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# RELEVANT FACTORS INFLUENCING SEED SET IN COMMERCIAL SUNFLOWER (*HELIANTHUS ANNUUS*) IN SOUTH AFRICA

DPhil (BOTANY)

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# Relevant factors influencing seed set in commercial sunflower (Helianthus annuus) in South Africa

by

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## ABSTRACT

Low seed set and hollow seededness are amongst the main problems of the sunflower oilseed production industry in South Africa. This study of eighteen sunflower cultivars on seventeen different localities was conducted to address these and other problems. It was undertaken in collaboration with the national sunflower trials.

The study has revealed that a lack of pollinators had the least significant influence on seed set. Neither could the percentage hollow seeds at each locality be attributed to a low number of insect pollinators. Commercial sunflowers in cages with honeybees gave a seed yield of 1,859 kg ha-1 (72% seed set) compared to 1,221 kg ha-1 (45% seed set) in cages with no insect pollinators.

The percentage unfilled seeds differed significantly between localities and cultivars. However, the incidence of both hollow seeds and unfilled seeds differed highly significantly with the increase in plant density from 15,000 plants per ha to 90,000 plants per ha. Yields also decreased as plant density increased.

Although the boron levels differed between soil, leaf and floret samples for the variables of locality, cultivar and plant density, no relationship was found between the boron levels and differences observed in the percentage hollow seeds and unfilled seeds between the eighteen sunflower cultivars. Slow pollen movement was identified as the main cause responsible for hollow seededness. A lack of nutrients resulted in the high levels of unfilled seeds found. Recommendations are given regarding these problems.

Viability and vitality of the sunflower pollen was good. It was, however, not possible to germinate the trinucleate sunflower pollen in vitro using conventional palynological methods and techniques. It is hypothesized that an unidentified "pollen growth factor", occurring on the stigmas, regulates germination of sunflower pollen.

# CHAPTER 1

# INTRODUCTION

## 1.1. Characteristics of commercial sunflower

The common sunflower (<u>Helianthus annuus</u> L.) belongs to the Asteraceae, which is one of the largest flowering plant families. Plants in this family have many small flowers aggregated in a head (capitulum). On the rim of the capitulum a single row of bright yellow sterile flowers, the ray florets are borne. The remainder of the capitulum consists of up to 2000 disc florets. Each disc floret has the potential to develop a one seeded fruit, also known as an achene.

The sunflower originates from the southwestern USA and was initially cultivated as an important oil-rich crop in Russia early in the nineteenth century (Heiser, 1955; Hurd, La Berge & Linsley, 1980; McGregor, 1976). Sunflower breeding intensified in America during the 1940's and was mostly aimed at earlier maturity, higher oil content of the seeds and disease resistance. These earlier open-pollinated cultivars were, however, very variable in development. Different breeding concepts were subsequently applied and these initiated the modern programme of hybrid sunflower seed production. The most revolutionary idea developed since 1970 is the cytoplasmic male-sterile system used

in hybridization. Since then we distinguish between sunflower planted for commercial purposes and that for hybrid seed production. Sunflower hybrid seed production is totally dependent upon insects to carry pollen from the male-fertile plants to the male-sterile plants (Radford & Rhodes, 1978).

Although the level of self-pollination of modern commercial hybrid sunflowers is better than that of the open pollinated varieties, the level of self-fertility is questioned. According Lewis (1979), plants have a natural resistance to to self-fertilization (autogamy). In sunflowers, self-pollination and self-fertility are controlled by a time difference in the availability of pollen and receptiveness of the stigma. Cross-pollination is therefore favoured, although selfpollination and consequently sib-fertilization, i.e. pollen received from another floret on the same head, is possible. Researchers therefore agree that, to achieve a high level of seed set of 70% or more, pollinators are still needed (Birch, Van der Sandt, Herrmann & Johannsmeier, 1985; Freud & Furgala, 1982; Furgala, Noetzel & Robinson, 1978; Krause & Wilson, 1981).

## 1.2. Seed set and yield

Poor seed set is one major factor affecting the sunflower crop in South Africa. Seed set and yield is determined by the separate but vital processes of flower initiation, pollination, fertilization and seed development (Birch, 1981). Birch (1981)

and Herring (1981) discussed the factors that influence these processes in South Africa. These include climate, cultivar characteristics, insect pollinators and plant nutrition. Birch (1982) revealed that South Africa was the only large sunflower producing country which lacked the relevant research on bees as pollinators. Preliminary research to investigate the influence of honeybees on yield was consequently undertaken (Birch et al., 1985). The effects of Apis mellifera scutellata Lepeletier (African honeybee), Astylus atromaculatus Blanchard (spotted maize beetle), <u>Heliothis</u> <u>armigera</u> Hubner (American bollworm larvae) and <u>Musca</u> domestica L. (house fly) on seed set in commercial sunflower were established by Du Toit (1990). Commercial sunflowers in cages with honeybees gave a seed yield of 1,859 kg ha<sup>-1</sup> (72% seed set) compared with 1,221 kg ha<sup>-1</sup> (45% seed set) in cages with no insect pollinators. A seed set of 73% was achieved in open control treatments. Spotted maize beetles were found efficient as pollinators (76% seed set). However, American bollworm larvae (44% seed set) and house flies (38% seed set) were insignificant as pollinators of sunflower.

### 1.3. Pollination in plants

Certain criteria in pollination are necessary for a plant insect relationship to evolve (Faegri & Van der Pijl, 1979; Kevan, 1983). The bodies of anthophilous insects should be covered with setae, upon which pollen can readily cling. An ability to recognize and imprint plant form, as well as a communication system, is advantageous. The pollinator should not be too specialized since specialization towards utilizing a single food source can lead to decreased flexibility without the particular food source. Insect-pollinated plants should have nectar of good quality and quantity available as a reward. More advanced bee plants have their nectar hidden in narrow, protective tubes, as in the Asteraceae. A nectar reward for pollination has led to various degrees of plant-insect relationships, a very specialized form being found, for example in orchids pollinated by euglossine bees.

According to Kevan (1983) the Coleoptera as a group are the most primitive holometabolous pollinators. Diptera are also regarded as primitive anthophilous insects. Adult Lepidoptera, which visit flowers for their nectar as an energy source, play some role in pollination. Hymenoptera are important pollinators, with members of the Superfamily Apoidea being the most important. It is estimated that the Apoidea are responsible for 80% of all pollination by insects. Heteroptera are the most common hemimetabolous anthophilous insects but their importance as pollinators is uncertain, at best minor.

## 1.4. Pollination of sunflower

Insects visit the flowering capitula of commercial sunflower to seek food, mating sites or shelter (Hurd <u>et al.</u>, 1980). During

their activities on the capitulum some of these insects transfer pollen to the stigmas.

The conspicuous sunflower capitulum is a good example of plant adaptation to insect pollination (Faegri & Van der Pijl, 1979; Kevan 1983). The bright yellow ray florets serve as a visual attraction, while the disc florets provide a highly-clustered energy reward. Crowding of the florets ensures that the maximum number of florets are pollinated by a single insect visit.

Wind is of almost no importance in the pollination of sunflower, as the pollen is heavy and sticky (Robinson, 1978). Although modern hybrid sunflower cultivars are bred to have а self-pollination efficiency of up to 50%, insects are internationally reported to be the primary cross-pollinators of sunflower: in the United States (McGregor, 1976; Hurd et al., 1980), in eastern Europe (Benedek, Manninger & Nagy, 1972), in India (Goyal & Atwal, 1973), in Argentina (De Diez, 1979) and in Australia (Langridge & Goodman, 1974; Radford, Nielsen & Rhodes, 1979 a+b). Pollinators listed by these authors include not only beneficial honeybees and solitary bees, but also agricultural pests such as American bollworm (larvae and moths) and spotted maize beetles. A wide range of other unimportant pollinators (based on their abundance and behaviour), including butterflies, flies and wasps, have been recorded as visitors to sunflower capitula.

The diversity, abundance and behaviour of insects on flowering

capitula have been studied in depth in South Africa by Du Toit (1988) and Du Toit & Holm (1992 a+b). This was based on three study sites in the Transvaal during 1985-1987. The mean number of insects per 100 capitula was 45, 147 and 64 at Settlers, Pretoria and Hartbeesspruit, respectively. Indigenous honeybees were the most common in all study areas (71%, 86% and 46% respectively). Other well-represented taxa included American bollworm larvae, spotted maize beetles, Diptera (flies) and Hemiptera (bugs). American bollworm moths, which were most active from dusk till 20:00, were the most abundant nocturnal insect species (76%), with a mean of 6,3 moths per 100 receptive capitula.

The leafcutter-bee (Eumegachile pugnata Say) has only relatively recently been discovered as the primary pollinator of wild sunflower in its natural habitat in America (Parker and Frohlich, 1983). The South African survey revealed that solitary bees were not frequent visitors of commercial sunflower. They comprised less than 1% of all insects observed on receptive capitula (Du Toit & Holm, 1992a). R. Watmough (personal communication, 1987) states the reasons for the insignificance of indigenous solitary bees as pollinators are mainly because of the difficulty in establishing breeding colonies (low reproductive rates). Furthermore, their season of activity does not correspond with the main sunflower bloom in South Africa, which is from February to April.

