

CHAPTER 3

RATE OF DEVELOPMENT OF *S. VAGANS* AT CONSTANT AND FLUCTUATING TEMPERATURES

3.1 ABSTRACT

The development of the ladybird *S. vagans* was assessed at seven constant (from 10 to 35°C) and variable (12.7-32.1°C) temperatures. Immature stages were reared in modified sealable petri dishes and were supplied with excess prey of two-spotted mite, *T. urticae*. Observations were made 12 hourly. Larvae developed through 4 larval instars and a pre-pupal stage. The mean development times from egg to adult were 65.2 ± 0.8 , 33.2 ± 0.6 , 18.2 ± 0.5 , 13.1 ± 0.4 , and 9.2 ± 0.3 days at constant temperatures of 12, 15, 20, 25, and 30°C respectively, while 15.4 ± 0.3 days was calculated for fluctuating temperatures (varying from 12.7 to 32.1°C). Developmental rate was calculated at each temperature for each stage. There was a strong positive correlation ($r = 0.99$) between temperature and rate of development. The lower development threshold temperature for egg, 1st, 2nd, 3rd, 4th larval instars, pupa and all immature stages combined was 10.1, 9.5, 9.5, 9.1, 8.2, 8.0, and 9.1°C respectively. Degree day (DD) accumulation was also calculated for each stage as well as for all stages combined. It was estimated to be between 189.2 ± 4.8 - 207.8 ± 6.9 from 12 - 30°C constant and 189.1 ± 5.0 at fluctuating temperatures (12.7-32.1°C) respectively for immature stages to complete their development.

Average egg incubation period decreased from 16.5 ± 0.84 to 2.18 ± 0.24 days with increasing temperatures from 12 to 30°C respectively and was 4.1 ± 0.5 at fluctuating

temperatures (12.7-32.1°C). No egg hatch was observed at 10° and 35°C. Eggs appeared to develop normally at 35°C, but larvae were unable to emerge. While eggs did not develop at 10°C, they were able to survive a long period of exposure to this temperature. Of approximately 200 eggs that were placed at a constant temperature of 10°C for 60 days then exposed to $\geq 15^\circ\text{C}$, more than 120 eggs hatched.

Total mortality of immature stages was 28.2, 13.9 and 21.9 % at 12 25 and 30°C respectively. Mortality was highest in the egg stage and 1st larval instar, and lowest in the 4th larval instar at a constant temperature of 12°C. No mortality was observed in 4th instar, pre-pupal and pupal stages at any other temperatures. Diapause was not observed in any stage in any treatment.

3.2 INTRODUCTION

Temperature and relative humidity are probably the most important physical environmental factors influencing rate of development and survival of living organisms. Degree day (DD) modelling is used to predict developmental events for plants, insects and other poikilothermic organisms (Arnold 1959; Higley *et al.* 1986). A degree day thermal scale is more accurate in predicting events than a chronological scale.

Rate of development and DD models have been developed for a number of insects: e.g., corn leaf aphid, *Rhopalosiphum maidis* (Elliott *et al.* 1988), navel orange worm *Amyelois transitella* (Sanderson *et al.* 1989), Mexican bean beetle *Epilachna varivestis* (Fan *et al.* 1992), and the coccinellids *Hippodamia sinuata* (Michels & Behle 1991), and *Stethorus bifidus* (Peterson *et al.* 1994). No data have been published on the degree day requirement development in the genus *Stethorus*, except for *S. bifidus*.

To understand the influence of biotic factors on development of *S. vagans* experiments were conducted at constant as well as fluctuating temperatures, to assess its effect on the development of immature stages. The goal was to obtain developmental data over a wide range of constant and fluctuating temperatures, which could be used to calculate the lower developmental threshold temperatures and to construct a degree day model for all life stages of *S. vagans*. This study was undertaken to also assist interpretation and prediction of seasonal development of *S. vagans* in the field.

3.3 MATERIALS AND METHODS

3.3.1 Field Collection and Stock Colony

Adult *S. vagans* were collected from the field on potted French bean plants (*P. vulgaris* cv Redland Pioneer) infested with two-spotted mites *T. urticae*. Infested bean plants were transferred to 15cm diameter pots from the glasshouse (Chapter 2) and all potted plants in the field were replaced after two weeks. These plants were examined every two days for beetle collection and were watered every day. Field collected beetles were brought to the laboratory and identified to species, because *Stethorus* species other than *S. vagans* were also present in the field. A colony was established from field collected *S. vagans* which was maintained at a constant temperature 25°C with a photoperiod of 16:8 (L: D) and relative humidity 46-75 % in a controlled temperature cabinet in the laboratory. All collections and investigations were carried out at the Centre for Horticulture and Plant Sciences, University of Western Sydney, Hawkesbury, Richmond (33° 36'S, 150° 44'E), New South Wales.

