VG333
Improving the control of Botytis in onions

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Know-how for Horticulture™

VG333

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I. Introduction

Tasmanian production accounts for approximately 70% of Australian onion exports, with 170 growers in the industry. The 1992/93 total value of the Tasmanian onion export industry is estimated to be $25 million. The total Tasmanian production was 72000 tonnes with 57000 tonnes exported (1992/93). Roberts Vegetables exports 6000 tonnes (16%).

Botrytis allii is the major casual pathogen in storage losses of onions and has been identified at Roberts Vegetables as a major limiting factor to reducing costs of production and international competitiveness. Major problems are encountered at grading due to *Botrytis* despite fungicidal seed treatments. Problems include increased packing costs and reduced packout, where it is not uncommon to have 10% rots. According to Roberts Vegetables figures, a 5% packout loss equates to a $60,000 cost to growers in terms of wastage. Packing costs can increase by $20/tonne, equating to $120,000 in a season. Claims at out turn vary from season to season but can range from $10,000 to $220,000.

This project aimed to develop an IPM strategy for controlling this disease. All aspects of onion production were considered under the following objectives:

A. To confirm the main disease causing problems as *B. allii*.
B. To review current management practices in relation to disease control.
C. To predict potential levels of Botrytis in crops by field sampling.
D. To evaluate selected fungicides with respect to efficacy against Botrytis species.
E. To compare fungicide efficacy with respect to application timing.

A number of fungal species are listed in literature as onion diseases however *B. allii* has been well documented as the major pathogen causing rots in onions. This predominantly appears as a neck rot, however the disease may infect the base and through wounds in the side. (Snowdon 1991, Maude 1988)

It has been accepted that seed as the major source of infection. For this reason seed is currently treated with a dusting of benomyl and thiram at the rate of 5g/kg and 4g/kg of seed respectively. Maude & Presly (1977), found that in 1972 and 1973 the level of *B. allii* in seed tested at Wellesbourne 39.5% and 71.4% respectively. Maude and Presly (1977) describe the process of infection from infected seed as mycelial invasion of the cotyledon leaf tip from seed coat. The attack is symptomless. Conidiophores develop after leaf tissue senesces and becomes necrotic. Seed is now tested routinely at Roberts Vegetables. and rejected if *B. allii* is present (levels are greater than 0).

Direct harvesting and drying systems have been shown to reduce disease levels (Maude *et al.* 1984). *B. allii* requires a moist environment for growth. Drying of the bulbs ensures the necks no longer support hyphal growth. The temperature at which bulbs are dried can significantly influence the spread of disease. Alderman and Lacy (1984) found growth of *B. allii* was significantly reduced above 30^{o}C. Harrow and Harris (1969) conclude that the maximum safe temperature for artificial curing is 38^{o}C. At higher temperatures cracking and splitting of skins increases. The outer fleshy tissue cooks, breaks down and is invaded by bacteria. Harrow and Harris (1969) found that 30^{o}C-37^{o}C for 7 days killed mycelium but not spores of *Botrytis allii.* Maude *et al.* (1984) found that disease levels were significantly reduced if topped onions were dried at 30^{o}C. This was found to be most effective if the crop was removed from the field within 48 hours of topping. The project investigated artificial drying at 30^{o}C along with mechanical and hand topping, and compared to the current practice of field curing.

Maude *et al.* (1984) suggested that damage to bulbs and leaves during crop growth provides entry points for spores of *Botrytis allii.* Some crop damage unavoidably occurs from tractor wheels as growers travel through the crop for pesticide and fertiliser application. Onions from traffic areas and non traffic areas were assessed for *Botrytis* infection.

Trichelaar (1967), claimed that infection occurred symptomlessly early in the life of a crop. Various techniques were applied to assess levels of *Botrytis* in the field at different stages in the life of the crop. Early detection of the disease will enable a more accurate targeting of control measures. When reliable methods of estimating levels of *Botrytis* are established action thresholds may be fixed for control measures.

Maude and Presly (1977), have described the infection process in the field, suggesting that the disease enters the leaf tips and grows downwards as the leaves senesce. The disease can exist saprophytically on old leaves during the growing season and becomes parasitic invading the necks of onions at, or close to, harvest.

Kritzman (1983) found foliar applications of fungicides protected onions from infection from *B. allii.* The timings of fungicide applications are unclear, since the disease may be present symptomlessly early in the life of the crop. An important component of this project was to determine when to apply fungicides to the crop and to assess various fungicides for efficacy against *Botrytis.* Determining periods of infection allows for a reduction in the number of sprays than would be required if the crop were to be sprayed repeatedly over the entire season.

II. Objective A

To confirm the disease causing problems as *Botrytis allii.*

A. Materials and Methods

Pathogens from diseased onion bulbs were cultured on PDA plates. Sub cultures were made in an attempt to isolate species of fungus. Fungal growth was observed and compared with a pure cultures of *B. allii* using microscope.

Healthy bulbs were inoculated with spores from a pure culture and stored. Effects observed. The pathogen from the inoculated bulbs were then re cultured and isolated.

B. Results

Isolation of fungi from diseased bulbs was extremely difficult due to the presence of opportunistic fungal species. *Penicillium* was one of the most commonly isolated pathogens. Fungal growth resembling *Botrytis* was observed. In attempts to subculture this fungus however, *Botrytis* was not able to be isolated.

The healthy bulbs inoculated with *B. allii* spores exhibited the same type of symptoms of rotting as breakdown observed in shipments at outturn. Flesh from these rotting bulbs was then cultured on PDA and *Botrytis* was isolated.

C. Discussion

All indications are that the disease is *B. allii.* It is apparent that more isolations are required to fully prove that *B. allii* is the disease causing problems. An attempt was made to infect growing plants with spores of *B.* allii to show that bulbs from these plants would break down in storage. However, the plants that were growing in pots were destroyed before the bulbs could be harvested and assessed.

III. Objective B

To review current management practices in relation to disease control.

A. Materials & Methods

Climatic conditions for Roberts Vegetables lifting and harvesting were examined for two previous seasons (91/92 and 92/93). Levels of *Botrytis* in field bins for individual growers was taken from quality control records for each season.

1. The level of Botrytis in each crop was compared to:

1.the number of days with rain from lifting to harvest
2.total rainfall for the five days after lifting
3.average maximum temperature lifting to harvest
4.total rainfall lifting to harvest

2. A trial was conducted to determine the value of artificial curing compared to field curing and hand machine topping prior to the curing process. Treatments were selected accordingly:

1. Artificial Drying Tops On
2. Artificial Drying Hand Topped
3. Artificial Drying Machine Topped
4. Field Cured Tops On
5. Field Cured Hand Topped
6. Field Cured Machine Topped

Six sites were selected encompassing early, mid and late season onion crops:

2 early - Hubbard and Cooper
2 mid season - Franks and Cook
2 late - Stuart and Plapp

Five replicates of each treatment at each site were applied. Plots of 1 metre of onion bed were randomly selected. The two late season crops (Stuart and Plapp) were harvested before samples could be taken and were not included in the trial.