Foraging honeybees and other insects discriminate between crops and even cultivars of the same crop when a choice is offered (Free, 1993). These preferences among cultivars are determined by quantity and/or quality of nectar (Burmistov, 1965; Vansell, 1934) as well as accessibility of the nectar and attractiveness of capitula (Cirnu, Dumitranche & Hociota, 1974; Shein, Sargent & Miko, 1980). These factors indirectly influence pollination These cultivar characteristics were and the eventual yield. reported on for seventeen South African sunflower cultivars by Du The sugar value, determined by nectar Toit & Coetzer (1992). and percentage sugar in volume solids the nectar, was significantly different among the cultivars. Significant differences in corolla length were also measured between cultivars. Long corollas prevent honeybees from reaching all the nectar in certain cultivars. Nectar was generally inaccessible to short-tongued solitary bees. Pigmentation of the corolla tube did not differ significantly. Pigmentation of the stigma did differ significantly but was considered a possible discriminating factor in one cultivar only.

## 1.5. Honeybees as pollinators of sunflower

The role of honeybees as pollinators of sunflower is well established. Researchers quoted in the previous section all agree that the honeybee must be considered the major pollinator of cultivated sunflower. Two reasons are given for this fact. First, honeybees are relatively domesticated as they are kept in manageable hives that can be brought into a extensive monoculture otherwise too large for sufficient pollination. Second, the areas planted to sunflower are usually intensively cultivated, where agricultural activities have led to a reduction in natural vegetation and therefore also the local honeybee and solitary bee populations. In South Africa the indigenous African honeybee should be regarded as the most important pollinator of commercial sunflower (Birch et al., 1985).

Du Toit & Holm (1992 a+b) studied honeybee activity and foraging behaviour in commercial sunflower fields at Settlers, 100 km north of Pretoria. Foraging activity was studied by means of the conventional strip count method. It commenced after sunrise and reached a peak of 38 foragers per 100 capitula at 10:00. Α moderate decline followed between 11:00 and 15:00 with a mean number of 32 foragers per 100 capitula. Thereafter, another foraging peak was reached two hours before sunset. Examination of foragers returning to the hive revealed that pollen foraging foraging prevailed until 09:00 and nectar predominated afterwards. Cross-pollination of honeybee foragers was efficient as more than 50% landed on the outer ring of florets. They then moved to the inner ring where fresh pollen was available, before flying to the next head. Movement between heads was indiscriminate, which enhanced pollen movement.

# 1.6. Economics of sunflower production in South Africa

Commercial sunflower cultivation in South Africa before World War II was entirely for seed production as poultry feed. After the war a worldwide shortage of vegetable oils led to the development of sunflower cultivars with a high oil content. Worldwide, the production of sunflower seed as a source of vegetable oil resulted in a decline in the use of sunflower seed as food. In 1974 South Africa followed the world trend of using hybrid cultivars that originated in the early 1970's in the United States (Birch & Engelbrecht, 1978). Today more than 40 hybrid cultivars are available on the South African market, developed to suit a wide range of climatic conditions.

In spite of this variety of cultivars available, commercial sunflower planting throughout South Africa is not extensive. Most sunflowers were planted only when weather conditions do not favour the cultivation of other fixed-price crops. In South Africa, sunflower is a short-season crop and can therefore be planted late in the growing season. Before 1983 the country was self-sufficient in its local demand for sunflower seed. However, since 1983 demands could not be satisfied, mainly because of drought. Van Zyl (1985) predicted a dependance on imports, based on the previous record harvest of 517,000 tons in 1982. The local demand in sunflower seed showed a steady increase from 240,000 tons in 1977 to 430,000 tons in 1986. The 1998 crop of

just below 570,000 tons was one of the biggest in the last ten years. A 35% increase (800,000 tons) in the 1999 crop is expected, based on the total area planted, National Crop Estimates Committee (1998). The current annual demand is estimated at 900,000 tons. This shortage therefore requires an increase in production per unit area as well as in the total area planted. New oil seed crops, such as canola, may well find favour with crop producers and will contribute to eliminate the shortfall in vegetable oil.

The aim of this study was to investigate seed set of commercial sunflower in South Africa by researching certain plant characteristics. The occurrence of hollow seededness and papery seeds as contributing factors to poor seed set was investigated at different localities, within different sunflower cultivars and at different plant densities. The possible influence of pollen quality on seed set was also examined. Lastly recommendations for the improvement of seed set were made.

# CHAPTER 2

# INVESTIGATION INTO SEED SET OF COMMERCIAL SUNFLOWER CULTIVAR SO323 AT SEVENTEEN DIFFERENT LOCALITIES IN SOUTH AFRICA

## 2.1. Introduction

Poor yields from commercial sunflower in South Africa were obtained during the period 1981 to 1984 (Birch, Van der Sandt, Herrmann & Johannsmeier, 1985). This was attributed to a lack of sufficient pollinating insects in large areas of monoculture, sometimes in access of 2000 ha, and the occurrence of hollow seededness in some years. Less than three honeybees per 100 flowering heads were observed. The activity and behaviour of pollinating insects on sunflowers, with special references to the role of honeybees, were well researched from 1985 to 1988 (Du Toit, 1990; Du Toit & Coetzer, 1992; Du Toit & Holm, 1992 a+b). Du Toit (1990) demonstrated the importance of indigenous honeybees and spotted maize beetles in sunflower pollination using insect-proof cages, simultaneously proving the insignificant role in pollination played by house flies and

American bollworm larvae. This was complemented with studies on the general abundance of honeybees and their activity in sunflower fields, establishing their importance beyond doubt (Du Toit & Holm, 1992 a+b). The morphological attractiveness of the different sunflower cultivars was further researched and found not be a limiting factor to attract insect pollinators under present cultivation practices in South Africa (Du Toit & Coetzer, 1992). I frequently recorded hollow seededness of between 20 and 40% during 1987 in the North West Province. Since sufficient numbers of honeybees - a mean of 26 honeybees per 100 capitula was recorded, poor pollination could not be blamed for the hollow seededness. A range of factors influence the eventual seed set and yield in sunflower, which is controlled by the vital but separate processes of flowering initiation, pollination and fertilization (Birch, 1981). Birch (1981) and Herring (1981) discussed the following factors influencing yield: climate, cultivar characteristics, insect pollinators and plant nutrition.

The aim of the present study was to determine the extent and nature of occurrence of hollow seededness and unfilled seeds in a commercial sunflower cultivar at different localities in South Africa.

### 2.2. Methods

# 2.2.1. Description of National Sunflower Cultivar Trials

Permission was obtained from the Grain Crops Research Institute, Potchefstroom, to make use of the National sunflower cultivar trials that are conducted annually at various localities in the sunflower producing areas of South Africa. The main objective of these trials is to supply the producer with reliable information on the performance of the commercially available cultivars (Loubser, Grimbeek & Steinmann, 1990). The extent of these trials therefore ideally suited the present investigations because all other climatic and cultivar data would be available for analysis.

The National sunflower cultivar trials were planted using conventional hand planting techniques and thinning to the required stand. The trials were planted in the various sunflower producing areas of Gauteng, Northern Province, North-West Province, Mpumalanga and Free State (Tables 1 and 2), from November 1989 to January 1990 with some late plantings (Loubser, et al., 1990). The density varied from 22000 to 78000 plants per hectare for dryland trails and 56000 to 78000 plants per hectare for irrigated trials (Table 2). Fertilizer was applied so that plant nutrition could not be considered as a factor limiting yield (Table 3). Weeds were effectively controlled by using

either herbicides, hand hoeing or mechanical means (Table 2). No disease control was practised. The trials were protected from bird damage, either by planting in birdproof cages, or covering the harvest rows with plastic netting, or planting the experiment in large sunflower fields and harvesting as soon as possible.

The cultivars were planted in a randomized block design with 19 different cultivars and three replicates at each locality. Six rows were planted per plot. The spacing and nett plot sizes are shown in Table 2. Agronomic and climatic data were recorded and collated by the Grain Crops Research Institute and are summarized in Tables 2, 3 and 4. This included uniformity of emergence, numbers of days from planting to 50 percent flowering, plant height, off-types and male sterility, stalk and root lodging and final density. Seed was harvested by hand after physiological maturity to determine yield and seed mass (Loubser, et al., 1990).

## 2.2.2. Occurrence of insect pollinators

All insects visiting the sunflower cultivar SO323 were recorded per 100 flowering capitula at these localities to determine insect diversity and their abundance. The conventional strip count method was followed, as described by Free (1964) and Parker (1981). Species diversity and abundance were determined for randomly-selected plant rows. During counting, capitula from the adjacent row were inspected, at a distance of 0.5-0.7 m, so as

not to disturb the insects. At each locality each replicate served as one count of 100 open capitula, taken between 09:00 and 11:00 at 40 to 60% flowering stage. This was found by Du Toit & Holm (1992b) to coincide with peak daily and seasonal honeybee activity in commercial sunflower fields, as well for being the most favourable time of day for successful pollination to occur.

## 2.2.3. Boron status of soil, leaves and florets

Boron fertilizer as Solubor was applied as soil application or foliar application (Table 6). To determine top soil boron status, a soil sample of approximately one kg soil was obtained after removal of the first 300 mm of loose soil with a shovel. The soil sample was placed in a plastic bag because other containers, e.g. metal and glass easily leech boron. After drying of the soil sample, its boron status was determined according to the ethanol extraction procedure for soil samples (Bessinger, 1988) by the Institute for Soil, Climate and Water, Private Bag X79, Pretoria, 0001.

The second sunflower leaf from the top was hand picked from five randomly selected sunflower plants of the cultivar SO323 to determine the leaf boron status. This was done for each three replicates at the seventeen localities under investigation. To determine the floral boron status, about 200 florets were removed by hand from five capitula of each of the three replications. These florets were from the area on the capitulum next to, but

outside, the newly opened florets, i.e. they represented florets that most probably had been pollinated and where fertilization should already had occurred 24 hours earlier.