3.3.2 Development at Constant Temperatures

Beetles were randomly selected from the mass colony and paired on 2.5 cm leaf discs infested with *T. urticae* over moist sponge in 5cm diameter modified sealable petri dishes (Chapter 2). These dishes were maintained in modified large plastic containers with dimensions of 35 x 30 x 6 cm. These were placed in different illuminated incubators (122.0 x 52.5 x 43.0 cm) at seven constant temperatures (i.e. 10, 12, 15, 20, 25, 30, and 35°C) in the laboratory (Chapter 2). Eggs of *S. vagans* were collected from the above constant temperatures every 12 hours from the paired beetles and placed singly on a 4 cm diameter dry filter paper in modified petri dishes. Every egg was placed at the same temperature from which it was collected. A cohort of 50 eggs (replicates) per temperature treatment was

allocated to each constant temperature. The photoperiod for all temperatures was 16: 8 (L: D) hours and relative humidity ranged from 44 to 66 %. Separate experiments for higher temperatures (30 and 35°C) were conducted at higher relative humidity (70-85%), because no egg hatching was observed at this lower relative humidity. All dishes were monitored at 12 hourly intervals to assess egg hatching, larval moulting and survival. The presence of exuviae in conjunction with head capsule measurements (Chapter 4) was used to determine development to various larval stages. Developmental changes were recorded, and unhatched eggs or dead individuals were removed. These immature stages were supplied daily with excess prey mites obtained by brushing infested bean leaves with a mite brushing machine (Chapter 2).

3.3.3 Development at Fluctuating Temperatures

A separate experiment was also conducted using fluctuating temperatures, to compare rate of *S. vagans* development with that recorded at constant temperatures.

All adults were randomly selected from the laboratory-established colony and paired over infested bean leaf discs in the modified petri dishes. As oviposition was observed, leaf discs containing an egg were carefully cut out and placed in similar petri dishes as previously described for other studies. These dishes were maintained in two uncovered plastic containers on a table at ambient temperature and relative humidity in a non-climate controlled laboratory. Larval instars were supplied daily with all stages of *T. urticae* mite as described in section 3.3.2. Temperature and humidity was constantly logged hourly using data loggers (Tini tags[®], Hastings Data Loggers, Kempsey, NSW Australia), which were placed beside the petri dishes in the containers. The insect developmental data were recorded at ambient temperatures at 12 hourly intervals as occurred for constant temperatures.

3.3.4 Mortality

During the life cycle study any dead individuals observed were recorded and immediately removed while any immature stage killed accidentally during observation of feeding was removed and excluded from the data.

3.3.5 Data Analysis

The duration of development for each individual was recorded in hours and converted into days for analysis. Standard errors were calculated for all life stages using Excel 5.0 (Microsoft Office, SYBEX Inc., 2021 Challenger Drive, Alameda, CA 94501, USA). The effect of temperatures on the developmental rate of stage specific and combine stages were calculated by analysis of variance (ANOVA) using CoStat (CoHort Software P.O.Box 19272 Minneapolis, MN 55419, USA).

Developmental time of each life stage and all stages combined was expressed as the reciprocal of their duration (as proportion /day). The relationships between temperature and developmental rate were estimated by linear regression analysis using Origin 4.1 (Software for Technical Graphics and Data Analysis for Windows. Microcal Software. Inc., Northampton, MA 01063, USA). The lower developmental threshold temperatures for each specific stage as well as for all stages combined were derived from the regression equation, $y = a + bx$, where as “y” is the developmental rate (expressed as 1/days) at temperature “x”; and “a” and “b” are estimates of the “y” intercept and slope respectively (Sokal & Rohlf 1995).

Degree-days (DD) were computed for development of each life stage and total stages using the method outlined by Price (1997):

$DD = D (T-t)$, where

DD = Degree-days needed for development at a specific temperature.

D = Mean numbers of days required for development at a certain temperature.

T = Temperature at which the development was observed.

t = Minimum threshold temperature for development.

The mean DD required for development of each life stage was obtained by averaging its DD associated with all temperature regimes separately for eggs, larval instars and pupae. The mean DD for total development (egg-adult) used data obtained for all stages combined.

3.4 RESULTS

3.4.1 Developmental Time

The rate of development of all immature stages and total stages of *S. vagans* was inversely related to temperature, within the range tested. The mean total development times from egg to adult emergence were 65.2 ± 0.8 , 33.2 ± 0.6 , 18.2 ± 0.5 , 13.1 ± 0.3 , and 9.2 ± 0.3 days at constant temperatures of 12, 15, 20, 25, and 30°C respectively (Table 3.1).

The mean egg developmental period for eggs varied from 16.5 ± 0.9 days at 12°C to 2.2 ± 0.3 days at 30°C, although in the latter case eggs only hatched at the higher relative humidity tested (70-85%). No eggs hatched at either humidity level at 10°C and 35°C. Eggs appeared to develop normally at 35°C at the higher relative humidity until just prior to hatching, but the larvae failed to emerge. At 10°C, eggs survived for a long period without developing. Of 200 eggs that were initially exposed to 10°C for 60 days, none showed signs of development. However more than 120 of these eggs hatched within the normal development period when they were subsequently exposed to temperatures of 15 and 20°C.

The duration of the four larval instars were 7.8 ± 0.6 , 7.7 ± 0.7 , 8.0 ± 0.9 and 9.13 ± 0.6 days at 12°C, and 0.97 ± 0.3 , 0.96 ± 0.3 , 1.04 ± 0.3 , 1.2 ± 0.3 days at 30°C, respectively. The developmental time for the pupa was 16.3 ± 0.5 , 9.5 ± 0.4 , 4.4 ± 0.5 , 3.4 ± 0.4 , 2.8 ± 0.3 days at 12, 15, 20, 25, and 30°C, respectively (Table 3.1). There were significant differences in all immature stages combined and for stage specific development times at different temperatures ($0.0001 < p < 0.0008$) (Table 3.3).