An insulated drying chamber was constructed. Samples to be artificially cured were placed in this chamber which was maintained at 30^{o}C and 50% RH. Dried samples were then transferred to a fantainer to be stored prior to assessment. Field cured onions placed in the fantainer after harvest.

Samples were assessed after 150 days in storage in the fantainer.

3. The influence of damage from traffic through the crop was assessed. At two sites, onions were sampled from a bed within a spray run. When spraying, the tractor straddles the bed and some injury is inflicted on onion bulbs in outer rows of the bed. Onions in the outer rows were marked for identification. Immediately prior to harvesting a sample was taken of onions from within the bed and in the outer rows. After 195 days in storage onion samples were assessed for signs of *Botrytis*.

B. Results
Results from comparisons of climatic data are included in Appendix One.

Tabulated data for the drying trial is included in Appendix Two. Graphical representation of data is presented in figures 1 to 3.

Figure 1:

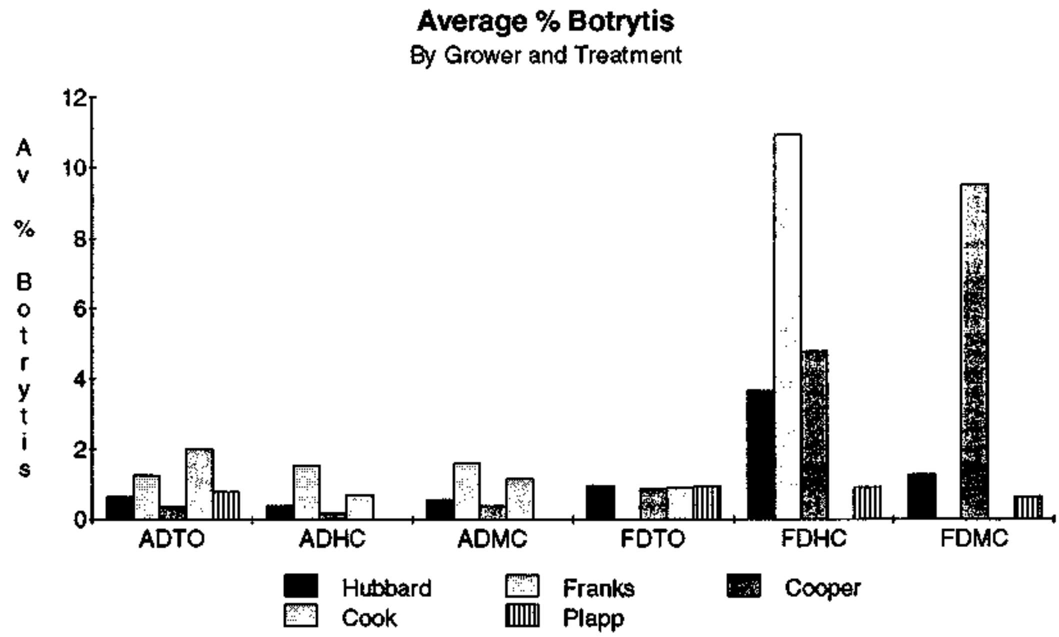


Figure 2:

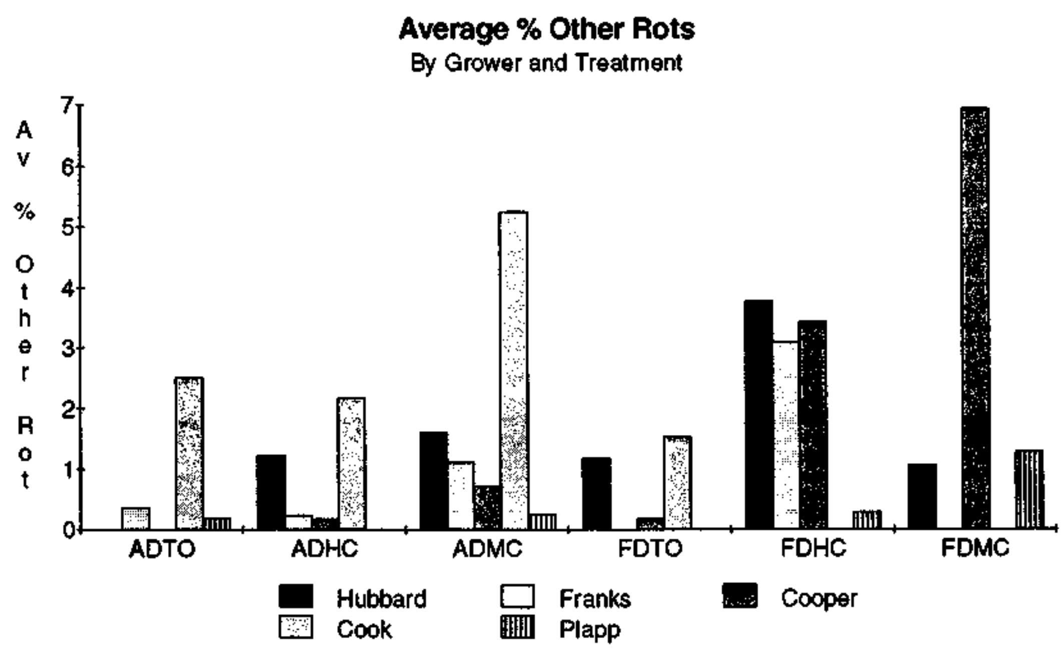
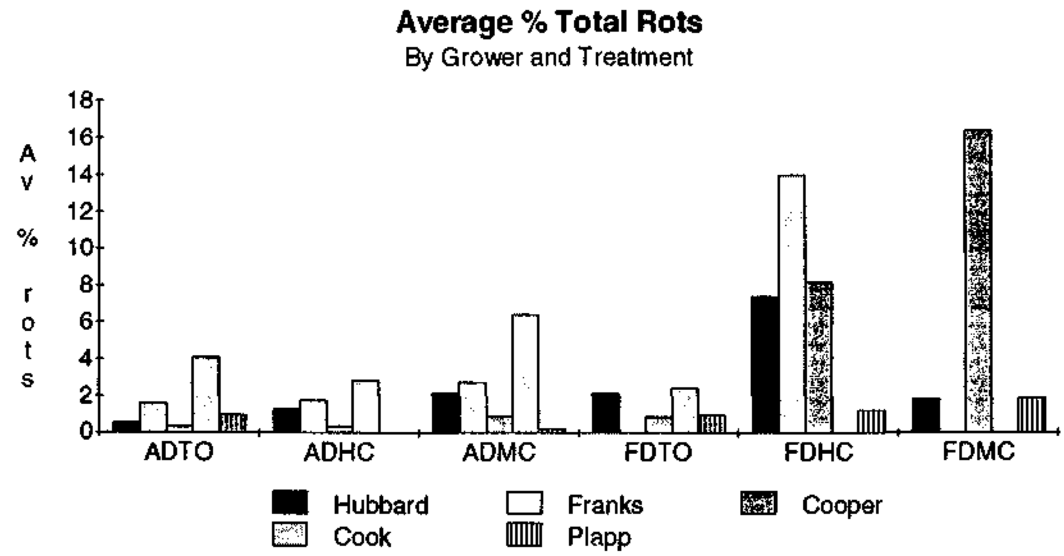