Leaf and floret samples from the three replicates at each locality were pooled. Samples were air-dried at room temperature for several days during which time they were turned around by hand to prevent moulding due to excessive moisture. When they were cracking dry, the samples were milled to powder with a handoperated grinder to obtain 30 g of leaf and floral powder. The boron content of the samples was determined by the Institute for Soil, Climate and Water using the ethanol extraction method for plant material (Bessinger, 1988). The results are presented in table 6.

# 2.2.4. Determination of seed set and hollow seeds and papery seeds

To investigate the occurrence and extent of hollow seededness at the different localities, the sunflower cultivar SO 323 was used as standard cultivar, as it was considered one of the most popular among farmers. Ten mature sunflower heads with a diameter of not less than 120 mm were harvested by hand from each of the three replications. Threshing was done by hand and after drying, the achenes were separated by hand into filled, hollow and under-developed (dull or papery) achenes. Filled achenes

consisted of the oil-bearing seed or kernel and the surrounding pericarp. Hollow achenes developed a normal pericarp, which was occasionally smaller than the filled ones, but containing no seed. These achenes also had a much lower mass than filled achenes and are usually blown out with the chaff during mechanical harvesting. Under-developed achenes consisted of a papery under-developed pericarp only, with no seed. The percentage of filled, hollow and under-developed achenes was calculated. Results are given in table 7.

## 2.2.5. Statistical analysis

A Kruskal-Wallis analysis of variance with random replication was carried out for the following variables: head diameter, percentage hollow seeds, percentage papery (dull) seeds, percentage filled seeds (seed set), and 1000 seed mass. The degrees of freedom were: 16 localities with 3 replicates. The median, inter quartile range and class total were calculated. The Chi-Squared value and an adjusted Chi-Squared value were calculated to test whether differences existed among treatments. Pairwise comparisons of the treatments (different localities) were done on these variables using Dunn's method.

Statistical analysis was performed using the GENSTAT statistical analysis programme (Alvey, Galvey & Lane, 1982) on a Burroughs computer.

Localities	Where conducted						
Dwaalboom	On the farm Stellenbosch of Mr J Botes						
Frankfort	Free State Co-op, Frankfort						
Heilbron	On the farm Vlakkuil of Mr G Welse						
Immerpan (2 trials)	On the farm Rietkuil of Mr Z van der Bank						
Kempton Park(2trials)	Hartbeesfontein Research Farm						
Klerksdorp	Pioneer Research Farm, Klerksdorp						
Koster	On the farm Loraine of Mr J Theron						
Lichtenburg AS	On the farm of Dr AJL du Toit						
Potchefstroom	Oil and Protein Seed Centre						
Potgietersrus	Northern Transvaal Co-op Research Farm						
Petrus Steyn	Free State Co-op, Petrus Steyn						
Vaalharts	Vaalharts Research Station						
Vermaas	Central-West Co-op Res. Farm, Sterkstroom						
Warm Baths (2 trials)	Towoomba Research Station						

Localities	Planting date	Population (x1000)	Spacing (cmxcm)	Nett plot size (m <sup>2</sup> )	Weed control
Dwaalboom	90-02-05	33	100 x 30	40.0	Hand hoeing
Frankfort	89-11-02	-	-	18.0	Lasso
Heilbron	89-11-20	37	90 x 30	28.8	Lasso, Hand hoeing
Immerpan (Early)	89-11-23	30	-	38.0	Hand hoeing
Immerpan (Late)	90-01-17	30	-	38.0	Hand hoeing
Kempton Park (Early)	89-10-17	56	90 x 20	36.0	Lasso, mechanical
Kempton Park (late)	89-12-15	56	90 x 20	36.0	Lasso, mechanical
Klerksdorp	89-12-28	24	90 x 45	39.6	Hand hoeing
Koster	89-12-07	32	90 x 35	23.9	Hand hoeing
Lichtenburg AS	89-12-24	56	91 x 20	8.0	Treflan, mechanical
Potchefstroom	89-12-19	56	90 x 20	25.2	Lasso, Hand hoeing
Potgietersrus	89-12-20	30	-	38.0	Hand hoeing
Petrus Steyn	89-11-17	30	100x27.7	40.0	Dual
Vaalharts	89-12-06	56	90 x 20	36.0	Lasso
Vermaas	90-01-29	37	90 x 30	25.2	Lasso, Hand hoeing
Warm Baths (Early)	89-12-22	31	100 x 32	35.8	Hand hoeing
Warm Baths (Late)	90-03-02	31	100 x 32	35.8	Hand hoeing

Table 2Production practices used at the different localities

**Table 3** Soil classification, topsoil analysis and fertilizer used at the different localities

			То	. <u></u>				
Localities	Soil classi- fication	рН (H <sub>2</sub> 0)	P ppm	K	Ca ppm	Mg ppm	Na ppm	rtilizer
Dwaalboom	Arcadia	7.2*	3	271	6000+	1500+	24 None	
Frankfort	Westleigh	4.0*	31	155	348	56	12P	+ Lime
Heilbron	Arcadia	5.5*	15	176	514	160	- 25N	
Immerpan (early)	Arcadia	-	-	-	-	-		
Immerpan (late)	Arcadia	-	-	_	-	-		
Kempton Park (early)	Hutton	5.6	13	105	728	243	- 4N,	7P, 4K
Kempton Park (late)	Hutton	5.8	15	111	733	251	- 4N,	7P, 4K
Klerksdorp	Hutton	6.5*	33	171	812	327	12 92N	
Koster	Hutton	5.8	25	280	440	89	- 35N,	8P, 4K +8
Lichtenburg AS	Avalon	-	-	-	-	-	- 17N,	11P
Potchefstroom	Hutton Shorrocks	6.6	50	74	314	99	9 90H, + 8	36P, 14K
Potgietersrus	Hutton	-	-	_	-	-		
Petrus Steyn	Bainsvlei	4.5*	35	162	410	85	- 29N,	19P + 8
Vaalharts	Hutton Mangano	-	-	-	-	-		
Vermaas	Hutton	5.7	28	299	654	138	6 48N,	25P
Warm Baths (early)	Arcadia	6.5	19	420	9 260	1 150	- None	
Warm Baths (late)	Arcadia	6.5	19	420	9 260	1 150	- None	

\* pH (KCl)

Table 4Rainfall and irrigation information for the differenttrial localities, 1989/90

	Monthly rainfall (mm)								_		
Localities	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Total	Irrigation	Total
Dwaalboom	0	111	161	111	124	53	105	20	685	0	685
Frankfort	60	100	52	65	68	120	85	0	550	0	550
Heilbron	44	115	134	92	74	111	78	0	648	0	648
Kempton Park	47	160	70	64	118	54	109	8	630	0	630
Klerksdorp	26	136	99	54	48	100	0	0	463	63	526
Lichtenburg AS	90	106	86	41	33	75	76	0	507	0	507
Petrus Steyn	83	87	41	34	80	116	47	7	495	0	495
Potchefstroom	0	147	79	47	163	62	106	8	612	75	687
Vaalharts	0	42	54	36	74	45	84	2	337	200	537
Vermaas	17	97	115	51	59	133	88	16	576	0	576
Warm Baths	33	129	206	44	94_	114	38	10	668	25	693

#### 2.3. Results

## 2.3.1. Occurrence of insect pollinators

No hived honeybee colonies were introduced to the study sites to enhance the number of insect pollinators. Twenty four honeybees per 100 flowering capitula are regarded as the minimum number of pollinators required for satisfactory pollination and seed set to occur in sunflowers. This number was not met at five of the seventeen localities under investigation (Table 5). The lowest number of honeybees was recorded at Petrus Steyn and Frankfort with a mean of only 2 and 3 honeybees per 100 flowering capitula respectively. At Dwaalboom and Immerpan (early and late) means of 22, 20 and 21 honeybees respectively were recorded per 100 capitula. The highest number of honeybees was counted at Vermaas namely 314 honeybees per 100 capitula. More than 100 honeybees capitula were also observed at Potchefstroom, 100 per Potgietersrus and Warm Baths (early), with respective means of 113, 144 and 148 honeybees per 100 flowering capitula (Table 5).

Spotted maize beetles, which are also highly regarded as a pollinator of commercial sunflower, was frequently observed (Table 5). Beetle numbers exceeded 1,000 beetles per 100 capitula at Frankfort (1226), Immerpan (late) (2997), Kempton Park (early) (3984), Klerksdorp (1256), Lichtenburg (1066) and Petrus Steyn (2843). No spotted maize beetle was recorded at Dwaalboom. The lowest number of spotted maize beetles was

recorded at Immerpan (late) with a mean of 37 beetles per 100 flowering capitula.

The number of other insects, including solitary bees, were low and therefore probably contributed little to the sunflower pollination (Table 5). Their numbers were below an average of 10 insects per 100 capitula, except for two localities. Twenty and 93 other insects were encountered at Vermaas and Potgietersrus respectively. These insects were mostly solitary bees of the family Halictidae.

### 2.3.2. Boron status of soil, leaves and florets

Different agronomical practices were followed at the various localities concerning boron application (Table 6). These varied from no boron application at all, to soil application and one or two leaf applications prior to flowering. The amount of free boron in the soil ranged from a low of 0.33 mg.kg<sup>-1</sup> at Vaalharts to the highest level of 1.76 mg.kg<sup>-1</sup> at Heilbron (Table 6). These levels were, however, not correlated with a boron application or the mode of application, as the highest boron levels were encountered at Heilbron where no boron was applied. Furthermore, they also did not correlate with soil type, as illustrated in Hutton soils where they ranged from the lowest level of 0.33 mg.kg<sup>-1</sup> at Vaalharts to moderately high levels of 0,98 mg.kg<sup>-1</sup> at Potchefstroom. Arcadia soils generally showed higher levels of boron (Table 6).