The mean developmental time of all stages combined was 15.4 ± 0.2 days at fluctuating temperatures (from 12.7 to 32.1°C, and computed to be an average of 21.4°C). Mean development times for eggs, 1st, 2nd, 3rd, 4th larval instars and pupae were 4.1 ± 0.2 , 1.6 ± 0.3 , 1.7 ± 0.4 , 1.6 ± 0.3 , 1.7 ± 0.4 and 4.7 ± 0.4 days respectively at the above temperature.

3.4.2 Developmental Rate

The reciprocals of mean development time in days at both constant and fluctuating temperatures were calculated as a percentage of developmental per day (Table 3.2). The development rate for each stage and for all stages combined increased as the temperature increased. The development rate of the egg stage increased from 6.1% per day at 12°C to 46% per day at 30°C. The daily rate of development recorded for all four larval instars combined was 18 % per day at 12°C and 96 % per day at 30°C, and was 6.2% and 36% per day for the pupal stage at the same temperatures respectively. The daily rate of development for eggs, larval and pupal stages was 24.6, 60.1, 21.5 % respectively at fluctuating temperatures. The mean daily development rates of all immature stages at constant temperatures of 12 and 30°C and for fluctuating temperatures were 6%, 36% and 21% respectively.

The figures for mean development rate were plotted against temperature for each stage and for all stages combined to show the effect of temperature (Figs 3.1-3.4). Based on the linear regression equations the lower developmental threshold temperatures were estimated to be 10.2, 9.5, 9.5, 9.1, 8.2, and 8.0°C for egg, 1st, 2nd, 3rd, 4th instars, and pupal stages respectively. The mean lower developmental threshold temperatures for all stages combined was 9.1°C (Table 3.3).

The linear regression statistic (intercept and slope) described the relationship between the developmental rate (y) and temperature (x) for each stage and for all stages combined in *S. vagans*. The correlation coefficient (r) for each stage and all stages combined was very high (from 0.98 to 0.99) indicating a good fit of data to the linear degree-day model within the temperature range of 12 to 30°C (Table 3.4 and Fig. 3.1-3.4). There were no significant differences ($\alpha = 0.05$) among immature stages at the lower threshold as the 95% confidence intervals overlapped broadly. Therefore the lower threshold temperature for all stages combined was used to determine the number of degree-days required to complete development for each stage and for all stages combined.

3.4.3 Degree Day (DD)

Degree-day (DD) requirements (Table 3.4) were calculated for each stage of *S. vagans* and for all immature stages combined from the developmental data and the mean threshold temperature (9.1°C) (Section 3.4.2) (Table 3.3). Total mean DD estimated for development from egg to adult were 189.2 ± 4.8 , 195.8 ± 7.4 , 198.4 ± 5.6 , 207.8 ± 6.9 and $191.7.1 \pm 5.9$ at constant temperatures of 12, 15, 20, 25 and 30°C respectively, while it was calculated to be 189.1 ± 5.0 DD under fluctuating temperatures. The DD computed for eggs was 47.9, 58.5, 56.4, 61.1, and 45.6 at 12, 15, 20, 25, and 30°C, and 49.9 at fluctuating temperatures respectively. The DD calculated for the four larval instars ranged between 19.5 to 26.5 per instar over the range of constant and fluctuating temperatures, while for the pupal stage it varied from 47.2 to 58.1 DD at the same temperatures.

3.4.4 Mortality

Mortality throughout the developmental period for all the immature stages combined was highest (34.9%) at 12°C, followed by 15°C (22.2%) and 30°C (21.9%), while the lowest mortality (13.9%) occurred at optimum temperatures (20-25°C). Egg mortality was 9.3, 6.7, 5.6, 5.6, and 8.6% at 12, 15, 20, 25, and 30°C respectively. Larval mortality was highest during the 1st instar, with 12.8% and 12.5% at 12°C and 30°C respectively, but lower at other temperatures, i.e. 9.5, 5.9, and 5.9% at 15, 20, and 25°C respectively. Mortality for the 2nd larval instars ranged from 3.1% at 25°C to 8.8% at 12°C and for 3rd instars from 6.5% at 12°C to 3.8% at 30°C. In the 4th larval instar, mortality was only observed in the 12°C treatment (3.5%). No mortality was observed for pre-pupal or pupal stages in any temperature treatment (Table 4.6).

For fluctuating temperature (12.7-32.1°C) total mortality (ie. all immature stages combined) was 5.9%, which was significant lower than that for the same parameter at constant temperatures. Mortality was highest (5.6%) at the egg stage, followed by the 1st and 2nd instars (3.0% each). No mortality was observed in any other stage.

Table 3.1 Development time (days) of immature stages of *S. vagans* at constant and fluctuating temperatures.

Stage	Constant Temperature					Fluctuating Temperature
	12°C	15°C	20°C	25°C	30°C	
Egg	16.5 ± 0.8	9.9 ± 0.7	5.2 ± 0.5	3.8 ± 0.4	2.2 ± 0.3	21.4°C (12.7-32.1°C) 4.1 ± 0.3
1 st instar larva	7.8 ± 0.6	3.4 ± 0.7	2.2 ± 0.6	1.3 ± 0.4	1.0 ± 0.3	1.6 ± 0.3
2 nd instar larva	7.6 ± 0.7	3.3 ± 0.5	2.1 ± 0.6	1.4 ± 0.4	1.0 ± 0.3	1.7 ± 0.4
3 rd instar larva	8.0 ± 0.9	3.5 ± 0.6	2.2 ± 0.6	1.4 ± 0.4	1.0 ± 0.3	1.6 ± 0.3
4 th instar larva	9.1 ± 0.6	3.6 ± 0.5	2.2 ± 0.7	1.6 ± 0.4	1.2 ± 0.3	1.7 ± 0.3
Pupa	16.26 ± 0.5	9.5 ± 0.5	4.42 ± 0.5	3.41 ± 0.4	2.78 ± 0.3	4.65 ± 0.4
All stages combined	65.24 ± 2.3	33.18 ± 0.6	18.2 ± 0.5	13.07 ± 0.4	9.17 ± 0.3	15.37 ± 0.3

Table 3.2 Rate of development of immature stages of *S. vagans* (proportion /day).

