1981

Investigations of ryegrass toxicity.

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1981 - SUMMARY OF RESULTS OF FIELD EXPERIMENTS

INVESTIGATIONS OF RYEGRASS TOXICITY

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DEVELOPMENT OF TOXICITY

EXPERIMENTS: 81KA47, 81NA47, 81MO47

LOCATION: C. Butterworth, Katanning
D. Holmes, Katanning
A. Price, Corrigin
J. Follett, Wongan Hills

AIM: To monitor the amount of toxin present in galls from the time of first head emergence until maturation of the ryegrass pastures. To relate the onset and development of toxicity to plant growth stages and climatic conditions in the different regions affected by annual ryegrass toxicity. To relate the level of toxicity to pasture density and to the number of galls present in the pasture.

METHODS: At each experimental site, an area known to be affected with annual ryegrass toxicity was subdivided into four blocks each comprising 10 plots measuring 1 x 2 m. The sites were innoculated with 175 galls per sq.m. to help achieve a uniform heavy infection. Approximately 14 per cent of the galls were colonised by bacteria, the remainder contained nematodes.

At each site four plots, one from each block, were harvested at intervals of one week from head emergence until all plots were sampled.

The total number of ryegrass heads, their weight, and the number of galls containing either nematodes or bacteria were estimated for each sample, and the growth stage of the ryegrass was recorded.

Toxicity tests are presently being done on chemical extracts from the infected material collected at each sampling. A sensitive microassay based on the ability of the toxin to inhibit the growth of bacteria *in vitro* has been developed and is being used for these tests.

RESULTS: The following table shows the density of pasture and level of infection at each site during the season. These results show that there was very little difference in the density of pasture between the sites although there was more than a tenfold difference in the degree of infection. The proportion of galls colonised by bacteria are not given here because they were extremely variable and showed no consistent pattern either within or between sites.

COMMENTS: Variation in the levels of infection observed at different sites presumably reflect the level of innoculum in the pasture before the experimental inoculation. Hopefully, it should be possible to correlate these measurements with the levels of toxicity once the toxin assays are completed.

Density of pasture and the number of galls present in pasture at different sites during the season.

Date	Site			
	Katanning (Butterworth)	Katanning (Holmes)	Corrigin (Price)	Wongan Hills (Follett)
	Number of emerged heads/m^2			
September 9	-	-	-	121
14	-	-	-	189
22	11	23	87	272
29	133	62	171	425
October 6	373	447	392	699
13	807	614	609	901
20	789	689	665	647
27	729	740	665	729
November 3	608	638	725	584
10	738	570	637	515
17	688	513	548	-
24	639	534	678	-
Total from 10 m^2 sampled	5515	4830	5177	5082
	Number of galls/m^2			
September 9	-	-	-	752
14	-	-	-	871
22	28	560	91	1442
29	971	1528	17	2591
October 6	1400	7237	392	5522
13	1089	7273	1005	3692
20	1816	6338	333	4883
27	1931	8584	299	3170
November 3	4496	4849	399	2044
10	1365	5874	1083	1158
17	447	5158	466	-
24	1182	4167	305	-
Total from 10 m^2 sampled	14725	51568	4390	21126

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LIFE CYCLE OF THE NEMATODE, ANGUINA AGROSTIS IN RELATION TO PLANT GROWTH

EXPERIMENTS: 81KA47, 81NA47, 81MO47

LOCATION:
C. Butterworth, Katanning
D. Holmes, Katanning
A. Price, Corrigin
J. Follett, Wongan Hills

AIM: As part of the series of experiments done to monitor the amount of toxin present in pasture at a number of sites throughout the seasons, an apportunity was taken to follow the life cycle of Anguina agrostis in relation to plant growth.