Leaf analysis revealed that boron content did not necessarily correspond with the additionally applied boron, either through fertilizer or other application (Table 6). The lowest boron levels in the leaf samples were found at Frankfort and Kempton Park (early) with 27.3 mg.kg<sup>-1</sup> and 29.5 mg.kg<sup>-1</sup> respectively. This did not correlate with either mode of application or soil type. At Kempton Park (early and late), two foliar treatments of Solubor that were applied, did not reflect in any of the tested the boron levels (Table 6). The highest leaf boron levels were obtained from plantings on Arcadia soils, with figures as high as 175.0 mg.kg<sup>-1</sup>.

Boron levels in florets showed lesser variation compared with leaves (Table 6). They ranged from 47.8  $mg.kg^{-1}$  at Petrus Steyn to 72.0  $mg.kg^{-1}$  at Koster. Again this did not correspond with the mode of application or whether any additional boron was applied.

Boron levels in leaves and florets could not be correlated with the percentage seed set (Table 6).

### 2.3.3. Seed set and hollow seededness

#### 2.3.3.1. Head diameter

Head diameter was used as a parameter to ascertain variation between localities and the replicates at each locality. The inter quartile range within the treatments (localities) was low

which suggests high homogeneity (Table 8). A Chi-Squared value of 259.2215 was calculated. The largest heads were found at Kempton Park (late) (235 mm diameter) and the smallest at Dwaalboom (135 mm diameter). Table 9 shows the level of significance based on paired comparison among the seventeen localities investigated.

#### 2.3.3.2. Hollow seededness

Little heterogeneity was found amongst the replications at each locality for the parameter of percentage hollow seededness as illustrated by the low interquartile range (Table 10). A Chi-Squared value of 124.6083 was calculated. Hollow seededness of 5% and above was found at four localities, i.e. Vermaas (5.05%), Kempton Park (late) (6.35%), Immerpan (early) (6.50%) and Koster (7.70%). Percentage hollow seededness of above 4% was found at a further five localities. The lowest level of hollow seededness (1.80%) was found at Kempton Park (early). Table 11 reveals the level of significance for paired comparisons among the seventeen localities studied, using Dunn's method.

#### 2.3.3.3. Papery seeds

The percentage papery (dull or under-developed) seed was highly heterogenous within the replications at the seventeen localities as indicated by the high interquartile range (Table 12). This intra-locality heterogeneity was highest at Dwaalboom (12.10%) and was also high at Frankfort (8.65%) and Vermaas (6.40%), while being almost uniform at Vaalharts (1.55%). A Chi-Squared value of 205.7843 was calculated. More than 10% papery or underfifteen of the seventeen developed seeds were found at localities. Percentages of above 20% papery seeds were found from three localities, Warm Baths (late), Frankfort and Dwaalboom with 20.65%, 35.75% and 42.45% papery seed respectively. The lowest percentage papery seeds was found at Warm Baths (early) and Vaalharts (7.15% and 9.35% respectively). Table 13 reveals the level of significance for paired comparisons among the seventeen localities studied using Dunn's method. Dwaalboom and Frankfort with high percentages of papery seeds and Warm Baths (early) and Vaalharts with low levels of papery seeds differ significantly from most other localities (Table 13).

#### 2.3.3.4. Filled seeds

Some intra-locality variance is shown when the percentage filled seeds (equal to seed set) is analysed (Table 14). Replicates homogeneous at Vaalharts (1.55%) almost and were most heterogenous at Dwaalboom (10.70%). The average seed set ranged from as low as 52.5% at Dwaalboom to 88.3% at Warm Baths (early). The percentage seed set was above 85% in only two localities (Warmbaths [early] - 88.3% and Vaalharts - 86.0%). It was above 80% at another seven localities and above 75% at a further six localities. A Chi-Squared value of 189.3600 was calculated. Comparison in pairs of the level of significance among the seventeen localities is illustrated in Table 15 using Dunn's method. Dwaalboom and Frankfort differed significantly in the

number of filled seeds from most other localities.

#### 2.3.3.5. Seed mass

Variance in the 1000 seed mass between replications at the seventeen localities investigated is very evident from the wide inter quartile range (Table 16). This was more than 5% for all but two localities (Frankfort - 4.25% and Kempton Park early - 4.75%) with a highest variance of 13.45% at Kempton Park (late). A 1000 seed mass of above 60 g was recorded at eight localities with the highest at Kempton Park (late) (78.9 g) (Table 16). At a further five localities the 1000 seed mass was found to be above 50 g. At Dwaalboom, Petrus Steyn, Frankfort and Vermaas the 1000 seed mass was below 50 g, being 48.20 g, 47.80 g, 47.15 g and 44.30 g respectively (Table 16). A Chi-Squared value of 201.6995 was calculated (Table 16). Dunn's method of paired comparisons was used to illustrate the level of significance of 1000 seed mass for the seventeen localities investigated (Table 17).

## 2.4. Discussion

It is believed that the numbers of pollinators at the seventeen localities studied was generally sufficient for satisfactory pollination to have occurred. Where the number of honeybees was low, this was complemented with high numbers of spotted maize beetle (Table 5). Seed set was significantly lower at Dwaalboom and Frankfort where insect abundance was also recorded to be very low. A correlation could therefore be drawn between unsatisfactory seed set and the number of pollinators. However, the percentage hollow seeds found at the different localities could not be attributed to low numbers of insect pollinators.

Sunflowers have a high boron requirement but little is known about the quantity of boron necessary in the plant tissue under field conditions (Blamey, Mould & Nathanson, 1978). The most severe symptom of boron deficient plants was malformation of the capitulum that led to areas of no seed set. Boron deficiency and toxicity inhibit ATP activity in sunflower roots and cell suspensions (Ferrol, Belver, Roldan, Rodriquez-Rosales & Donaire, 1993). Though the boron content of soils ranges from 20 to 200 mg.kg<sup>-1</sup>, it remains unavailable to plants (Bessinger, 1988) as it is a constituent of tourmaline. Plants can only utilise the water soluble boron. This occurs mainly as boric acid, and its concentration in soils varies from 0.4 to 5.0 mg.kg<sup>-1</sup> (Bessinger, 1988).

Boron has an essential function in the successful completion of the life cycle of most plants. One function of this microelement is that it has an advantageous effect on the reproductive cycle of plants. It affected the reproductive processes of pollination (Dickenson, 1978), pollen tube growth (De Wet, Robbertse & Groeneveld, 1989; Robbertse, Lock, Stoffberg & Coetzer, 1990) and seed and fruit set (Coetzer, Robbertse,

Stoffberg, Holtzhausen & Barnard, 1990; Mengel & Kirkby, 1982). The stigma, style and ovaries have a rich supply of boron, which combines with pectin to form a pectin-borate complex that encourages pollen tube growth and also the pollination process (Vasil, 1964). It has been suggested that the pollen tube absorbs boron as it grows through the stigma. It was also demonstrated that pollen germination is dependant on the presence of satisfactory levels of boron in the stigma exudate (Schmucker, 1935). Various researchers have identified boron deficiency as one of the contributing factors in poor sunflower yields (Birch, 1981). Blamey, Mould & Chapman (1979) reported a 48% increase in sunflower production after boron application. Researchers working in India found that sunflower seed yield increased linearly with increasing levels of boronated superphosphate (Ateeque & Malewar, 1992; Ateeque, Malewar & More, 1993; Tamak, Sharma & Singh, 1997; Vasudevan, Virupakshappa, Venugopal & Bhaskar, 1997). Foliar application of boron with four different boron fertilizers resulted in an increase in seed yield with all treatments compared with the untreated control (Droba, 1993). Sarkar & Ghosh (1992) demonstrated that both a boron deficiency and toxicity condition would result in decreased sunflower yield.

Shorrocks (1984) demonstrated that flowers treated with boron are more frequently visited by honeybees. This was due to an increase in nectar secretion of the flowers after boron application. This make the flowers more attractive to bees and will enhance pollination.

In spite of these significant results the specific effect of boron in sunflowers is still not understood. Various questions still arise, e.g. what are the optimum requirements, regarding methods and stage of application and how much boron should be present in leaves and flowers. Blamey et. al. (1978) calculated the minimum boron level in the topmost, mature leaf at flowering to be 57 ppm on an Avalon medium sandy loam soil. The results obtained from the present study indicate that we know too little, especially of boron uptake and translocation in plants, to make any meaningful conclusion about the levels required to increase sunflower production. However, researchers have already established the importance of boron on dryland agronomical crops on sandy soils in South Africa (Van de Venter & Farina, 1972). Application of boron increased grain yield dramatically by 90%. Boron seems to have no effect up to the time of kernel formation, but only become evident during maturation. This was associated with a purple colouring of stems.

The physiology of hollow seededness is still unexplained. One possiblity is that where cross-pollination is inefficient, a high level of self-pollination could occur, resulting either in poor fertilization and/or the abortion of the seed after fertilization. It is likely that both these phenomena could Self-pollination in sunflowers occurs occur and contribute. where the bilobed stigma elongates and curls back far enough, after several days of no cross-pollination, to come into contact with its own pollen (Birch & Van der Sandt, 1985). Hollow seeds were dissected by L. Coetzer and J. Robbertse (personal

communication, 1992) and the remains of aborted embryo sacs were observed. These indicated that fertilisation did not take place, which could be related to interruption of pollen tube growth. The germinating pollen did, however, provide a stimulus for seed development. High levels of hollow seededness, between 12-24%, were reported in caged sunflower without insect pollinators (Birch <u>et al</u>, 1985; Du Toit, 1990). In adjacent, open pollinated fields this was found to be less than 5% (Du Toit, 1990). It must therefore be concluded that cross-pollination will reduce the incidence of hollow seededness. Slow or poor pollen movement which in the end would result in self-pollination, would lead to higher levels of hollow seededness.