Stage	Constant Temperature					Fluctuating Temperature
	12°C	15°C	20°C	25°C	30°C	21.4°C (12.7-32.1°C)
Egg	0.061	0.100	0.193	0.260	0.459	0.246
1 st instar	0.129	0.298	0.463	0.752	1.031	0.600
2 nd instar	0.132	0.303	0.474	0.709	1.041	0.559
3 rd instar	0.125	0.286	0.463	0.694	0.962	0.617
4 th instar	0.110	0.277	0.459	0.610	0.806	0.578
Pupa	0.062	0.105	0.226	0.293	0.360	0.215
All stages	0.015	0.030	0.055	0.077	0.109	0.065

Table 3.3 Regression of rate of development (1/y) with calculated values of correlation coefficient (r), probability (p) and minimum threshold development temperature (t) at constant temperatures.

Stage	Regression equations	r	P	t (°C)
Egg	$Y = - 0.21223 + 0.02092 x$	0.98	0.0006**	10.14
1st Instar	$Y = - 0.46514 + 0.04898 x$	0.99	0.0003***	9.50
2nd Instar	$Y = - 0.46034 + 0.04855 x$	0.98	0.0006**	9.48
3rd Instar	$Y = - 0.4076 + 0.04478 x$	0.99	0.0001***	9.10
4th Instar	$Y = - 0.30784 + 0.03735 x$	0.99	0.0003***	8.24
Pupa	$Y = - 0.13447 + 0.01689 x$	0.98	0.0008**	7.96
All stages	$Y = - 0.04655 + 0.00509 x$	0.99	0.0001***	9.07

** Significant at $p \leq 0.05$ *** Significant at $p \leq 0.01$

Table 3.4 Degree-days (DD) needed for development of immature stages of *S. vagans* at constant and variable temperatures.

Stage	Constant Temperature					Fluctuating Temperature
	12°C	15°C	20°C	25°C	30°C	
Egg	47.91	58.47	56.35	61.06	45.56	21.4°C (12.7-32.1°C) 49.94
1 st instar larva	22.53	19.82	23.54	21.15	20.27	20.17
2 nd instar larva	21.92	19.47	23.00	22.42	20.06	20.54
3 rd instar larva	23.20	20.65	23.54	22.90	21.74	19.93
4 th instar larva	26.48	21.30	23.76	26.08	25.92	21.28
Pupa	47.15	56.05	48.18	54.22	58.10	57.20
All stages	189.94	195.76	198.38	207.81	191.65	189.05

Table 3.5 Mortality (%) of life stages of *S. vagans* at different temperatures.

Stage	Constant Temperature												Fluctuating Temp.			
	12°C			15°C			20°C			25°C			30°C		12.7-32.1°C	
	Mortality %	Survive n	Mortality %	Survive n	Mortality %	Survive n	Mortality %	Survive n	Mortality %	Survive n	Mortality %	Survive n	Mortality %	Survive n	Mortality %	Survive n
Egg	9.30	39	6.67	42	5.56	51	5.56	41	8.57	32	5.56	34	5.56	32	5.56	34
1 st instar	12.82	34	9.52	38	5.88	48	5.88	38	12.5	28	5.88	33	3.03	28	3.03	33
2 nd instar	8.82	31	5.26	36	4.17	46	3.13	34	7.14	26	3.13	32	3.13	26	3.13	32
3 rd instar	6.45	29	2.78	35	2.17	45	0.00	32	3.8	25	0.00	32	0.00	25	0.00	32
4 th instar	3.45	28	0.00	35	0.00	45	0.00	31	0.00	25	0.00	32	0.00	25	0.00	32
Total Larva (1-4)	28.20	28	16.67	35	11.76	45	8.82	31	21.88	25	5.88	32	5.88	25	5.88	32
Pre-pupa	0.00	28	0.00	35	0.00	45	0.00	31	0.00	25	0.00	32	0.00	25	0.00	32
Pupa	0.00	28	0.00	35	0.00	45	0.00	31	0.00	25	0.00	32	0.00	25	0.00	32
Adult	0.00	28	0.00	35	0.00	45	0.00	31	0.00	25	0.00	32	0.00	25	0.00	32
Total (egg-adult)	34.88	28	22.22	35	16.67	45	13.89	31	21.88	25	5.88	32	5.88	25	5.88	32