Figure 3:



Data from traffic damage to crop assessment:

Site	Sample Taken	No. Onions	Botrytis	Other Rots
West Pine	inner row	81	2	2
	outer row	88	0	0
Moorleah	inner row	119	2	0
	outer row	137	3	3

C. Discussion

No obvious pattern was observed from the climatic data.

There was a very large variation both within and between growers in the drying trial. Such variation makes statistical analysis difficult. While the results appear inconclusive, it is apparent that any form of topping associated with field drying results in high levels of *Botrytis* following storage. The level of *Botrytis* in all crops was low for the season of this project, making it difficult to observe a significant difference between treatments. Generally the treatment "artificially dried hand cut" had low levels of *Botrytis*. In a poor season the value of this treatment would be evidenced.

No clear conclusions may be drawn from data obtained from crop damage assessments. There is no discernible difference in percentage of rots, either *Botrytis* or other rots, between inner and outer rows of onions in traffic beds. *Botrytis* levels in these samples correspond with low levels observed in other trials for the season 1993/94.

A larger scale survey should be conducted in order to accurately determine the impact of wheel damage to onions.

IV. Objective C

To predict potential levels of *Botrytis* in crops by field sampling.

A. Materials and Methods

Random samples of plants were collected fortnightly from three crops.

Samples were incubated in sealed plastic bags at 20^{O}C until fungal growth was apparent. Any fungi present were examined under microscope and sub cultured on PDA plates.

Two attempts of spore trapping were made at 2 sites - one currently sown with *Botrytis* infected seed and the other at the site of a previous years crop which had significant levels of *Botrytis*. 1/2 strength PDA plates were held to the prevailing wind at a number of positions across the field for a count of 30 seconds. The plates were then inoculated for 1 week at 20^{O}C.

B. Results

No plant samples were found to have *Botrytis* after inoculation in bags.

Only one occurrence of *Botrytis* was found on spore trap plates. This was on a plate taken from the site of the previous years crop, approximately one metre from a self seeded onion plant.

V. Objective D

To evaluate selected fungicides with respect to efficacy against *Botrytis* species.

A. Materials and Methods

A trial was carried out in order to screen a number of fungicides both individually and in combination. The trial was conducted within a commercial crop. The seed for this crop was treated with 2g/kg Benlate[R] and 5g/kg Thiram[R]. There were seven treatments:

1. Control
2. Folicur 600 ml/ha
3. Folicur 1.2 L/ha
4. Folicur 3 L/ha
5. Folicur 600 ml/ha + Bravo 3 L/ha
6. Sumisclex 2 L/ha
7. Benlate 2 kg/ha + Bravo 3 L/ha

Each treatment was applied twice. Firstly on 7/10/93, at the 1-2 leaf stage and then again 7 days later. A randomised complete block design was used, with three replicates per treatment. Each replicate consisted of one bed of onions, 10 metres long.

At harvest a random sample of 200 onions per treatment replicate were taken then stored. After 141 days samples were assessed for signs of *B. allii.*

B. Results

Disease Assessment After 141 Days in Storage

Treatment No	% *Botrytis*
1	1.5
2	1.84
3	1.0
4	0.84
5	1.84
6	1.34
7	2.0

In an earlier assessment the untreated control showed 1.17% *Botrytis* infection after 64 days in storage.

There are no significant differences between treatments.

C. Discussion

Botrytis levels were low with *Botrytis* levels in the control no higher than in other treatments. Whilst there was no significant result, there is an indication of a decreasing trend in *Botrytis* levels with increasing rates of Folicur (treatments 1-3). Also at higher Folicur[R] rates, *Botrytis* levels tend to be lower than in the Benlate[R] treatment.

VI. Objective E

To compare fungicide efficacy with respect to application timing.

A. Materials and Methods

A trial was conducted to assess effect of timing of fungicide application on *B. allii*. The trial area was planted with untreated seed with an infection of 2-3% *B. allii*. Nine treatments were applied:

Treatment	Timing*
1. Control	
2. Benlate 2 kg/ha + Ronilan 2 L/ha	A
3. Benlate 2 kg/ha + Ronilan 2 L/ha	B
4. Benlate 2 kg/ha + Ronilan 2 L/ha	C
5. Benlate 2 kg/ha + Ronilan 2 L/ha	A+
6. Benlate 2 kg/ha + Ronilan 2 L/ha	A+C
7. Benlate 2 kg/ha + Ronilan 2 L/ha	B+C
8. Folicur 600 mL/ha + Bravo 3 L/ha	A
9. Benlate 2 kg/ha + Ronilan 2 L/ha	A

*Timing
A - 22/08/93 (1 to 2 true leaf stage)
B - 24/12/93 (4 to 5 true leaf stage)
C - 11/01/94 (early bulbing)

Treatments 1 to 8 consisted of two applications, the first being according to the stage of the crop and the second seven days later. Treatment 9 began at the 1 to 2 true leaf stage of the crop and applications occurred every ten days until tops fell.

A randomised complete block design was used, with four replicates per treatment. Each replicate consisted of one bed of onions, 10 metres long.

At harvest a random sample of 200 onions per treatment replicate were taken then stored. After 131 days samples were assessed for signs of *B. allii*.

B. Results

Disease Assessment After 131 Days in Storage

Treatment No	% *Botrytis*
1	12.38
2	7.13
3	9.0
4	6.75
5	6.5
6	7.38
7	6.38
8	5.38
9	8.38

The untreated control showed 6.63% *Botrytis* infection after 54 days in storage compared with 12.38% at 131 days. This is evidence of the late development of disease symptoms

Botrytis infection of treatment eight is significantly lower than that of other treatments.

C. Discussion

In this timing trial, the treatment of Folicur + Bravo applied at the 1-2 leaf stage showed significant *Botrytis* control when compared to the untreated control. No other treatments showed significant control.

The failure of Benlate + Ronilan applied at ten day intervals (treatment 9) is of concern. This treatment far exceeds what would be considered a commercial strategy for *Botrytis* control.