METHODS: Galls containing nematodes were separated from the material collected to monitor the development of toxicity which was harvested at weekly intervals at each site. The experimental design and sampling procedures are given in the summary for "Development of toxicity". From 20 heads selected at random from each sampling, 10 galls were dissected and the numbers of adults, eggs and parasitic larvae were recorded. The numbers of eggs and parasitic larvae per female at each sampling were estimated from these records.

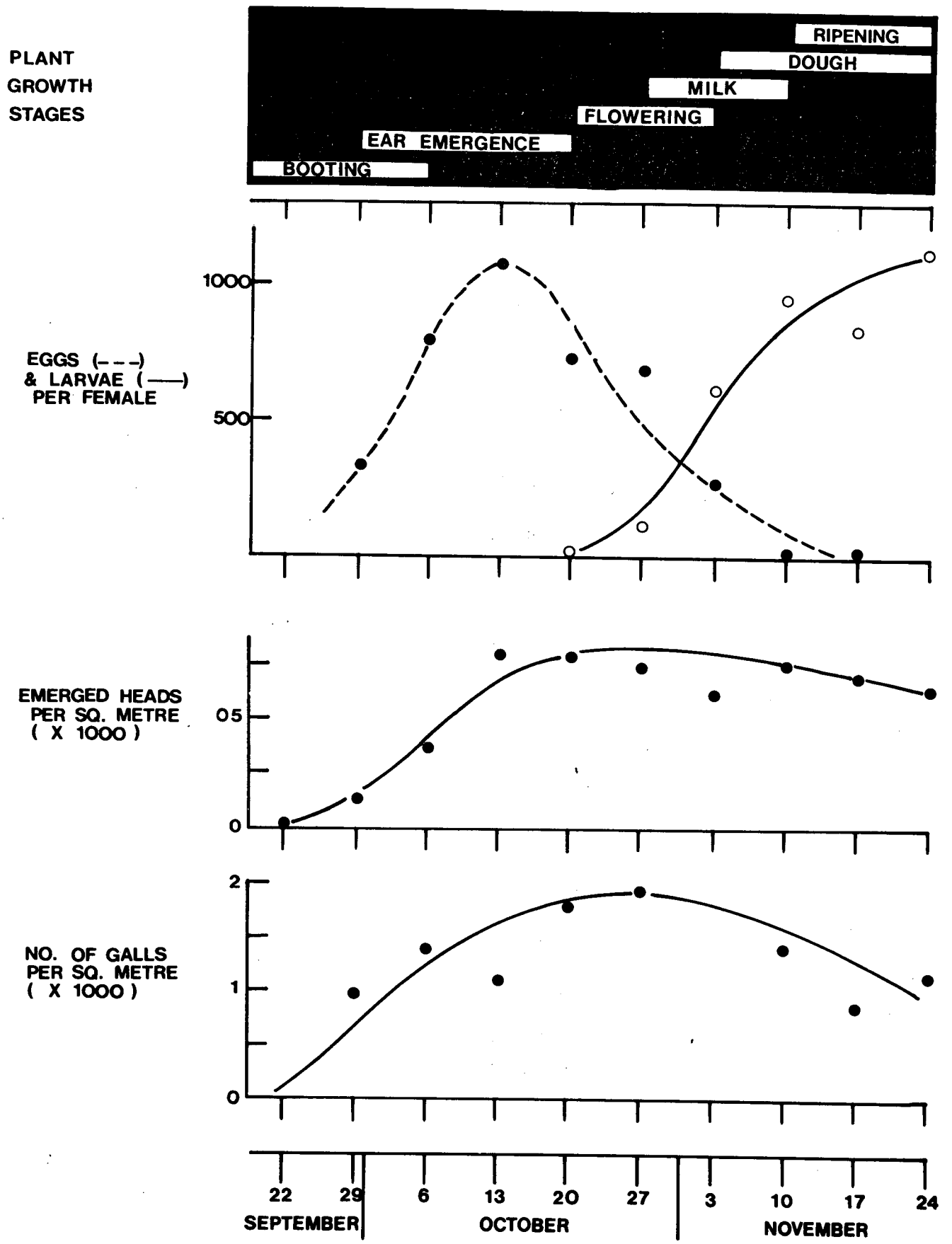
RESULTS: At each site egg laying commenced one to two weeks prior to ear emergence and continued for about five weeks. Hatching started just prior to and continued during flowering. By the end of flowering, almost all the eggs had hatched and the nematodes had developed into infective larvae.