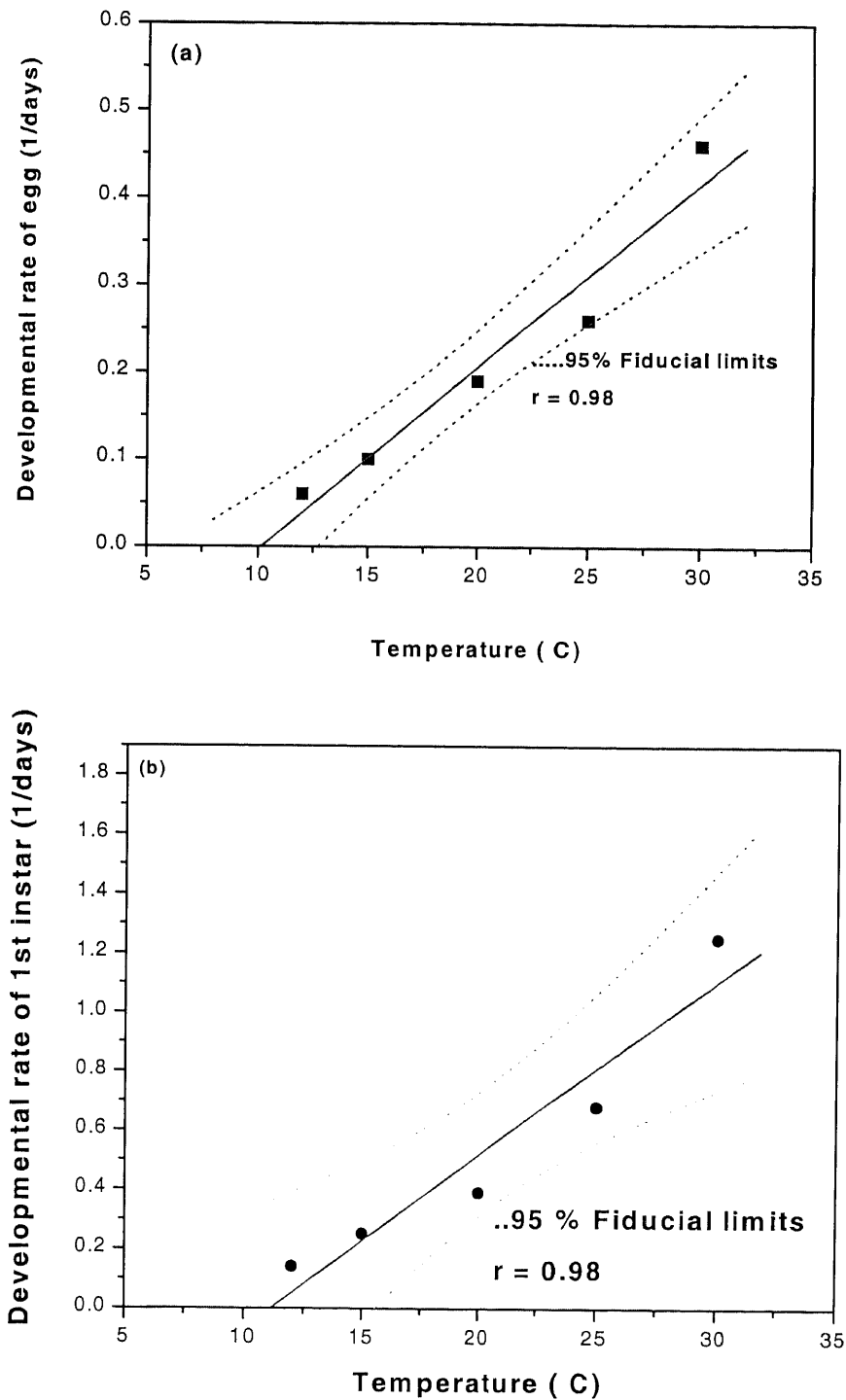


Fig. 3.1 Effect of various constant temperatures on the rate of development of immature stages of *S. vagans*: (a) Egg, (b) 1st larval instar.