Khanna (1972) reported that papery seeds (dull or under-developed seeds) in sunflowers are mainly caused by competition for water and nutrients. A physiological nutrient scarcity develops when good seed set is achieved. Abortion in the central, youngest florets is a general phenomenon in sunflower and results in poor or no seed set of these florets (Khadiikar & Mahajan, 1974). Herring (1981) reported that sterility in the centre of the head was normally overlooked when seed set was determined. Although precautions could be taken to ensure sufficient numbers of insect pollinators (honeybees) in sunflower fields, the level of dull seeds could still be above 20% (Du Toit, 1990).

## 2.5. Conclusions

- Sufficient numbers of pollinating insects were observed at the different localities, which excludes a lack of pollinators as reason for differences in seed set. These comprised mainly honeybees and spotted maize beetles. Solitary bees played an insignificant role in pollination due to their low numbers.
- Self-pollination, which results because of inadequate or to slow pollen transfer from other florets, will induce hollow seededness.
- 3. Boron levels differed significantly in soil samples, leaves and florets. Interpreting these results was difficult, suggesting that our knowledge on the interaction of boron in sunflowers is still lacking. The role of boron in the various physiological processes associated with anthesis and fertilization should be studied further.
- Flower head diameter differed significantly in the cultivar SO323 at different localities for the cultivar SO 323.
- 5. Seed set differed significantly when compared in pairs. The highest seed set was recorded at Warm Baths (86%) and the lowest at Dwaalboom (53%).

- 6. The percentage hollow seededness reached its highest levels of above 5% at three localities only, namely Immperpan (early) (7%), Kempton Park (late) (6%) and Koster (8%).
- 7. The percentage of papery seeds (dull or unfilled seeds) differed significantly from locality to locality. This parameter is the main cause for concern as it is much overlooked and severely reduces the yield potential of commercial sunflower.
- 8. The 1000 seed mass differed significantly between the studied localities. It showed no relationship with the number of filled seeds (seed set). It therefore confirms other researchers results that sunflower have the ability to compensate in seed mass when seed number is reduced.
- 9. Understanding the interaction between the different parameters which determine the potential yield in sunflower was difficult. Further investigation of the physiology of these processes is needed.

Table 5Mean number of insects per 100 flowering sunflowerheads recorded at the different localities during the 1989/90sunflower season

Locality	Honey-	Spotted maize	Other
	bees	beetle	insects
Dwaalboom	22	0	6
Frankfort	3	1226	3
Heilbron	48	419	3
Immerpan	20	2997	3
Immerpan	21	37	3
(irrigation)			
Kempton Park	46	3984	4
(early)			
Kempton Park	46	132	10
(late)			
Klerksdorp	88	1256	6
Koster	27	199	2
Lichtenburg As	24	1066	1
Potchefstroom	113	153	7
Potgietersrus	144	149	93
Petrus Steyn	2	2843	4
Vaalharts	38	166	2
Vermaas	314	309	20
Warm Baths	148	301	3
(early)			
Warm Baths (late)	98	172	6

**Table 6** Boron application, soil type, free boron in soil, boron in leaves and florets (mg.kg<sup>-1</sup>) and % seed set, at the different localities

Localities	Boron application	Soil type 1	Free boron in soil mg.kg <sup>-1</sup>		mg.kg <sup>-1</sup> Florets	
Dwaalboom	None	Arcadia	0.94	70.8	57.5	538
Frankfort	None	Westleigh	0.39	27.3	50.0	61%
Heilbron	None	Arcadia	1.76	130.0	64.7	77%
Immerpan (early)	None	Arcadia	1.14	175.0	67.0	79%
Immerpan (late)	Foliar	Arcadia	1.14	121.0	58.5	84%
Kempton Park (early)	2x Foliar	Hutton	0.66	29.5	58.7	81%
Kempton Park (late)	2x Foliar	Hutton	0.77	81.3	71.0	76%
Klerksdorp	None	Hutton	0.50	97.2	71.5	81%
Koster	Soil	Hutton	0.49	77.5	72.0	78%
Lichtenburg	None	Avalon	0.46	75.4	61.0	81%
Potchefstroom	Soil	Hutton	0.98	77.7	68.7	838
Potgietersrus	Soil	Hutton	0.52	88.4	49.8	848
Petrus Steyn	Soil	Bainsvlei	0.56	45.0	47.8	798
Vaalharts	None	Hutton	0.33	116.0	63.5	86%
Vermaas	None	Hutton	0.66	46.5	66.5	80%
Warm Baths (early)	None	Arcadia	0.76	120.5	61.2	88%
Warm Baths (late)	None	Arcadia	0.76	91.0	63.5	75%

Table 7 Mean head diameter (mm), number of filled, hollow and papery seeds, 1000 seed mass (g), % seed set and % hollow seeds in the sunflower cultivar SO 323 at seventeen different localities during the 1989/90 sunflower season

Locality	Head	Filled	Hollow	Papery	1000	<pre>% seed</pre>	Seed`	% hollow
	dia-	seeds	seeds	seeds	seed	set	Yield	seeds
	meter				mass		kg/ha	
	(mm)				(g)			
Dwaalboom	135	652	60	489	48.20	53	552	4
Frankfort	153	940	56	544	47.15	61	1924	3
Heilbron	190	1137	77	256	59.45	77	1908	5
Immerpan	172	1146	145	194	67.55	79	1131	7
(early) Immerpan (late)	190	1251	74	191	66.76	84	818	4
Kempton Park (early)	173	1425	49	283	52.25	81	2096	2
Kempton Park (late)	235	1588	144	352	78.90	76	2269	6
Klerksdorp	215	1600	86	290	66.75	81	928	4
Koster	205	1617	158	288	56.00	78	2437	8
Lichtenburg	165	1091	71	196	64.20	81	2487	5
Potchefstroom	187	1212	68	229	60.65	83	2628	4
Potgietersrus	195	1733	70	253	62.70	84	975	3
Petrus Steyn	155	1102	72	225	47.80	79	1188	5
Vaalharts	205	1512	93	170	66.30	86	3651	5
Vermaas	172	1200	87	263	44.30	80	1392	5
Warm Baths (early)	195	1434	96	168	56.10	88	2280	3
Warm Baths (late)	180	1434	96	168	52.90	75	1754	3

Table 8Kruskal-Wallis analysis of head diameter of thesunflower cultivar SO 323 at seventeen localities during the 1989/90season

			· · · · · · · · · · · · · · · · · · ·
Locality	Median	Inter quartile	Class total
•		range	
Dwaalboom	13.50	01.00	1089.50
Frankfort	15.35	01.15	3493.00
Heilbron	19.00	02.50	7476.00
Immerpan (Early)	17.25	01.75	6256.50
Immerpan (Late)	19.00	02.00	8219.00
Kempton Park	17.25	01.25	6185.00
(Early)			
Kempton Park	23.50	02.00	13303.50
(Late)			
Klerksdorp	21.50	01.75	11576.50
Koster	20.50	01.00	10695.50
Lichtenburg (AS)	16.50	01.75	5401.00
Potchefstroom	18.75	00.75	8868.00
Potgietersrus	19.50	02.75	9616.50
Petrus Steyn	15.50	01.00	3481.00
Vaalharts	20.50	01.00	10968.00
Vermaas	17.25	01.00	5714.00
Warm Baths (Early)	19.50	01.00	9813.50
Warm Baths (Late)	18.00	01.50	8148.50

Chi-Squared value = 259.2215 16 Degrees of freedom Adjusted Chi-Squared value = 260.0800 16 Degrees of freedom Table 9Significance for all pairwise comparisons amongseventeen localities for head diameter of the sunflower cultivar SO323, using Dunn's method.

Dw Fr He I(e) I(l) K(e) K(l) Kl Ko L AS Po Pg Pe Va Ve W(e) Fr He \* \* I(e) \*\* I(1) \*\* \*\* K(e) \*\* K(l) \*\* \*\* \*\* \* \* ++ Kl \* \* \* \* \* \* Ko \*\* \* \* \* \* L AS \* Ро \* \* Ρg \*\* \* \* \* Pe \*\* \*\* \* \* Va \* \* \* \* \* \* \*\* \* \* \* \* Ve \*\* \* \* + W(e) \*\* \* \* \* \* \* \*\* W(1) \*\* \*\* \*\*

\* : significant at the 5% level

\*\* : significant at the 1% level

Table 10Kruskal-Wallis analysis of the percentage hollowseeds in the sunflower cultivar SO 323 at seventeen localitiesduring the 1989/90 season

Locality	Median	Inter quartile	e Class total
		range	
Dwaalboom	3.65	2.05	6705.00
Frankfort	2.75	1.15	5598.00
Heilbron	4.55	1.75	8504.00
Immerpan (Early)	6.50	3.65	11324.50
Immerpan (Late)	4.30	1.85	8067.50
Kempton Park	1.80	0.30	3365.00
(Early)			
Kempton Park	6.35	1.95	10847.50
(Late)			
Klerksdorp	3.90	0.95	7531.50
Koster	7.70	2.70	11381.00
Lichtenburg (AS)	4.55	1.20	8985.00
Potchefstroom	3.85	1.30	6398.50
Potgietersrus	3.15	1.05	5280.00
Petrus Steyn	4.70	1.70	7704.00
Vaalharts	4.55	1.45	8731.00
Vermaas	5.05	2.05	8462.50
Warm Baths (Early)	2.90	2.15	6224.50
Warm Baths (Late)	2.65	0.75	5168.50