The first figure shows the typical pattern of egg laying and of hatching in relation to plant development in the Katanning area in 1981. The second figure shows how different seasonal conditions influenced the rate of ryegrass development and how the life cycle of the nematode followed closely the plant development.

COMMENTS: This series of experiments shows that the life cycle of the nematode is synchronised with the growth and development of the ryegrass plant., This suggests that in any region and under different climatic conditions it should be possible to predict what stage of development the nematode is at by observing the plant growth stage.

Recognising that the infective larvae are the least vulnerable stage in the nematode life cycle it can be expected that control measures implemented before flowering will have the greatest chance of limiting the size of the nematode population carried over from one season to the next. This is clearly illustrated in the results of the spray top trial given in this report.

Embryonic and post embryonic development of Anguina agrostis in relation to plant growth and the seasonal patterns of head emergence and gall production



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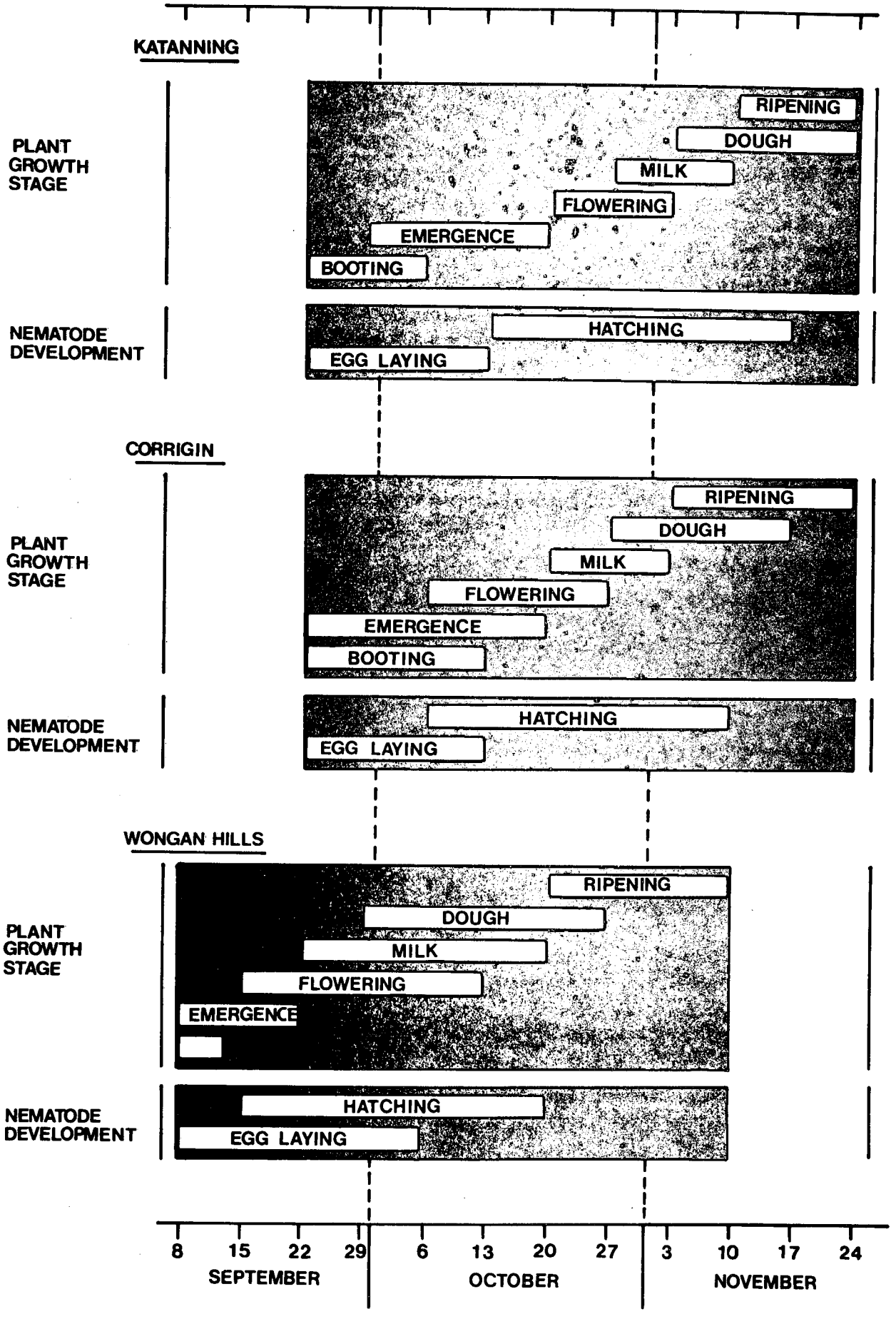
The effect of different seasonal conditions at Katanning, Corrigin and Wongan Hills on embryonic and post embryonic development of Anguina agrostis