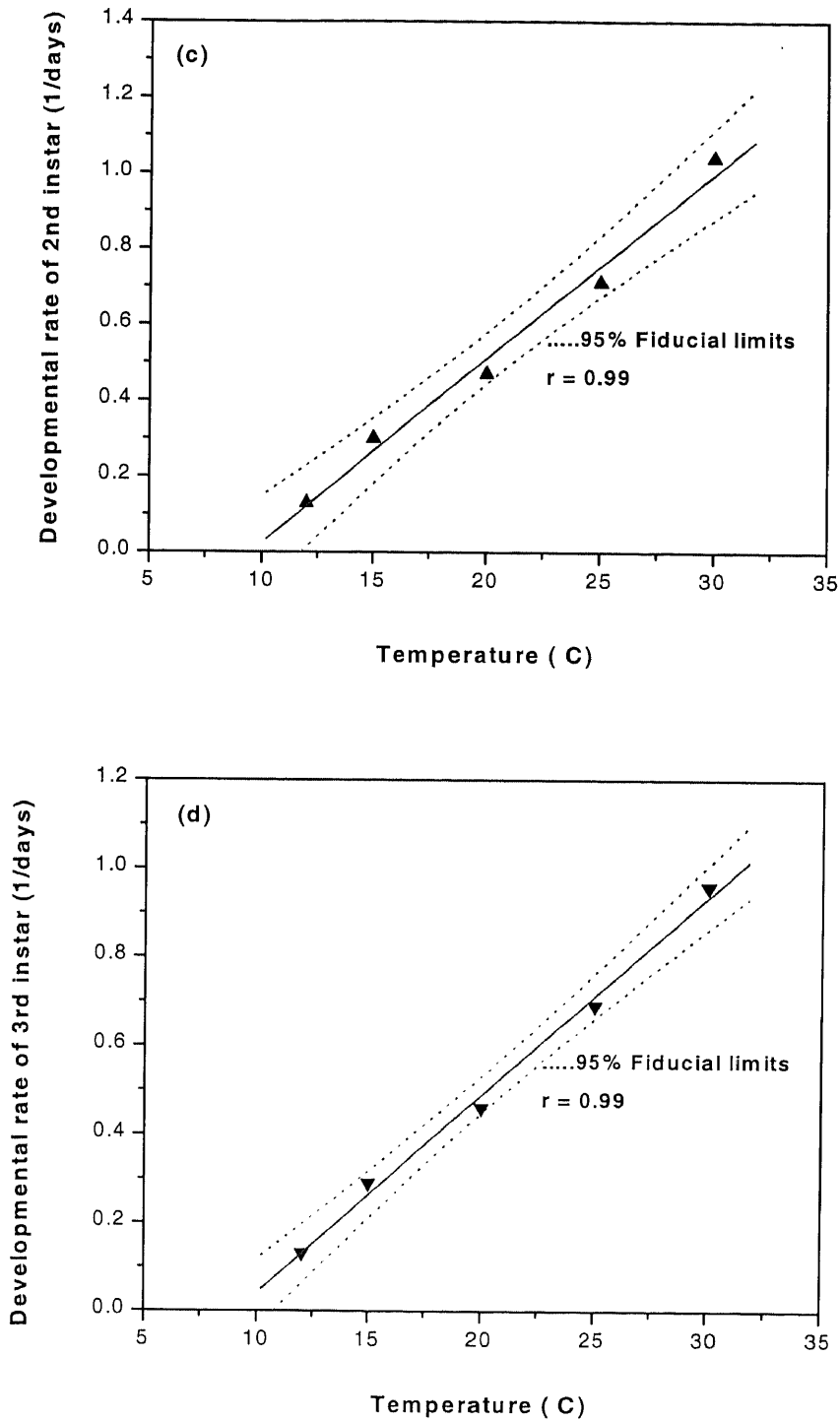


Fig. 3.1 Effect of various constant temperatures on the rate of development of immature stages of *S. vagans*: (c) 2nd larval instar, (d) 3rd larval instar.

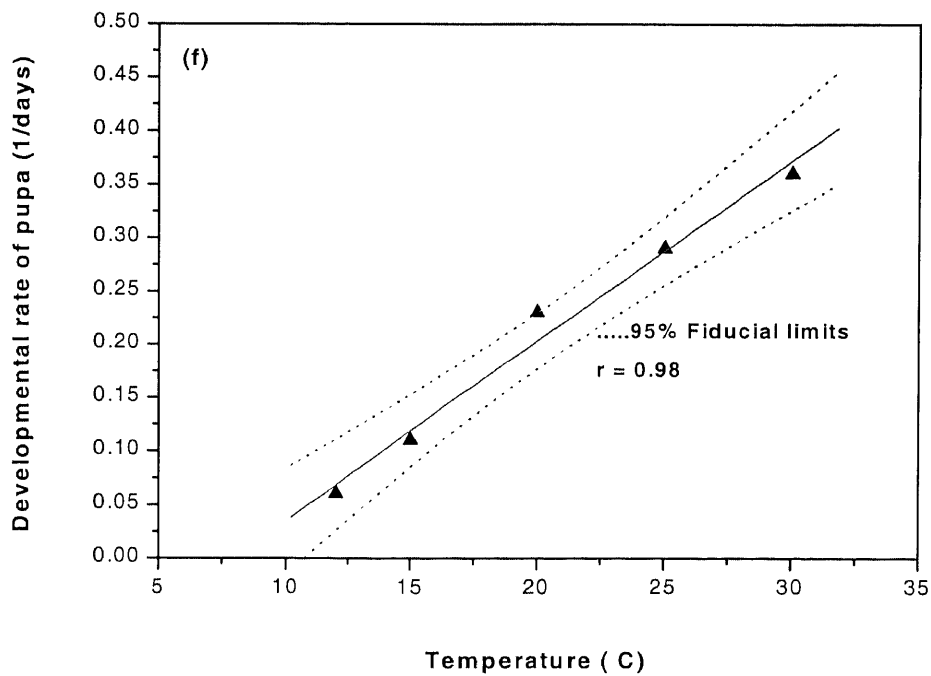
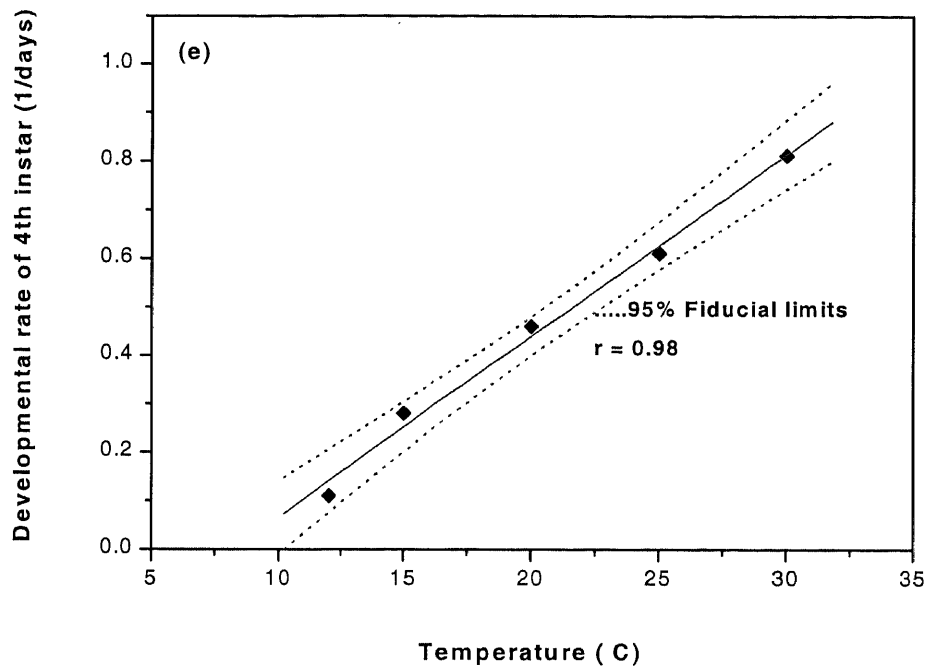