Botrytis levels in the timing trial were quite high when compared to the screening trial. The levels of up to 12% are quite unacceptable. The contribution of seed dressing to *Botrytis* control is evident and should be further investigated. Foliar treatments alone may not provide adequate *Botrytis* control. The fungicides used in the timing trial may not be effective or timing may need to be modified.

Although results are limited, a trend may be interpreted from both the timing and screening trials indicating low efficacy from Benlate. Further investigation may be required to determine its efficacy or if resistance of *B. allii* to Benlate exists.

VII. Summary

Botrytis allii innocuously infects the growing onion in the crop. Symptoms of bulb rotting do not become apparent until harvest or during storage. Crops infected with *B. allii* lead to increased production costs and reduced out turn through the need for increased inspection time and bulb rotting.

The project has identified *B. allii* as the probable causal agent producing major observed storage rots. More isolations are required to fully prove this however.

The presence of poor weather conditions does not necessarily lead to *Botrytis* infection. If *Botrytis* is present however its severity is increased.

The topping trials showed that when damage occurs through topping and the bulbs are allowed to field dry, the incidence of *Botrytis* increases. This infection is not so prevalent when bulbs are artificially dried as the bulb is dried more quickly preventing growth of the fungus in the neck.

Although results were not significant, the fungicide screening trials indicated that increasing Folicur^R rates may provide increasing control of *Botrytis.* Botrytis levels were low in this trial indicating the benefits of seed treatment in the control programme.

In timing trials applications of Folicur^R + Bravo^R showed significant *Botrytis* control. Both the screening and timing trials showed low efficacy from Benlate^R.

VIII. Recommendations

Future research should be directed at:

A. Further identification of the disease.

B. Developing an efficient system for forecasting likely disease events (eg downy mildew and botcast).

C. Investigating combination of seed treatment and foliar fungicide applications.

D. Screening fungicides in the laboratory to evaluate their relative efficacy against *Botrytis*.

E. Investigating possible infection after lifting and in storage

Much of the work carried out in this project should be repeated over an extended period to allow for seasonal variations in disease levels.

