is the range of host suitability found within both ploidy levels (Fig. 1). Clearly the distribution of egg-masses/plant is different in the two ploidy levels. Whilst diploids have more resistant plants than tetraploids the latter show not only more susceptibles but a greater degree of susceptibility. Distribution of egg-masses/plant within ploidy over this wider range of material from different sources is similar to that reported from restricted testing of diploid and tetraploid material (York, in press, *a*).

A similar linear relation of reproduction with numbers of egg-masses holds as found previously. Root weight at the end of the experiment was not related to numbers of egg-masses. Whilst eggs/egg-mass ranged widely, presence or absence of egg-masses should allow sufficient discrimination between susceptible and resistant plants for screening purposes. Although a less sensitive test, with more susceptibles slipping into the five class, when selecting only egg-mass free plants a simple visual assessment should suffice, regardless of plant size.

Great emphasis is not placed on the differences between accessions within ploidy as relatively few plants of each were examined. However some general trends in relation to source and history of material are worth comment.

Distribution of resistance in the South African material should be fairly representative of what is available in diploids in the wild state. Pioneer was selected to suit Australian conditions, reputedly arising from diploid material originating in South Africa (Barnard, 1972) : it falls within the range observed in this wild material. Nzoia (Cg29, and improved Nzoia Cg21) was a popular variety in Kenya (Bogdan, 1969). The slight disparity between these two accessions in numbers of zero rated plants may relate to the vegetative propagation of Cg29 for this test. Nzoia is very similar in appearance to the South African material (York, in press, *b*). Rhodes grass was first brought into cultivation in South Africa where diploids are more common (Rhind, 1977). In Kenya tetraploids are more prevalent. Possibly European farmers introduced Nzoia or its progenitor to Kenya (Bogdan, 1969). However the difference between Cg29 and Cg21 on the one hand and Cg53 to Cg57 on the other in host status to *M. javanica* is striking. This may represent an accidental selection especially as Nzoia had received some breeding attention. Nzoia could be a useful alternative to Katambora as a root-knot control pasture; but is more upright and less spreading than Katambora, thus less effective in suppressing weed invasion.

Whilst diploids are generally more resistant than tetraploids, the danger of relying on that generalisation is illustrated by the behaviour of Cg2. That tetraploids are generally more susceptible is evident; but also there is apparent scope for selection within these. Large numbers of plants must be screened as with the Zambian selection Mt. Makulu 56 (York, 1989) and perhaps more for some accessions than others; compare Samford and Mbara for example.

With a greater degree of susceptibility amongst tetraploids host suitability relationship trends are more difficult to detect. Samford derived from CPI16144 (Barnard, 1972) which was introduced to Australia via Sierre Leone originally from Kenya. Arising from survivors in Sierre Leone its genetic base may have been limited and these results may reflect an accidental selection towards greater resistance. CPI16144 originally was classed as diploid (Hutton, 1961); but later found to be tetraploid (Pritchard & Gould, 1964; Barnard, 1972). No accidental selection towards resistance occurred in the case of Elmba or Boma, selected respectively from Mbarara and Masaba (Boonman, 1978) on the basis of

earliness of flowering; nor in the case of Pioneer. The Zimbabwean strains of cv. Giant may vary in host suitability, cf. Cg71 and Cg40.

Comparison of the overall distribution of susceptibility (based on egg-masses) in the two ploidy states is more meaningful than attempting to draw too many variety comparisons or attaching too great a weight to those differences. Whilst different tests show general agreement, these accessions are mainly relatively unselected for agronomic characteristics and few have been stabilized. There has been no selection for root-knot resistance : Katambora was simply bulked from a collected sample (West, 1952) gauged relatively resistant (Martin, 1957) and found to give reasonable control (Shepherd, 1968).

The large range of numbers of egg-masses detected in both ploidy types indicates a polygenic or additive component for degree of susceptibility. The distributions are not normal so that resistance may be governed by one or few major genes. Partial dominance of major gene(s) for resistance could cause a range of response with a wider range occurring in tetraploids as seen here.

The nature of polyploidy in *Chloris gayana* has received little critical analysis. An early opinion (Moffet, 1944) was that morphological similarity indicated autoploidy. The diploids and tetraploids are very similar in appearance. Genetic analysis with a recessive genetic marker in tetraploids (Bogdan, 1963) indicated alloploidy, whilst recent cytological examination has suggested a degree of autoploidy (Nakagawa & Sato, 1986).

Progeny of selected material (2n and 4n) is being screened. Controlled paired crosses in different combinations of resistant and susceptible plants have been made. The progeny of these will be screened. Results will more clearly define the inheritance of resistance.

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REFERENCES

BARNARD, C. (1972). *Register of Australian herbage plant cultivars.* Div. Plant Industry, CSIRO, Australia : 98-102.

BOGDAN, A. V. (1963). *Chloris gayana* without anthocyanin colouration, *Heredity (London),* 18 : 364-368.

BOGDAN, A. V. (1969). Rhodes grass. *Herb. Abstr.,* 39 : 1-13.

BOONMAN, J. G. (1978). Rhodes grass breeding in Kenya. III. Seed and herbage yield in selections of four maturity classes based upon intravariety variation. *Euphytica,* 27 : 649-656.

HOLLBROOK, C. C. D., KNAUFT, D. A. & DICKSON, D. W. (1983). A technique for screening peanut for resistance to *Meloidogyne arenaria. Plant Dis.,* 57 : 1025-1028.

HUSSEY, R. S. & BARKER, K. R. (1973). A comparison of methods of collecting inocula for *Meloidogyne spp.,* including a new technique. *Pl. Dis. Reptr,* 57 : 1025-1028.

HUTTON, E. M. (1961). Intervariety variation in Rhodes grass (*Chloris gayana* Kunth). *J. Brit. Grassl. Soc.,* 16 : 23-29.

MARTIN, G. C. (1957). The common root-knot nematode. *Rhod. Farmer,* 27 : 24-26 [*11-1-57*].

MOFFET, A. A. (1944). Note on the cytology of Rhodes grass. *Rhod. J. Agric.,* 41 : 11-13.

NAKAGAWA, H., SATO, H. (1986). Cytological and morphological characteristics of the genus *Chloris. Proc. 15th Int. Grassland Congr. Kyoto, Japan, 1985* : 325-327.

PRITCHARD, A. J., GOULD, K. F. (1964). Chromosome numbers in some introduced and indigenous legumes and grasses. *CSIRO, Australia, Div. trop. Pastures, Techn. Pap. No. 2.*

VAN RENSBURG, H. J. (1969). Selection of productive strains of *Chloris gayana* in Zambia. *Rep. FAO OPEX Assignment, Min. rural Develop., Lusaka, Zambia.*

RHIND, J. M. L. C. (1977). A survey of mode of reproduction of certain grasses and legumes in South Africa. *Landbouuavorsing (S.A. Dept. Landboutegnise)* : 47.

SHEPHERD, J. A. (1968). A nematode survey of tobacco soils in Rhodesia and Zambia and the effects of grass-tobacco rotations on nematode populations. *Rhod. J. agric. Res.,* 6 : 19-26.

WEST, O. (1952). Promising new grasses for seeded pastures in Southern Rhodesia. *Rhod. Agric. J.,* 49 : 89-93.

YORK, P. A. (in press, *a*). Resistance to *Meloidogyne javanica* (root-knot nematode) in *Chloris gayana* (Rhodes grass). *Nematologica.*

YORK, P. A. (in press, *b*). Rhodes grass breeding. *Ann. Rep. Div. Livestock and Pastures, Dept. Res. and Specialist Services, Zimbabwe, 1987.*

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