Table 2 *Diosgenin content of seeds of C. speciosus*

Level of ploidy	Diosgenin content μ g in dry sample
Diploid	1101 6
Triploid	774.4
Tetraploid	773.4

stem and leaf of diploid, triploid and tetraploid races of *C. speciosus* at three different stages of growth revealed that the diosgenin content is higher at the flowering stage than vegetative and fruiting stages in almost all the parts of the plants. Diosgenin content estimated in rhizomes at the three different stages of growth also shows that the content of diosgenin is higher at the flowering stage. At the flowering stage only, the diosgenin present in the rhizome has been more than in the other parts such as root, stem and leaf. Moreover the variation in diosgenin content in root, stem and leaf at the different stage is not much of significance.

From the present investigation it is clear that the diosgenin content at the flowering stage is high and this supports the earlier findings of Sarin *et al*[5] and Gupta *et al*[6]. The diosgenin content goes up when rhizome sprouts with increasing tendency till flowering starts, reaches maximum when the flower buds appear, and after this the diosgenin content starts decreasing during the fruit formation. As a result of the foregoing observation, it can be concluded that the diosgenin synthesis starts soon after the sprouting and goes on increasing till it reaches maximum at the time of initiation of flowering. Later the biosynthesis of diosgenin probably stops in the rhizome when biodegradation starts because of the increased energy requirement of the plant during flowering and seed setting. This is the time when sugar moiety of the molecules is probably utilised for seed setting[7]. In the present study the maximum diosgenin content has been observed in the diploid race and the minimum diosgenin content has been found in the tetraploid. The triploid ranks next to diploid.

The diosgenin content of the seeds of the diploid, triploid and tetraploid is found to vary at Maduravoyal conditions. The content of diosgenin is maximum in the diploid followed by the triploid and tetraploid. Of the three races studied, the diploid race contains the maximum diosgenin which can be exploited commercially. Although the root, stem and leaf contain less diosgenin than the rhizome, the yield of these parts is more when compared to the rhizome which can also be exploited as a source of diosgenin. Extraction of diosgenin from the root, stem and leaf of *C. speciosus* (proved for the first time) is an additional source of diosgenin.

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INTERACTION BETWEEN *CERCOSPORIDIUM PERSONATUM*, *PUCCINIA ARACHIDIS* AND *ARACHIS HYPOGAEA*.

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GROUNDNUT rust caused by *Puccinia arachidis* Speg. and Tikka leaf spot caused by *Cercosporidium personatum* (B. & C.) Deighton (= *Cercospora personata*) are the most destructive of the foliar diseases of groundnut worldwide. While Tikka is known since long, rust appeared in India only in 1969 but quickly became pandemic throughout South India. Both appear on groundnut late in the growing season and persist till harvest. Under favourable conditions both the fungi develop together uninhibitedly. Plants are heavily defoliated to the extent that in some areas only withered stems are observed. The effect on yield appears more than additive[1,2].

Of the two pathogens *P. arachidis* is a biotroph (obligate parasite) while *C. personatum* begins its

parasitic life as a biotroph and turns necrotrophic later. Therefore the simultaneous occurrence of the two fungi with different modes of parasitism, has been a point of concern to the farmers and phytopathologists. The objective of the present investigation was to study the kind of interaction between the two groundnut pathogens on the one hand and between the pathogens and the host on the other.

C. personatum was grown on Czapek's agar (with 0.1 % yeast extract) medium for 10 days of which the last seven days were in light. (Sylvania 40 W-BLB lamps)[3]. Spores were collected by flooding the culture with sterile water and brushing the colony surfaces gently with camel hair brush to dislodge the conidia. Suspensions were filtered through cheese cloth layers to remove mycelial debris and spore clumps. Spore concentrations were adjusted to 50,000 spores per ml using a haemocytometer. *P. arachidis* was grown on detached groundnut leaves and the uredospores were collected on the 15th day after inoculation. Observing the usual precautions spore suspension was prepared and the concentration was adjusted to 50,000 spores per ml. Spores were allowed to germinate for 24 hr in petriplate pairs maintained in dark at 25 ± 1 C. At the end of the incubation period, the spores were examined under microscope and scored for germination. If the germ tube of *C. personatum* spore has exceeded at least twice the length of a conidium it was taken to indicate germination. For *P. arachidis* uredospores, differentiation of infection structures like germ tube and appresorium served the purpose. At least five replicates were maintained for each treatment and from each replicate at least a hundred spores were examined. Germination fluid was collected by centrifuging the suspensions containing germinating spores at 10,000 × g for 15 min. The supernatant was decanted and filter-sterilized for further use.

Fresh spore suspensions of *C. personatum* and *P. arachidis* were prepared as outlined above and were incubated for germination in cavities on microscopic slides. To each of the cavity with spores (0.2 ml) the germination fluid of *C. personatum* or *P. arachidis* or sterile distilled water (control) was added at the rate of 0.2 ml. Spores were examined after 24 hr and scored for germination. The results of two separate experiments are presented in tables 1 and 2.