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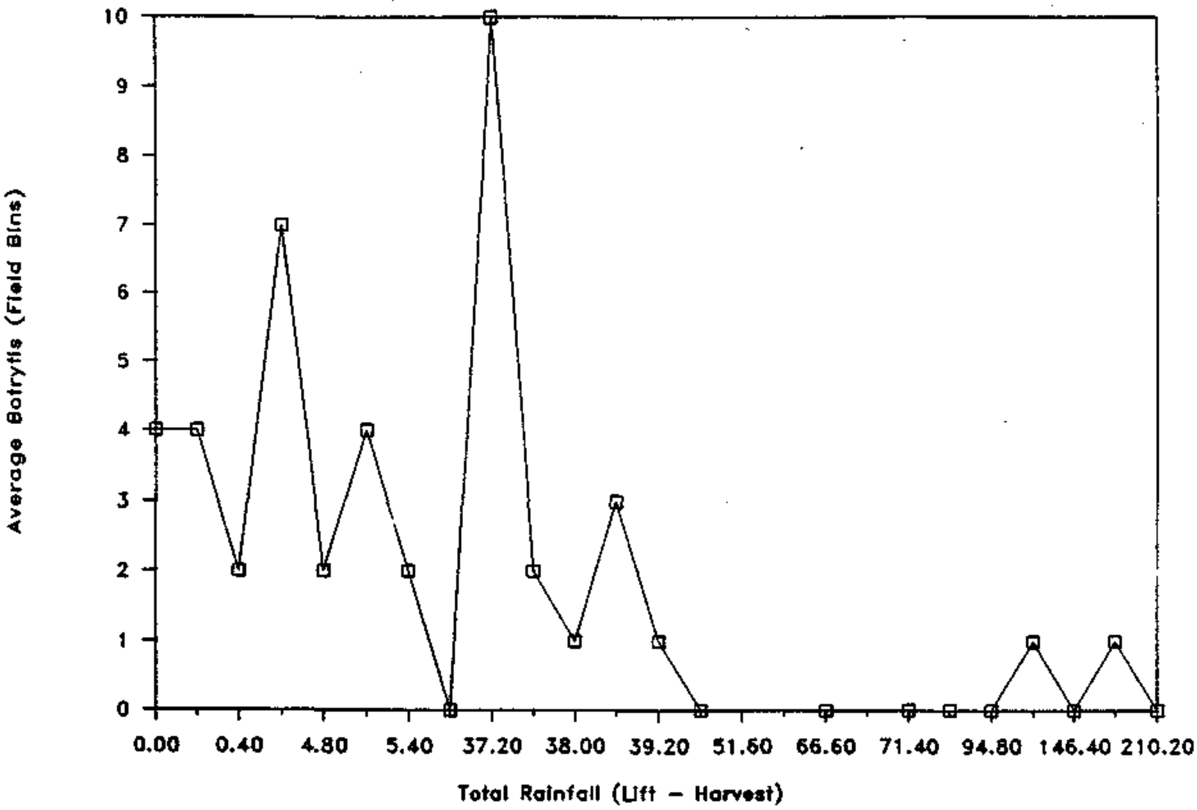
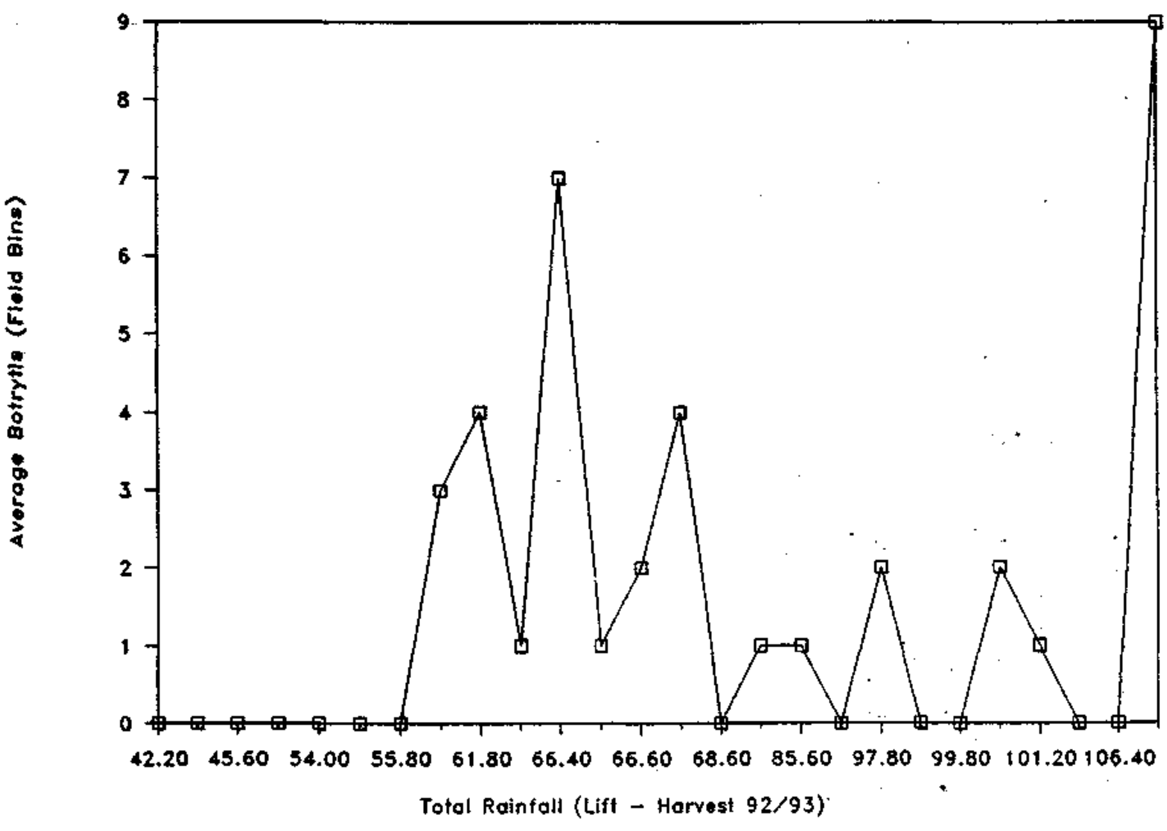
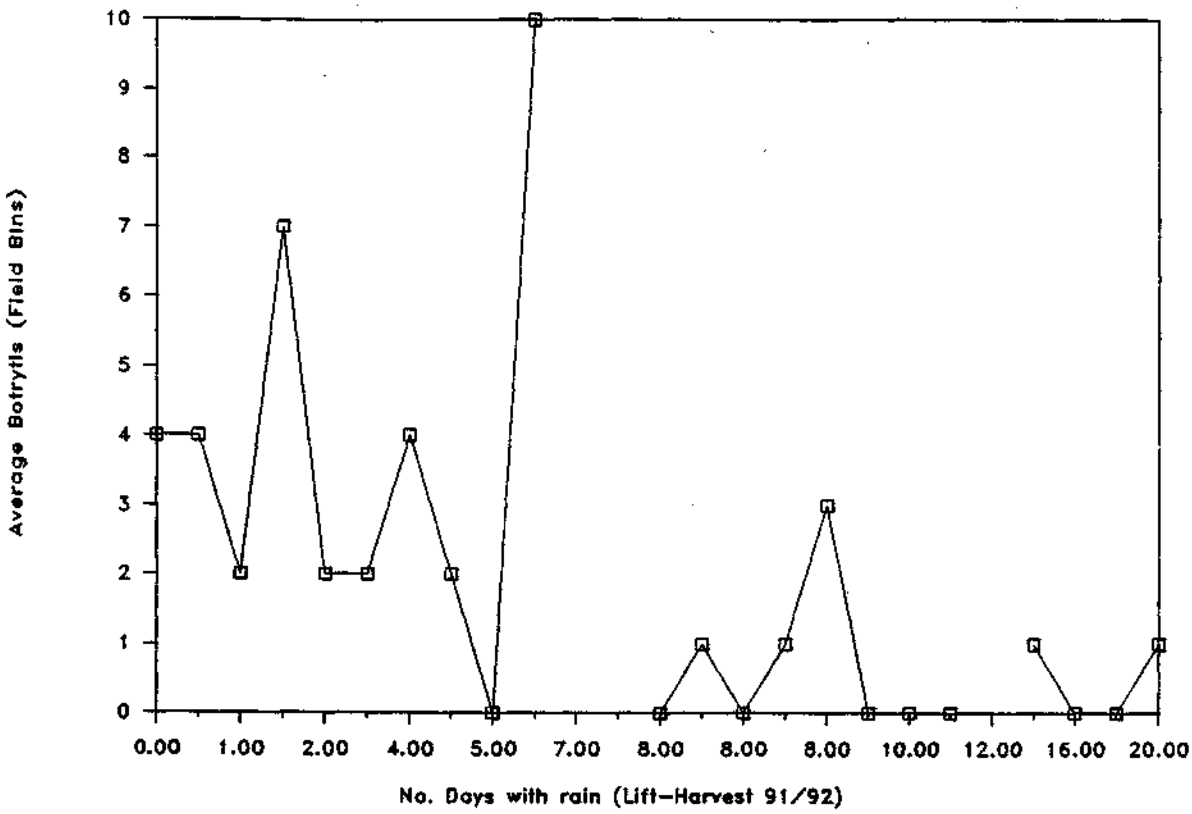
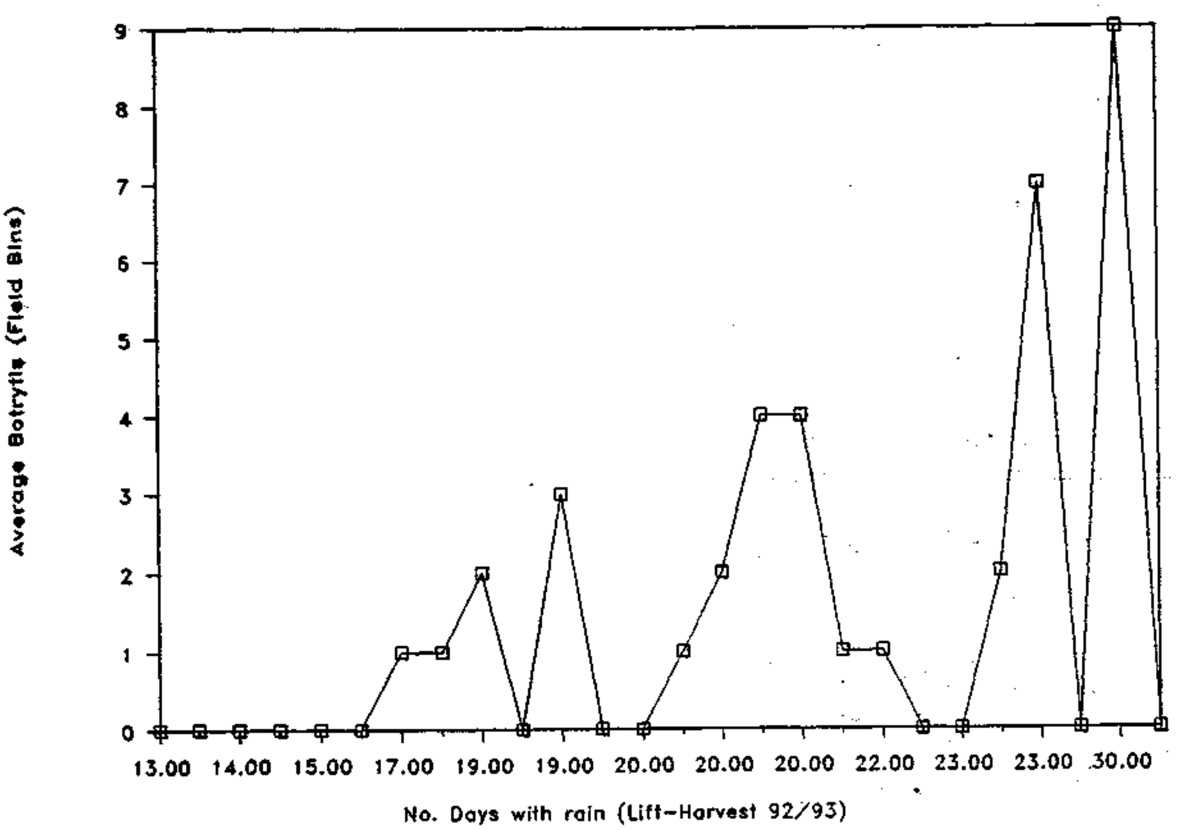
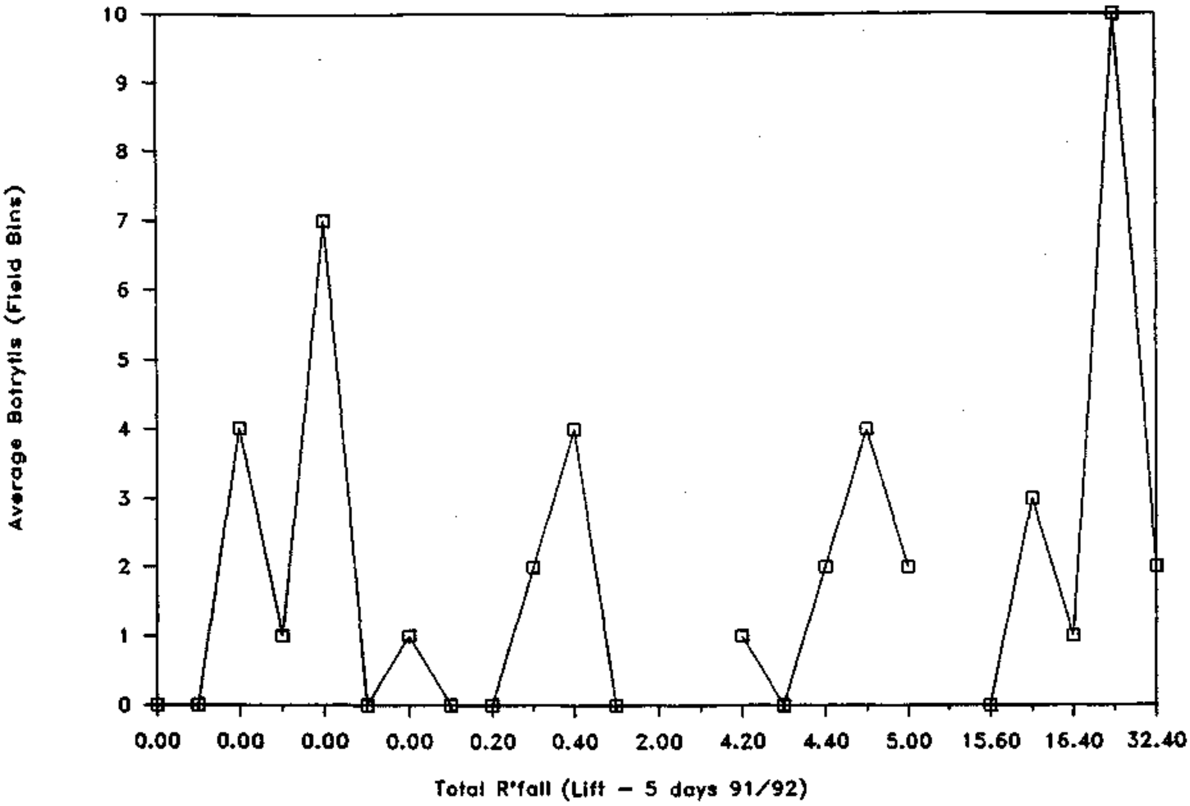
Appendix One - Climatic Data Comparisons