EFFECT OF PASTURE "SPRAYTOPPING" ON GALL DEVELOPMENT

EXPERIMENTS: 81KA48, 81NA45, 81MO45

LOCATION: C. Butterworth, Katanning
A. Price, Corrigin
J. Follett, Wongan Hills

AIM: Earlier trials at Katanning (79KA14, 80KA26) showed that a single spray of paraquat in pasture just after ear emergence will reduce the number of galls produced. A proportional reduction in toxicity of the pasture is expected.

The trials reported here were done to:
(1) determine whether the timing of spraying in relation to plant development and climate was the same in different areas of the State and in different seasons; and
(2) evaluate the effect this treatment has on nematode populations.

TREATMENTS: Experimental sites with a history of toxicity were selected at Katanning, Corrigin and Wongan Hills. Each site was inoculated with 175 galls/m^2 to help achieve a uniform, heavy infection. Approximately 14 per cent of the galls were colonised by bacteria. The remainder contained nematodes. The experimental design at each site comprised four randomised blocks of 10 plots measuring 1 x 2 m. Paraquat was applied at a rate of 550 ml/ha to one plot per block at intervals of one week from September 23 until the ryegrass matured. Six sprays were applied in this instance.

METHODS: At maturity, plant tops were harvested from 1.0 m^2 quadrats per plot and threshed to collect galls. The number of galls containing nematodes and those colonised by bacteria were estimated per unit area and per 100 g threshed grain. The galls containing nematodes were then dissected and the contents examined to determine the effect of spraytopping on the development of nematodes. The numbers of eggs, preparasitic larvae and of parasitic nematode larvae per gall, were determined from each spray treatment.

RESULTS: The inoculum was applied too late at the Wongan Hills site to adequately infect the plots; consequently, this site was abandoned. The following table summarises the results for Katanning and Corrigin. They show that spraying between September 30 and October 7 at Katanning and at Corrigin reduced the numbers of galls produced in the pasture by up to 95 per cent and 85 per cent, respectively. Furthermore, they show that the spray has a marked effect on the viability of nematodes within those galls that do develop. The number of infective larvae per gall was reduced by 78 per cent at Katanning and by 54 per cent at Corrigin. Consequently, as the table shows, the number of viable nematodes per unit area was reduced by 97 per cent at Katanning and by 89 per cent at Corrigin.

The optimum growth stage to spray corresponded to the first week of ear emergence.