Chi-Squared value = 124.6083 16 Degrees of freedom Adjusted Chi-Squared value = 124.6388 16 Degrees of freedom Table 11Significance for all pairwise comparisons amongseventeen localities for percentage hollow seeds in the sunflowercultivar SO 323, using Dunn's method

	Dw	Fr	Не	I(e)	I(l)	K(e)	K(l)	Kl	Ko I	_ AS	Ро	Pg	Pe	Va	Ve	W(e)	
Fr																	
Не																	
I(e)	**	**															
I(l)																	
K(e)			**	**	**		* *										
K(l)	*	* *															
Kl						*											
Ko	**	* *				**											
L AS	5					**											
Ро				**			*		**								
Pg				**			**		**								
Pe						*											
Va						* *											
Ve						* *											
W(e)				**			**		**								
W(1)				* *			**		**								

\* : significant at the 5% level

\*\* : significant at the 1% level

Table 12Kruskal-Wallis analysis of the percentage papery(dull) seeds in the sunflower cultivar SO 323 at seventeenlocalities during the 19989/90 season

			·
Locality	Median	Inter quart	ile Class total
		range	
Dwaalboom	42.45	12.10	13562.50
Frankfort	35.75	08.65	13419.00
Heilbron	17.25	04.15	8721.50
Immerpan (Early)	12.30	02.70	6071.50
Immerpan (Late)	10.75	03.50	5238.50
Kempton Park	15.35	02.90	8117.00
(Early)			
Kempton Park	18.15	04.25	8689.50
(Late)			
Klerksdorp	14.65	02.80	7319.00
Koster	13.15	03.35	6597.50
Lichtenburg (AS)	14.55	02.20	7141.50
Potchefstroom	14.70	03.30	7437.00
Potgietersrus	12.10	04.10	5510.00
Petrus Steyn	14.55	04.80	7585.50
Vaalharts	09.35	01.55	3092.50
Vermaas	14.55	06.40	7715.50
Warm Baths (Early	) 07.15	03.15	3337.50
Warm Baths (Late)	20.65	04.65	10749.50

Chi-Squared value = 205.7843 16 Degrees of freedom Adjusted Chi-Squared value = 205.7923 16 Degrees of freedom

Dw Fr He I(e) I(l) K(e) K(l) Kl Ko L AS Po Pg Pe Va Ve W(e) Fr He \*\* \*\* I(e) \*\* \*\* I(l) \*\* \*\* K(e) \*\* \*\* K(l) \*\* \*\* Kl \*\* \*\* Ko \*\* \*\* L AS \*\* \*\* Ро \*\* \* \* Ρq \*\* \* \* \*\* \*\* Pe Va \*\* \*\* \*\* + + Ve \* \* \*\* \*\* W(e) \*\* \*\* \* \* ++ \*\* \* \* \* W(1) \*\* \*\* \* \* \* \* \*

Table 13Significance of all pairwise comparisons among seventeenlocalities for percentage papery (dull) seeds in the sunflowercultivar SO 323, using Dunn's method

\* : significant at the 5% level
\*\* : significant at the 1% level

Table 14Kruskal-Wallis analysis of the percentage filledseeds (seed set) in the sunflower cultivar SO 323 at seventeenlocalities during the 1989/90 season

Locality	Median	Inter quartile	Class total
		range	
Dwaalboom	52.50	10.70	1787.50
Frankfort	61.40	07.55	2115.00
Heilbron	76.85	04.25	6627.00
Immerpan (Early)	79.10	06.35	7185.00
Immerpan (Late)	84.40	02.65	10105.00
Kempton Park	81.50	04.80	8927.50
(Early)			
Kempton Park	75.60	04.95	6047.00
(Late)			
Klerksdorp	81.50	03.15	8575.00
Koster	78.50	05.15	7280.50
Lichtenburg (AS)	80.95	02.75	8327.50
Potchefstroom	82.95	05.90	8616.00
Potgietersrus	83.70	05.25	10513.00
Petrus Steyn	79.50	04.10	7670.00
Vaalharts	86.00	01.55	11628.00
Vermaas	80.00	06.80	7554.50
Warm Baths (Early)	88.35	04.45	11704.00
Warm Baths (Late)	75.30	05.35	5642.50

Chi-Squared value = 189.3600 16 Degrees of freedom Adjusted Chi-Squared value = 189.3668 16 Degrees of freedom

Table 15Significance for all pairwise comparisons among seventeenlocalities for percentage filled seeds (seed set) in the sunflowercultivar SO 323, using Dunn's method

	Dw	Fr	Не	I(e)	I(1)	K(e)	K(l)	Kl	Ko	L AS Po	Pg Pe	Va	Ve W(e)	
Fr														
Не	**	*												
I(e)	**	* *												
I(1)	**	**												
K(e)	**	**												
K(1)	*													
Kl	**	**												
Ko	**	**												
L AS	; **	**												
Ро	**	**												
Pg	**	**					*							
Pe	**	**												
Va	**	**	**	*			**		*					
Ve	**	**										*		
W(e)	**	**	**	*			**		*				*	
W(1)					*						**	**	* *	

\* : significant at the 5% level

\*\* : significant at the 1% level

Table 16Kruskal-Wallis analysis of 1000 seed mass (g) in thesunflower cultivar SO 323 at seventeen localities during the1989/90 season

Locality	Median	Inter quartile	e Class total
	·····	range	
Dwaalboom	48.20	06.55	4243.50
Frankfort	47.15	04.25	3108.50
Heilbron	59.45	06.90	7651.00
Immerpan (Early)	67.55	07.90	10842.00
Immerpan (Late)	66.76	09.70	11104.00
Kempton Park	52.25	04.75	5221.00
(Early)			
Kempton Park	78.90	13.45	11808.00
(Late)			
Klerksdorp	66.75	07.75	10533.00
Koster	56.00	05.05	6551.00
Lichtenburg (AS)	64.20	05.70	9515.50
Potchefstroom	60.65	05.70	8877.50
Potgietersrus	62.70	09.50	8654.00
Petrus Steyn	47.80	08.95	4226.50
Vaalharts	66.30	05.35	10546.00
Vermaas	44.30	05.95	3548.50
Warm Baths (Early)	56.10	06.65	7395.50
Warm Baths (Late)	52.90	07.30	6479.50

Chi-Squared value = 201.6995 16 Degrees of freedom Adjusted Chi-Squared value = 201.7026 16 Degrees of freedom Table 17Significance for all pairwise comparisons amongseventeen localities of the 1000 seed mass (g) in the sunflowercultivar SO 323, using Dunn's method

	Dw	Fr	He	I(e)	I(1)	K(e)	K(l)	Kl	Ko	L AS	5 Po	Pg	Pe	Va	Ve V	W(e)
Fr																
Не		**														
I(e)	**	**														
I(l)	**	* *														
K(e)				* *	* *											
K(l)	**	**	*			**										
Kl	**	**				**										
Ko				*	**		**									
L AS	5 **	* *				*										
Ро	**	**														
Pg	*	* *														
Pe				* *	**		**	**		* *	**	*				
Va	**	* *				**							**			
Ve			*	**	**		**	**		**	**	* *		**		
W(e)		*					*									
W(l)				*	* *		**									

\* : significant at the 5% level
\*\* : significant at the 1% level

# CHAPTER 3

# INVESTIGATION INTO SEED SET IN EIGHTEEN SUNFLOWER CULTIVARS

#### 3.1. Introduction

Intensive sunflower breeding and selection, and competition between different seed houses, resulted in the availability of many different commercial sunflower cultivars on the South African market. These cultivars differ in their general plant characteristics such as growth period (calculated as number of days to 50% flowering), plant height, resistance to disease and to lodging, head diameter, seed mass and oil and protein content. All these factors, together with environmental conditions, determine the eventual seed yield.

Yield in sunflower is usually determined in agronomical terms as kilogram seed obtained per hectare. No previous studies have attempted to express the yield (seed set) in relation to seed loss through hollow seeds and unfilled seeds. Various researchers did however, recognise one or both of these factors as contributing to poor yield (Birch, 1981; Herring, 1981; Jain, Vaish, Gupta & Mathur, 1978; Khadiikar & Mahajan, 1974; Khanna, 1972).

The aim of the present study was to determine the extent and nature of the occurrence of hollow seededness and unfilled seeds and its influence on crop yield in different sunflower cultivars at Warm Baths, South Africa.

#### 3.2. Methods

#### 3.2.1. Description of trial and study area

The National Sunflower Cultivar trial at Towoomba was used to study seed set at a single locality amongst eighteen commercially available South African sunflower cultivars. Towoomba, near Warm Baths, was chosen as study locality as it is situated in the Springbok Flats that is one of the major sunflower producing areas in South Africa. With its favourable climate and deep black Arcadia soil ('turf'), crops are cultivated mainly under dry-land conditions with a small percentage under flood- or sprinkler-irrigation.

The general weather the Springbok Flats experienced from the 1970 to 1985 sunflower seasons is presented in figures 1 to 3. Data were summarized from the South African Weather Bureau Report on Meteorological Data (1970 - 1977) and the Monthly Weather Report (1978 - 1985) of the Towoomba Research Station (24"54'S, 28"20'E; altitude 1143m).

In general the Springbok Flats experience hot summers, as it is

protected by the Highveld from cold winds. Mild, dry winters are experienced, although the temperature can be minus 7°C on winter mornings. The mean annual rainfall is 622 mm. Heavy downpours occur, usually accompanied by thunder storms. In late spring hail can be severe. The region is almost frost free, with frost only on irrigated lands.