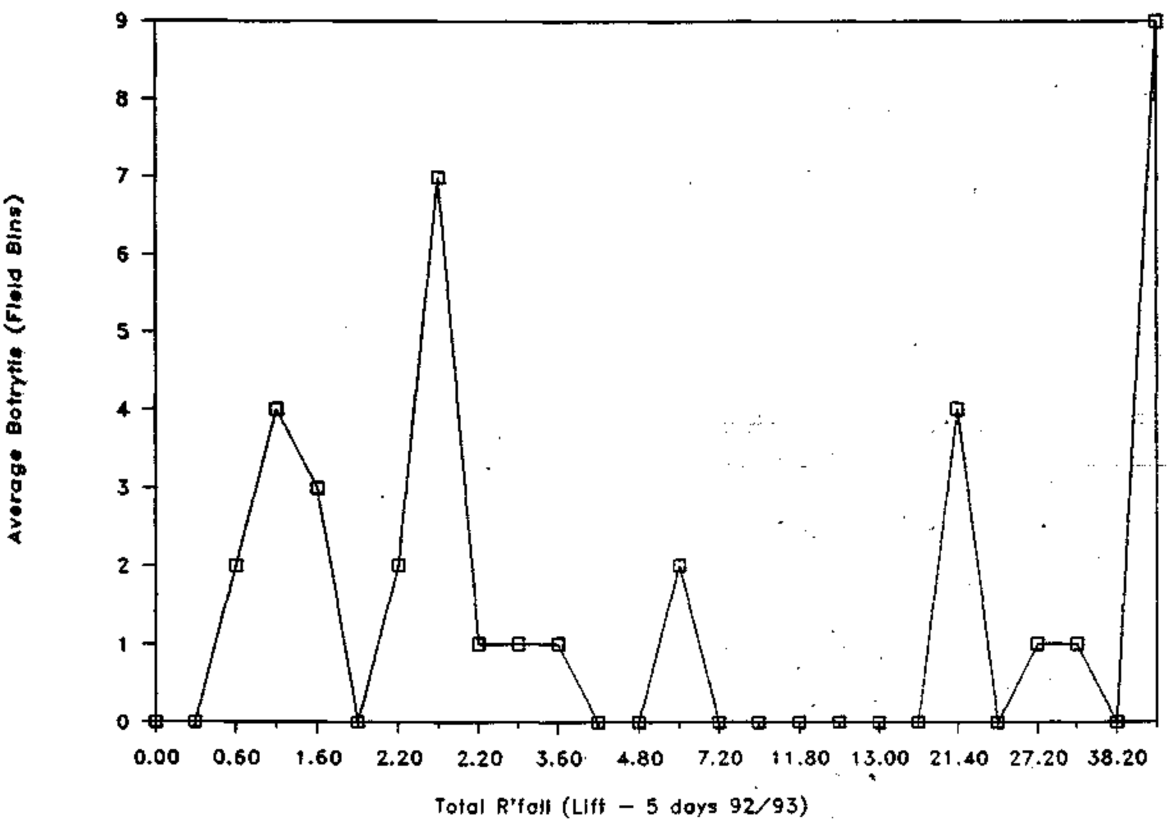



Average Botrytis (Field Bins)
0
1
2
3
4
5
6
7
8
9
10
0.00
1.00
2.00
4.00
5.00
7.00
8.00
8.00
8.00
10.00
12.00
16.00
20.00
No. Days with rain (Lift-Harvest 91/92)