COMMENTS: As was shown in earlier experiments, the time to Spraytop pasture is very critical. A maximum reduction in the number of galls developing and hence in the production of toxin is achieved by spraying between one and two weeks after the first ryegrass heads emerge from the boot. This has proved consistent at different sites for the last three seasons. If sprayed earlier gall production is still reduced although not to the same extent, while later sprays become successively less effective.

The degree of control achieved by spraying at the correct time should normally prevent pasture from becoming toxic to grazing animals. However, it should be borne in mind that control is not complete and hence sufficient galls could still develop in very heavily infected paddocks to affect grazing animals.

A further benefit from this treatment is the effect on nematode survival. Apart from the direct reduction in the number of galls produced, there is also a reduction in the viability of larvae in the galls that do develop. The numbers of viable larvae that survived in spraytopped areas were reduced by as much as 97 per cent. Consequently, there would be minimal infection or risk of toxic pasture in the seasons immediately following spraytopping.

The effect of spraytopping on the production of galls, on the viability of nematodes and on the survival of seeds at Katanning and Corrigin

Date of spray	Number of galls produced per m^2	Number of viable nematodes per gall	Estimated number of viable nematodes per m^2	Weight of ryegrass seed per m^2
		Katanning		
September 23	28^{b}	758^{bc}	18950	3.9^{bc}
30	5^{b}	457^{d}	2057	2.5^{c}
October 7	27^{b}	273^{d}	4505	2.4^{c}
14	91^{a}	343^{d}	24696	3.7^{bc}
21	102^{a}	492^{cd}	43640	5.3^{b}
28	87^{a}	12^{b}	64554	4.8^{b}
Nil	91^{a}	1255^{a}	91239	8.8^{a}
		Corrigin		
September 23	5^{b}	193^{c}	781	3.6^{c}
30	5^{b}	624^{ab}	1923	5.6^{bc}
October 7	8^{b}	353^{bc}	2142	6.4^{bc}
14	29^{ab}	397^{bc}	8200	7.4^{bc}
21	58^{a}	456^{abc}	19243	10.1^{b}
28	51^{a}	592^{ab}	20838	9.4^{b}
Nil	34^{ab}	766^{a}	17235	18.0^{a}

Values followed by the same suffix are not significantly different ($P < 0.05$)

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EMERGENCE OF NEMATODE LARVAE FROM GALLS

EXPERIMENTS: 81KA50, 81NA46, 81MO46

LOCATION: C. Butterworth, Katanning
A. Price, Corrigin
J. Follett, Wongan Hills

AIM: To study the emergence of larvae of Anguina agrostis from galls at three different regions in the State and evaluate any differences in the patterns of emergence in relation to any variations in plant growth and climate.

METHODS: Ten sampling trays, each comprising a 2 cm length of 8 cm PVC pipe closed off at one end with nylon mesh (1 mm aperture) and containing 10 nematode galls, were placed in the field at each site prior to opening rains and were lightly covered with soil.

At each site, one tray was collected at weekly intervals commencing six weeks after the opening rains. The numbers of larvae dissected from the galls in each tray were counted. The growth stage of ryegrass on the experimental plots was recorded at each sampling.

RESULTS: The following table gives the average number of larvae remaining in the galls from each sampling.

COMMENTS: Unfortunately, the nematode counts in these experiments may only be indicative of the patterns of emergence of nematodes in the different areas because of the difficulty in recognising the 10 original galls added to each sampling tray. Invariably, more than 10 galls were recovered from each tray when sampled, indicating some were present in the soil used to cover the trays. Because the age of these galls was unknown, their larval content biassed the final count. This experiment will be repeated in 1982.

Average number of larvae of Anguina agrostis present per gall during the growing season

Date		Site		
		Wongan Hills	Corrigin	Katanning
July	22	1680	1955	1025
	29	1817	1535	1215
August	4	1156	1451	1022
	11	1434	1081	1407
	18	1754	847	542
	26	1983	1389	239
September	2	1503	1868	519
	9	644	485	159
	16	400	978	575
	23	445	-	168

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