Germination of *P. arachidis* uredospores was inhibited by its own germination fluid and by that of *C. personatum*. Inhibition was more severe with the former. Germination fluids of *P. arachidis* and some other rust fungi are known to contain self-inhibitors of germination *i.e.* methyl *cis* 3,4-dimethoxy cinnamate. Of the rust spores those of *P. arachidis* were more sensitive to the self-inhibitor[4], perhaps explaining the lower rate of germination compared to controls.

Table 1 *Effect of germination fluid of P. arachidis uredospores and of C. personatum conidia on germination of groundnut rust uredospores.*

	Germination of spores (%)	
	Expt. 1	Expt. 2
P. arachidis uredospores in water (control)	72	76
P. arachidis uredospores + germination fluid of *P. arachidis*	53	53
P. arachidis uredospores + germination fluid of *C. personatum*	54	46

Table 2 *Effect of germination fluid of C. personatum conidia and of P. arachidis Uredospores on germination of C. personatum spores.*

	Germination of spores (%)	
	Expt. 1	Expt. 2
C. personatum spores in water (Control)	62	64
C. personatum spores + germination fluid of *C. personatum*	64	64
C. personatum spores + germination fluid of *P. arachidis*	64	61

There is reason to suspect the presence of an inhibitory compound in the germination fluid of *C. personatum* which could inhibit the germination of rust urediospores to some extent but not autoinhibitory (table 2). How these inhibitors interplay on host leaf surfaces is left to conjuncture and needs further investigation.

Experiments were designed to see how the two fungi formed characteristic lesions on host leaves when inoculated together. Spore suspensions of the two fungi were prepared as outlined earlier and sprayed onto the lower surfaces of the groundnut leaves one after the other. Control leaves were sprayed with spores of *P. arachidis* or *C. personatum*. The inoculated leaves were floated on distilled water in petriplates and incubated at 30°C. The number of spots of each fungus in every leaf was counted on the

tenth day. Lesions of both fungi were almost equal in number whether inoculated together or separately. In the case of the former, though they occurred closeby they were clearly separated from each other by rings of green tissue. The kind of inhibition observed during germination was not evident either in number or form of lesions. The observations strongly suggest that the two fungi can co-exist on leaves and therefrom exert a synergistic effect on host plants.

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ASSOCIATION OF RUST RESISTANCE WITH NUMBER OF TANNIN SACS IN GROUNDNUT

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FOURTEEN rust resistant lines were jointly released by the International Crops Research Institute for Semi Arid Tropics, Hyderabad and the United States Department of Agriculture, Georgia, twelve of them belonging to the botanical type valencia and two to the virginia type[1-3]. Among the 21 published and several unpublished wild species of *Arachis*, all, except *A. monticola*, are either immune or highly resistant to the rust disease[4]. Earlier attempts to find out whether resistance to diseases has any anatomical basis were inconclusive[5]. During the course of our studies on rust, a detailed study was made on leaf anatomical features of several groundnut varieties and wild species of *Arachis*.

Thirteen varieties, reported as resistant to rust and one resistant to *Aspergillus flavus*, 5 popular varieties which are moderately to highly susceptible to rust, and 13 wild species of *Arachis* were examined for the number and size of the tannin sacs in paradermal sections. Paradermal sections were taken of fixed material of mature leaves cut at a thickness of 12 μ. The sections were stained with Delafelds haematoxylin. The number of tannin sacs was counted in 5 micro-

Table 1 *Number and diameter of tannin sacs in wild species and cultivated varieties*

Wile species/cultivated variety	Mean No. of tannin sacs (mm^{-2})	Mean size of the tannin sacs (μm)
Wild species		
1. *A. villosa* (Coimbatore)	101	26.4
2. *A. duranensis* (PI 219823)	121	23.6
3. *A correntina* (PI 331194)	77	24.4
4. *A. chacoense* (PI 276235)	42	25.3
5. *A. monticola* (Dharwar)	84	30.0
6. *A. sp.* (PI 10038)	114	25.6
7. *A. batizocoi* (PI 338312)	76	34.0
8. *A. sp.* (*A. prostrata*)	100	33.2
9. *A. hagenbeckii* (Coimbatore)	105	20.4
10. *A. glabrata* (Coimbatore)	94	24.8
11. *A. marginata* (Coimbatore)	65	26.8
12. *A. pusilla* (PI 338449)	87	23.6
Mean	89	26.5
Resistant groundnut varieties		
1. ICG 7881 PI 215696	36	28.4
2. ICG 7882 PI 314817	43	22.0
3. ICG 7883 PI 315608	47	28.8
4. ICG 7884 PI 341879	47	24.9
5. ICG 7885 PI 381622	48	23.7
6. ICG 7886 PI 390593	52	28.0
7. ICG 7887 PI 390595	40	29.8
8. ICG 7888 PI 393516	56	29.6
9. ICG 7895 PI 393643	42	27.6
10. ICG 7896 PI 393646	41	23.1
11. ICG 7887 PI 405132	53	26.5
12. ICG 7898 PI 407454	48	26.2
13. ICG — PI 259747	42	29.6
14. ICG — PI 337394	44	22.8
Mean	45	26.5
Popular varieties		
1. J-11	26	25.8
2. POL-2	24	27.8
3. DH-3-30	35	22.0
4. Robut 33-1	33	26.5
5. M-13	27	27.6
Mean	27	25.9