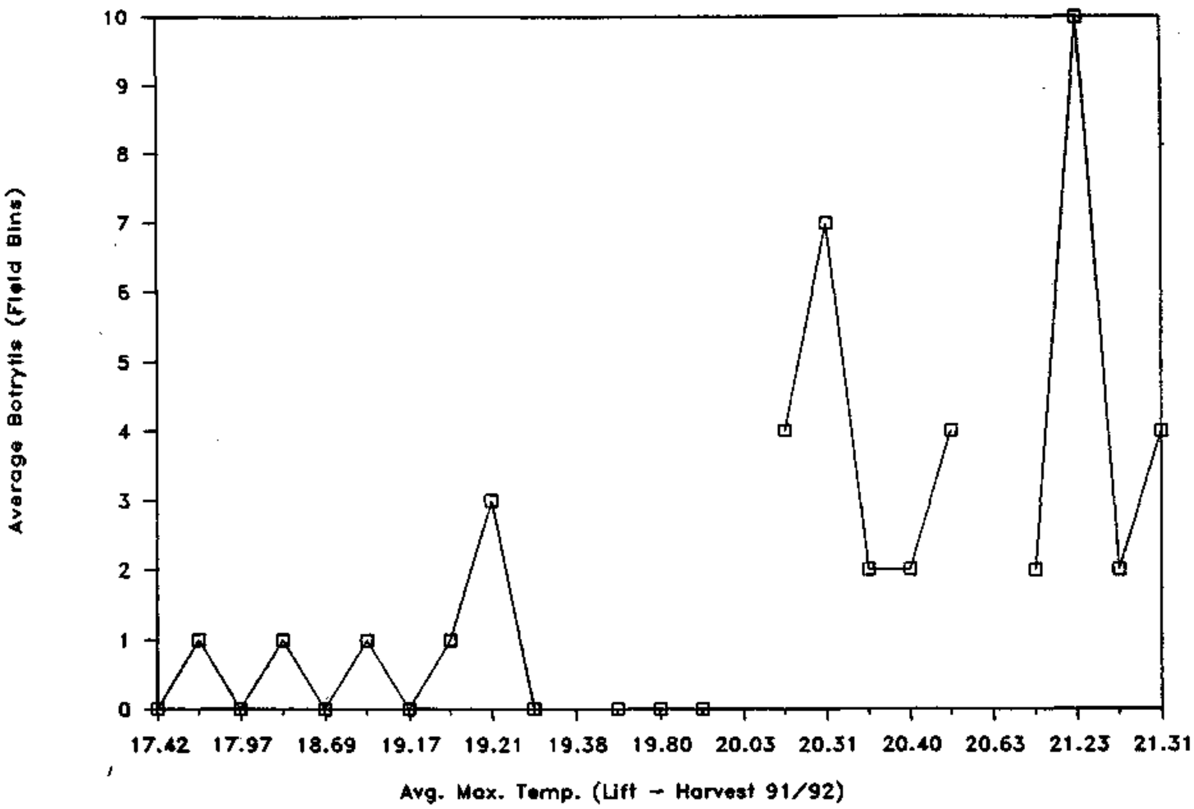

Average Botrytis (Field Bins)
0
1
2
3
4
5
6
7
8
9
13.00
14.00
15.00
17.00
19.00
19.00
20.00
20.00
20.00
22.00
23.00
23.00
30.00
No. Days with rain (Lift-Harvest 92/93)