The eighteen sunflower cultivars were planted on 22 December 1989 in a randomized block design with three replicates. The spacing was 100 cm x 32 cm with a nett sub plot size of 35.8  $m^2$  and a resultant final plant population of 31000 plants per ha. The Arcadia soil at Towoomba had a pH of 6.5 and top soil analysis revealed the presence of minerals to be 19 ppm P; 420 ppm K; 9260 ppm Ca, 1150 ppm Mg; 0.76 mg.kg<sup>-1</sup> boron and no Na. No additional fertilizer, including boron was applied after top the soil had been analysed. Weed control was done by means of hand hoeing. The number of days to 50 percent flowering, mean plant height, percentage male sterile plants and percentage root and stem lodging in each of the cultivars were recorded. The above agronomical data were recorded and collated by the Grain Crops Research Institute and are summarized in Table 18 (Loubser, Grimbeek & Steinmann, 1990).

Insect pollination was considered adequate as abundant numbers of pollinating insects were present. Means of 400 honeybees, 301 spotted maize beetles and 3 other insects per hundred capitula were recorded on the sunflower capitula (Table 5, Chapter 2). No managed bee hives were introduced.

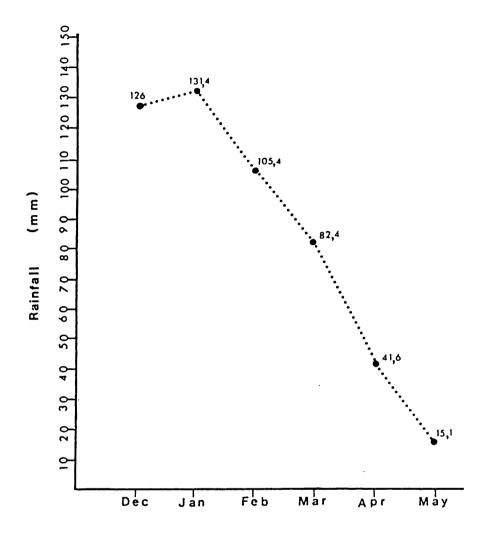


Figure 1 Mean monthly rainfall at Towoomba Research Station during 1970 to 1985 for the six months sunflower season

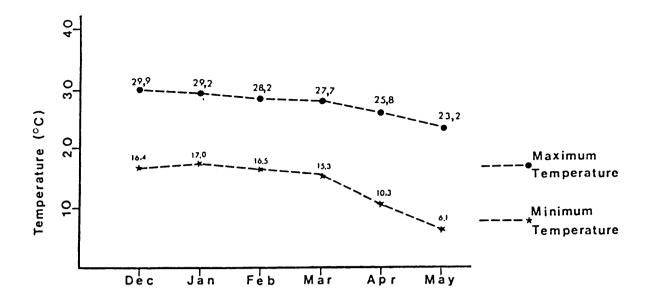


Figure 2 Mean monthly maximum and minimum temperatures at Towoomba Research Station from 1970 to 1985 during the six months sunflower season

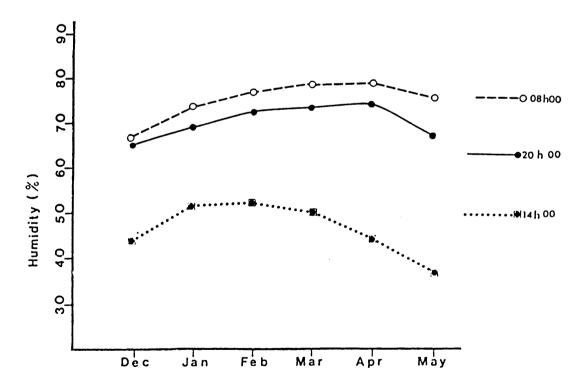


Figure 3 Mean monthly relative humidity at Towoomba Research Station from 1970 to 1985 during the six months sunflower season

Table 18The number of days from planting to 50 percentflowering, mean plant height, percentage male sterile plants,percentage off-types, percentage root lodging and percentagestalk lodging of each cultivar at Warm Baths during the 1989/90sunflower season (From: Loubser, Grimbeek & Steinmann, 1990)

Cultivar	days from planting	mean plant	% male sterile	% off- types	% root lodging	% stalk lodging
	to 50%	height	plants			
	flowering		- <u> </u>		·· ·	
AS 470	61	153	0	0	0	0
AS 505	64	174	2	0	0	0
AS 543	69	164	0	0	1	0
CAR 1012	65	154	1	0	0	0
CAR 1199	61	99	0	0	0	0
PNR 7204	54	147	0	0	0	1
PNR 7225	67	170	1	0	0	0
PNR 7369	65	154	0	0	0	0
PNR 7371	67	182	1	0	0	0
SNK 22	66	154	0	0	0	1
SNK 32	63	155	1	0	1	0
SNK 33	61	137	1	0	1	0
SNK 37	65	160	0	0	1	0
SO 222	56	156	0	0	1	2
SO 242	66	154	1	0	1	1
SO 306	66	165	1	0	1	1
SO 323	65	162	0	0	0	0
SUNKING	63	151	0	0	1	0

3.2.2. Determination of seed set, hollow seededness and unfilled seeds

The occurrence and extent of hollow seededness and unfilled seeds eighteen different sunflower cultivars within the were Ten mature sunflower heads with a diameter of not determined. less than 120 mm were harvested by hand from each of the three replications. Threshing was done by hand and after drying, the achenes were separated by hand into filled, hollow and underdeveloped (dull or papery) achenes. Filled achenes consisted of the oil-bearing seed or kernel and the surrounding pericarp. achenes developed a normal pericarp, which Hollow was occasionally smaller but contained no seed. These achenes also had a much lower mass as filled achenes and were usually blown out with the chaff during mechanical harvesting. Under-developed achenes consisted of a papery under-developed pericarp only, with no seed. The percentages of filled, hollow and under-developed achenes were calculated.

#### 3.2.3. Statistical analysis

A two-way analysis of variance (ANOVA), including cultivars and replicates as sources of variation were performed on the following variables: percentage filled seeds (seed set), percentage papery (dull) seeds and the 1000 seed mass for eighteen sunflower cultivars.

The following results and graphical output were also displayed by the computer program for each of these variables:

- (a) The effective standard errors of the means for the different cultivars and replicates.
- (b) The standard error among plants after removing the variation due to cultivars and replicates, as well as the corresponding coefficient of variation.
- (c) A histogram of the residuals for each of the variables, the main purpose being to see if the normality assumption is acceptable. Normality of the residuals was reasonable in all cases except % hollow seeds.
- (d) A plot of the residuals against the fitted values, as well as the results of different tests to check for the validity of the assumption of homogeneous variances among different cultivars.

For the variable percentage hollow seeds the non-parametric Kruskall-Wallis analysis of variance with random replication was used to analyse the data. This was carried out for all cultivars. The median, inter quartile range and class total were calculated. The Chi-Squared value and adjusted Chi-Squared value were calculated to test whether differences existed among treatments.

Finally, pairwise comparisons were made among the different cultivars using the non-parametric method of Dunn.

Statistical analysis was performed using the GENSTAT statistical analysis programme (Alvey, Galvey & Lane, 1982) on a Burroughs computer.

#### 3.3. Results

The means obtained from the primary data, which were used for further analysis, are presented in Table 19.

3.3.1. Head diameter

Head diameter was used as parameter to ascertain variances between cultivars, as well as within the three replicates of each cultivar under investigation. The largest heads were found in the cultivar CAR 1199 (mean 23,167 cm) while SNK 22 had the smallest heads (mean 18.333 cm). The calculated F-value was 12.533 (P <.001) for cultivars. This value points to highly significant variance among the eighteen cultivars under investigation (Table 21). The effective standard errors of means were calculated as 0.3684 for cultivars (Table 20). Standard error of the mean (SE) was calculated at 2.0180 with a coefficient of variation of 10.0% (Table 20). A histogram of the residual values showed a symmetrical distribution (Figure 4). The residual values were plotted against the fitted values and illustrated graphically in figure 5. The Miller-Levene test for homogeneity of variance was calculated as F = 1.50247 indicating no significant variation and the different cultivars could

therefore be evaluated and compared with one another.

#### 3.3.2. Filled seeds

Some inter cultivar variance was evident when the percentage filled seeds (equal to seed set) was analysed. The lowest seed set was recorded in PNR 7204 (79,75%) and the highest in SUNKING (90,35%). Another ten cultivars (AS 470; AS 505; AS 543; PNR 7225; PNR 7371; SNK 22; SNK 32 SNK 37; SO 242 and SO 323) had an average seed set of above 85%.

The calculated F-value in analysing the percentage seed set (filled seeds) was 10.086 (P <.001) for cultivars, indicating highly significant variation among the eighteen cultivars under investigation (Table 23). The effective standard errors of means were calculated as 0.857 for cultivars (Table 22). Standard error of the mean (SE) was calculated at 4.695 with a coefficient of variation of 5.5% (Table 22). A histogram of the residual values showed an even distribution (Figure 6). The residual values against the fitted values are presented graphically in Figure 7. The Miller-Levene test for homogeneity of variance was calculated as F = 4.07604 indicating significant variance amongst the different cultivars investigated. Comparison in pairs of the level of significance among the eighteen cultivars is illustrated in Table 24. The cultivars PNR 7204 and SUNKING differed significantly from most other cultivars (Tables 24).

#### 3.3.3. Hollow seededness

Insignificant inter cultivar variation was shown when the percentage hollow seededness among the cultivars was analysed. The two cultivars CAR 1199 and PNR 7204 differed significantly from most other cultivars (Table 25 & 26). A Chi-Squared value of 154.7611 was calculated (Table 25). This points to highly significant variance among most of the eighteen cultivars under investigation (Table 25). The lowest percentage hollow seeds was recorded in the cultivar AS 420 (2.50%) and the highest in CAR 1199 (10.30%) with the second highest in PNR 7204 (7.15%). Hollow seededness in all other cultivars was below 5%.