Fig. 3.1 Effect of various constant temperatures on the rate of development of immature stages of *S. vagans*: (e) 4th larval instar, (f) Pupa

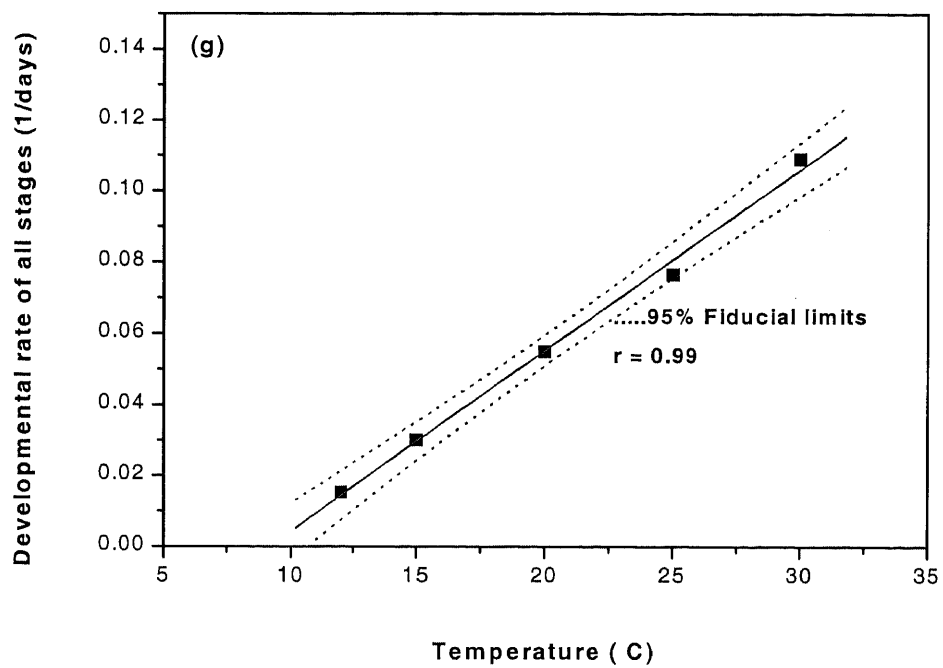


Fig. 3.1 Effect of various constant temperatures on the rate of development of immature stages and all immature stages of *S. vagans*: (g) All stages combined.

3.5 DISCUSSION

3.5.1 Effect of Temperature on the Development of *S. vagans*

The developmental time decreased linearly with increasing temperature in the range 10-35°C for all immature stages of *S. vagans* (Table 3.1). Total developmental time of preimaginal stages of *S. vagans* (33.2 ± 0.6 at 15°C, and 9.17 ± 0.3 days at 30°C) was different to that reported by Richardson (1977) for *S. nigripes* (52.3 ± 3.4 and 8.3 ± 0.4 respectively). This slight difference in development rates between these two Australian species may be because *S. vagans* commonly occurs in coastal and sub-coastal climates, while *S. nigripes* is largely restricted to hotter inland areas (Britton & Lee 1972; Readshaw 1975; Houston 1980). The total developmental time for *S. vagans* was also shorter than that reported for *S. punctillum* (Berker 1958; Jiang *et al.* 1982), *S. gilvifron* (Kaylani 1967; Ahmed & Ahmed 1989) and *S. loi* (Shih *et.al.* 1991). The reason may be that *S. vagans* is a smaller species and is also adapted to different climatic conditions. Another reason may be that some of these species have different primary hosts.

Developmental time for individual life stages also reflected the same trend. *S. vagans* eggs took 9.9 ± 0.7 days at 15°C and 2.2 ± 0.3 days at 30°C to complete development whereas *S. nigripes* took 14.3 ± 0.9 and 1.9 ± 0.3 days respectively (Richardson 1977). The developmental times for 1st, 2nd, 3rd, and 4th, larval instars and pupal stage of *S. vagans* (3.4 ± 0.7 , 3.3 ± 0.5 , 3.5 ± 0.6 , 3.6 ± 0.5 and 9.5 ± 0.5 days at 15°C and 0.97 ± 0.3 , 0.96 ± 0.3 , 1.04 ± 0.3 , 1.24 ± 0.3 , and 2.78 ± 0.3 days at 30°C respectively) were generally shorter than for *S. nigripes* (10.54 ± 1.8 , 3.95 ± 1.56 , 4.19 ± 0.91 , 7.67 ± 0.47 and 9.81 ± 0.59 days at 15°C and 1.26 ± 0.28 , 0.92 ± 0.19 , 0.85 ± 0.23 , 1.23 ± 0.28 and 2.03 ± 0.12 days at 30°C respectively) (Richardson 1977).