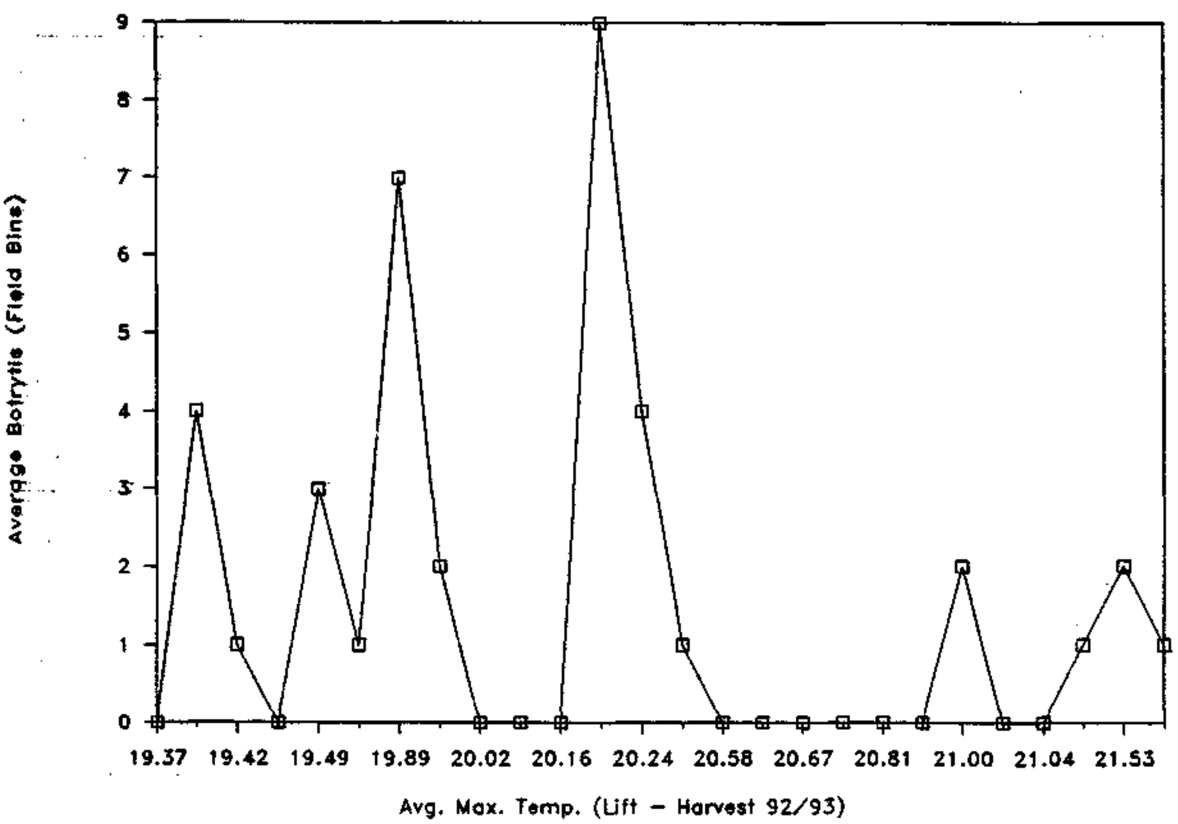


Average Botrytis (Field Bins)
10
9
8
7
6
5
4
3
2
1
0
0.00
0.00
0.00
0.00
0.20
0.40
2.00
4.20
4.40
5.00
15.60
16.40
32.40
Total R'fall (Lift – 5 days 91/92)



Average Botrytis (Field Bins)
9
8
7
6
5
4
3
2
1
0
0.00
0.60
1.60
2.20
2.20
3.60
4.80
7.20
11.80
13.00
21.40
27.20
38.20
Total R'fall (Lift – 5 days 92/93)


Average Botrytis (Field Bins)
10
9
8
7
6
5
4
3
2
1
0
17.42 17.97 18.69 19.17 19.21 19.38 19.80 20.03 20.31 20.40 20.63 21.23 21.31
Avg. Max. Temp. (Lift - Harvest 91/92)


Average Botrytis (Field Bins)
9
8
7
6
5
4
3
2
1
0
19.37 19.42 19.49 19.89 20.02 20.16 20.24 20.58 20.67 20.81 21.00 21.04 21.53
Avg. Max. Temp. (Lift - Harvest 92/93)

Appendix Two - Drying Trial Data

Average Percentage Total Rots by Grower and Treatment

GROWER		ADTO	ADHC	ADMC	FDTO	FDHC	FDMC
Hubbard		1.14	2.98	0	3.14	3.73	0
		0	0	1.25	1.64	4.35	4.23
		0	1.19	3.16	1.66	7.69	0
		1.39	2.22	4.62	1.83	5.26	5.04
		0	0	1.74	2.37	16.13	0
	av	0.506	1.278	2.154	2.128	7.432	1.854
Franks		0	0	0		17.82	
		1.86	2.63	1.08		7.62	
		0.88	3.89	3.53		18.18	
		3.6	1.15	4.08		7.21	
		1.74	1.15	4.88		19.35	
	av	1.616	1.764	2.714		14.036	
Cooper		0	1.62	0	1.68	4.34	8.82
		0	0	0	0.93	11.22	18.18
		1.72	0	1.37	0.98	3.19	15.94
		0	0	2.98	0.74	6.14	16.1
		0	0	0	0	16.26	23.23
	av	0.344	0.324	0.87	0.866	8.23	16.454
Cook		9.38	1	2.63	3.3		
		2.06	5.49	3.77	4.21		
		4.66	0	4.55	1.58		
		0	1.41	6.41	3.09		
		4.42	6.42	14.77	0		
	av	4.104	2.864	6.426	2.436		
Plapp		0	0	0	0	2.09	4.24
		0	0	0	0	0	0.78
		1.92	0	0	2.17	1.78	1.46
		0	0	0	0	0.55	2.22
		2.9	0	1	2.53	1.5	0.8
	av	0.964	0	0.2	0.94	1.184	1.9

ADTO = artificially dried tops on
ADHC = artificially dried hand cut
ADMC = artificially dried machine cut
FDTO = field dried tops on
FDHC = field dried hand cut
FDMC = field dried machine cut

Average Percentage Botrytis by Grower and Treatment

GROWER		ADTO	ADHC	ADMC	FDTO	FDHC	FDMC
Hubbard		1.14	1.49	0	1.57	2.24	0
		0	0	0	0.82	2.61	2.54
		0	0	1.05	0.83	1.92	0
		1.39	0	0	0	2.63	2.52
				1.74	1.55	8.87	
	av	0.6325	0.3725	0.558	0.954	3.654	1.265
Franks		0	0	0		11.88	
		0.93	2.63	1.08		5.08	
		0.88	3.89	0		17.27	
		3.6	1.15	2.04		5.41	
		0.87	0	4.88		15.05	
	av	1.256	1.534	1.6		10.938	
Cooper		0	0.81		1.68	2.17	7.35
		0	0	0	0.93	3.06	9.09
		1.72	0	0	0.98	2.13	10.14
		0	0	1.49	0.74	4.39	5.93
		0	0	0	0	12.19	15.15
	av	0.344	0.162	0.3725	0.866	4.788	9.532
Cook			0	0	1.65		
		2.06	1.09	0	0		
		2.33	0	1.14	0.79		
		0	1.41	1.28	2.06		
		3.54	0.92	3.41	0		
	av	1.9825	0.684	1.166	0.9		
Plapp		0	0		0	1.4	0.85
		0	0	0	0	0	0
		0.96	0	0	2.17	1.78	0
		0	0	0	0	0.55	1.48
		2.9	0	0	2.53	0.75	0.8
	av	0.772	0	0	0.94	0.896	0.626

Average Percentage Other Rots by Grower and Treatment

GROWER		ADTO	ADHC	ADMC	FDTO	FDHC	FDMC
Hubbard		0	1.49	0	1.57	1.49	0
		0	0	1.25	0.82	1.74	1.69
		0	1.19	2.11	0.83	5.77	0
		0	2.22	4.62	1.83	2.63	2.52
				0	0.82	7.26	
	av	0	1.225	1.596	1.174	3.778	1.0525
Franks		0	0	0		5.94	
		0.93	0	0		2.54	
		0	0	3.53		0.91	
		0	0	2.04		1.8	
		0.87	1.15	0		4.3	
	av	0.36	0.23	1.114		3.098	
Cooper		0	0.81		0.84	2.17	1.47
		0	0	0	0	8.16	9.09
		0	0	1.37	0	1.06	5.8
		0	0	1.49	0	1.75	10.17
		0	0	0	0	4.07	8.08
	av	0	0.162	0.715	0.168	3.442	6.922
Cook		9.38	1	2.63	1.65		
		0	4.4	3.77	4.21		
		2.33	0	3.41	0.79		
		0	0	5.13	1.03		
		0.88	5.5	11.36	0		
	av	2.518	2.18	5.26	1.536		
Plapp		0	0		0	0.69	3.39
		0	0	0	0	0	0.78
		0.96	0	0	0	0	1.46
		0	0	0	0	0	0.74
		0	0	1	0	0.75	0
	av	0.192	0	0.25	0	0.288	1.274