The lower developmental threshold temperature for preimaginal stages of *S. vagans* was 10.1, 9.5, 9.5, 9.1, 8.2, 8.0, and 9.1°C for egg, 1st, 2nd, 3rd, 4th larval instars, pupa and all stages combined respectively. Methodology may have had some influence on the higher threshold recorded for eggs. Because the eggs were kept with the portion of the leaf on which they were laid, and other immature stages were reared directly on dry filter paper, there may have been resultant higher humidity in those petri dishes. However differences in threshold temperatures for different immature stages such as those reported here are not uncommon in insect species. For example, Richardson (1977) recorded different lower threshold temperatures for different immature stages of *S. nigripes*, with the highest for the egg stage (11.5°C) and lowest for 2nd and 3rd instars (6.5 and 6.0°C respectively), and Nordin & O’Canna (1985) recorded lower thresholds for fall webworm, *Hyphantria cunea* of 14, 11, and 12°C for egg, larva, and pupa, respectively. Our results are also consistent with those recorded for *S. bifidus*, (11.9°C for egg stages and 9.4°C for 3rd instars) (Peterson *et. al.* 1994).

The lower developmental threshold temperature for eggs appears to be valid based on the data showing that eggs did not develop at 10°C (Section 3.4.1), although they remained viable when exposed to this temperature for a long time (61 days) and subsequently hatched in the normal time period when exposed to temperatures of $\geq 15^\circ\text{C}$. This characteristic may enable this species to survive winter in the egg stage and hatch when temperature and other climatic conditions become favourable. However, in the field very few eggs appear to hatch in winter even when the temperature rises above from 10°C, because field counts over a two year period indicated the abundance of motile *S. vagans* declined (Chapter 4).

Van de Vrie (1972) calculated a lower threshold temperature of 10°C for *T. urticae*, while Readshaw (1975) reported a lower threshold of around 9 to 10°C for eggs and immature

stages of this mite species. Therefore the lower threshold temperatures of prey and predator appear to be in harmony. The period of development reported for *T. urticae* is, however substantially shorter than that for *S. vagans* (i.e. from egg-to-egg 36.4, 16.6, and 7.3 days at 15, 20 and 30°C) (Bodman *et al.* 1993). The development period of *S. vagans* recorded was 43.8, 23.6, and 10.3 days at the same temperatures. The oviposition rate reported for *T. urticae* is also higher (i.e 200 eggs at a rate of 3-14 eggs /female /day in their life span of 3-4 weeks) (Bodman *et al.* 1993). However the rate of predation by *S. vagans* larvae and adults we recorded is much higher than the intrinsic rate of *T. urticae* increase (i.e. larval instars of *S. vagans* consume 27.9-152 eggs /day, and adult males and ovipositing females 63.5 – 142.7 eggs /day). This high predation rate appears to explain reported field observations of rapid declines in spider mite populations in the presence of *Stethorus* spp.

Eggs of *S. vagans* developed into larvae at a constant temperature of 35°C, but they could not survive for a long time. The temperature during summers in the Hawkesbury valley (the region in which the investigations were conducted) does not often rise above 35°C, although temperatures >40°C are occasionally recorded (Pat Hanson, climatic data recorder, UWS, personal communication). On the other hand temperatures in the microclimate within trees, shrubs and ground cover plants are frequently lower than the normal maximum ambient temperature, and humidity at the leaf surface is also higher than ambient conditions. This is likely to provide suitable environments for all *S. vagans* stages to survive and reproduce under otherwise inhospitable environments.

3.5.2 Degree Days (DD)

The degree-day (DD) model is the most widely used approach for describing insect development rate and in predicting insect developmental times as a function of temperature. There were slight differences between the DD calculation for preimaginal stages and all

stages combined depending on whether data was generated from different constant and/or fluctuating temperatures. These slight differences may be due to temperature fluctuations occurring when petri dishes were removed from the temperature cabinets for observation, although every attempt was made to minimise this influence. However, differences in DD resulting from constant and fluctuating temperatures has been reported by a number of authors. For example, Hanula *et al.* (1987) recorded different DD for immature stages of pine the coneworm, *Dioryctri amatella*, at different constant and fluctuating temperatures, and Tolley & Neimczyk (1988) also reported considerable variation in DD for the fruit fly, *Oscinella frit* calculated from eight constant temperatures. Our results (207.8 DD) recorded at a constant 25°C for all stages combined for *S. vagans* is quite close to that recorded for all stages combined of *S. bifidus* (217 DD) estimated at a constant 27.5°C (Peterson *et al.* 1994).

3.5.3 Mortality:

The total mortality observed for *S. vagans* (13.9%) at a constant 25°C was much less than that recorded by Richardson (1977) for *S. nigripes* (40%). Mortality in the egg stage and 1st larval instar was also comparatively lower than for *S. nigripes* (viz. egg mortality 9.2% and 1st larval instar 19.5%). However while mortality in the 2nd larval instar was not very different, no mortality was observed for 3rd instar, 4th instar, prepupal and pupal stages of *S. vagans* compared with 4.2, 3.3, 4.5, and 1.2% for *S. nigripes* receptively. The reason may be that Richardson (1977) used 2.5 cm diameter Munger cells with bean leaf discs over moist cotton balls, while we used 5 cm diameter petri dishes and brushed mite stages on dry filter paper. Richardson (1977) also reported that 3rd and 4th instar often become impaled at leaf disc trichomes and died. In the preliminary investigations for rearing *S. vagans* reported in Chapter 2, high mortality was also recorded. However our rearing techniques were modified to reduce mortality to a more acceptable level.