The Miller-Levene test for homogeneity of variance was calculated as F = 8.27758 indicating significant variance amongst the eighteen cultivars. Comparison in pairs of the level of significance among the eighteen cultivars is illustrated in Table 26.

#### 3.3.4. Papery seeds

Insignificant inter cultivar variation was shown when the percentage papery (dull) seeds among the cultivars were analysed. The lowest percentage papery seeds was recorded in the cultivar SUNKING (6.10%) and the highest in PNR 7204 (13.45%). The percentage papery (dull) seeds was above 10% in a further eight

cultivars (AS 505; AS 543; CAR 1012; PNR 7369; SNK 32; SNK 33; SO 222 and SO 306).

The calculated F-value for the percentage papery seeds (dull seeds) was 5.595 (P <.001) for cultivars. This points to highly significant variance among the eighteen cultivars under investigation (Table 28). The effective standard errors of means were calculated as 0.733 for cultivars (Table 27). Standard error of the mean (SE) was calculated at 4.013 with a coefficient of variation of 38.5% (Table 27). A histogram of the residual values showed an asymmetrical distribution (Figure 8). The residual values were plotted against the fitted values and illustrated graphically in Figure 9.

The Miller-Levene test for homogeneity of variance was calculated as F = 4.30735 indicating significant variance. Comparison in pairs of the level of significance among the eighteen cultivars is illustrated in Table 29. The cultivar SUNKING differed significantly from almost all other cultivars (Table 29).

#### 3.3.5. Seed mass

The 1000 seed mass factor was used as parameter to ascertain variation between cultivars with regard to potential yield in the cultivars under investigation. The highest 1000 seed mass was found in the cultivar SO 222 (mean 81.84 g) while another two cultivars (SNK 33 and SNK 37) had a 1000 seed mass of above 70

g (Table 30). The cultivar AS 505 had the lowest 1000 seed mass (mean 51.20 g). In another four cultivars, AS 470; PNR 7204; SNK 32 and SO 323, the 1000 seed mass was also below 60 g.

The calculated F-value was 13.655 (P <.001) for cultivars (Table 31), indicating significant variance among the eighteen cultivars under investigation. The effective standard errors of means were calculated as 2.270 for cultivars (Table 30). Standard error of the mean (SE) was calculated at 12.431 with a coefficient of variation of 19.1% (Table 30). A histogram of the residual values showed an even distribution (Figure 10). The residual values were plotted against the fitted values and presented graphically in Figure 11. The Miller-Levene test for homogeneity of variance was calculated as F = 2.38337 indicating significant variation amongst the eighteen cultivars.

## 3.4. Discussion

Sufficient numbers of insect pollinators were observed at the study locality for satisfactory pollination to have occurred. It was also concluded that soil fertility was not a restricting factor in the potential yield as demonstrated by top soil analysis (Table 3, Chapter 2) and an analysis of leaf and floral boron content (Table 6, Chapter 2). Other climatic conditions and agronomical practices were equal for all the cultivars investigated.

The eighteen cultivars showed insignificant differences for the parameters head diameter, seed set (filled seed), hollow seeds, papery (dull) seeds and the 1000 seed mass when looked at as a group. Significant variances between the cultivars for these parameters did however occur when they were compared with one The level of hollow seededness was of significant another. concern in two cultivars (CAR 1199 and PNR 7204) only. Of more importance was the fact that the percentage papery (dull or unfilled) seeds reached levels of above 10% in nine, i.e. 50%, of the cultivars investigated (AS 505; AS 543; CAR 1012; PNR 7204; PNR 7369; SNK 32; SNK 33; SO 222 and SO 306). Furthermore, the lowest percentage papery (dull) seeds (6.10% in SUNKING) was above the 5% level, which is the maximum plant breeders aim for. Herring (1981) reported that sterility in the head centre is one of the most overlooked sunflower cultivar characteristics. The results of this study indicate sufficient proof for plant breeders to give special attention to this phenomenon of dull seeds.

Competition between different seed houses led to the registration of between ten and twenty new sunflower in South Africa each year. Seed houses look for an improvement in yield through tolerance to drought and resistance to fungal diseases, bacterial diseases and bird depredation. Newly-developed cultivars may stay in the market for only one or two seasons, before being replaced by a cultivar. Popular cultivars may have a commercial life of perhaps five to six seasons. It is therefore very important for sunflower breeders to also take cognisance of

restricting factors such as hollow seededness and a sterile head centre. It is noteworthy that only four of the eighteen cultivars investigated in this study were still commercially available after five years (Agricultural Research Council, 1995), which clearly illustrates the high level of competition. External factors in the seed industry such as take-overs (e.g. Saffola by PANNAR), a shift on product focus (Asgrow focussing on vegetable seed) and new entrants to the South African market (Africa Pacific, Carnia, PHI Hi-bred, and Sandoz Seed) certainly played a significant role in this.

## 3.5. Conclusion

- Though head diameter among the eighteen cultivars differed these differences were not significant.
- Hollow seededness was of significance in two cultivars only. 12% in CAR 1199 and 8% in PNR7204.
- The number of unfilled seeds did not differ significantly among the eighteen cultivars investigated.
- The potential yield of the eighteen sunflower cultivars was mainly determined by differences in seed weight and not seed number.

Table 19 Mean head diameter (mm), number of filled, hollow and papery seeds, 1000 seed mass (g), % seed set and % hollow seeds in eighteen sunflower cultivars at Towoomba, Warm Baths during the 1989/90 sunflower season

Cultivar	Head	Filled	Hollow	Papery	1 000	8	Seed	8
	diameter	seeds	seeds	seeds	seed	seed	Yield	hollow
	(mm)				mass	set	kg/ha	seeds
					(g)			
AS 470	191	1397	51	123	51.75	89	2217	3
AS 505	203	1704	88	210	51.20	85	2107	4
AS 543	204	1661	67	200	63.11	86	1767	4
CAR 1012	216	1725	94	255	65.25	83	2157	5
CAR 1199	231	1537	240	154	65.90	80	1874	12
PNR 7204	187	1407	141	224	54.45	79	1579	8
PNR 7225	200	1504	63	154	68.09	87	1916	4
PNR 7369	197	1409	83	178	69.91	84	2250	5
PNR 7371	220	1755	88	203	67.58	86	2248	4
SNK 22	183	1432	90	165	60.78	84	2107	5
SNK 32	199	1585	88	188	58.13	85	2434	5
SNK 33	197	1473	88	234	71.29	82	2038	5
SNK 37	216	1726	65	186	79.01	87	2260	3
SO 222	198	1247	71	187	81.84	83	2068	5
SO 242	192	1450	68	179	69.65	85	1700	4
S0 306	189	1508	96	210	66.93	83	1782	5
SO 323	197	1434	69	168	58.30	86	2280	4
SUNKING	219	1612	69	122	69.40	89	2095	4

Table 20Table of means of head diameter in eighteen sunflowercultivars at Towoomba, Warm Baths during the 1989/90 sunflowerseason

Cultivar	Mean	
AS 470	19.133	
AS 505	20.250	
AS 543	20.350	
CAR 1012	21.583	
CAR 1199	23.167	
PNR 7204	18.700	
PNR 7225	19.983	
PNR 7369	19.667	
PNR 7371	21.950	
SNK 22	18.333	
SNK 32	19.910	
SNK 33	19.733	
SNK 37 SO 222	21.550 19.817	
SO 222 SO 242	19.233	
SO 306	18.917	
SO 323	19.717	
SUNKING	21.883	
<u></u>		
Minimum	13.50	
Maximum	28.00	
Total values	540	
Missing values	0	
Mean replicate 1		
Mean replicate 2		
Mean replicate 3		
Grand mean	20.215	
Effective standa	ard errors of t	the mean (ES
	Cultivars Re	plicates
Replicates	30	180
ESE	0.3684	0.1504

Stratum standard errors and coefficients of variation

Stratum	df	SE	CV%
*units*	520	2.0180	10.0

(ESE)

Table 21ANOVA table of head diameter for eighteen sunflowercultivars at Warm Baths during the 1989/90 season

Source of variation	df	SS	SS%	MS	VR	F PR
Cultivar	17	867.645	28.04	51.038 <b>1</b>	2.533	<.001
Replicates	2	108.930	3.52	54.465 <b>1</b>	3.375	<.001
Residual	520	2117.567	68.44	4.072		
Total	539	3094.142	100.00	5.741		
Grand Total	539	3094.142				
Grand mean		20	.215			
Total number of obser	vations	540	)			

-	-6.0	0	
-6.0 -	-5.4	1	*
-5.4 -	-4.8	4	***
-4.8 -	-4.2	5	****
-4.2 -	-3.6	8	*****
-3.6 -	-3.0	18	*****
-3.0 -	-2.4	22	*******
-2.4 -	-1.8	47	**********
-1.8 -	-1.2	41	*******
-1.2 -	-0.6	51	***************************************
-0.6 -	0.0	69	***************************************
0.0 -	0.6	72	***************************************
0.6 -	1.2	55	*****************
1.2 -	1.8	52	******************
1.8 -	2.4	38	* * * * * * * * * * * * * * * * * * * *
2.4 -	3.0	28	*********
3.0 -	3.6	9	*****
3.6 -	4.2	8	*****
4.2 -	4.8	5	****
4.8 -	5.4	6	****
5.4 -	6.0	1	*
6.0 -	6.6	0	
6.6 -		0	

Missing values : 0 Asterisk represents 1 unit

Figure 4 Histogram of residual for head diameter in eighteen sunflower cultivars at Towoomba, Warm Baths during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 17 and 504 degrees of freedom: F = 1.50247

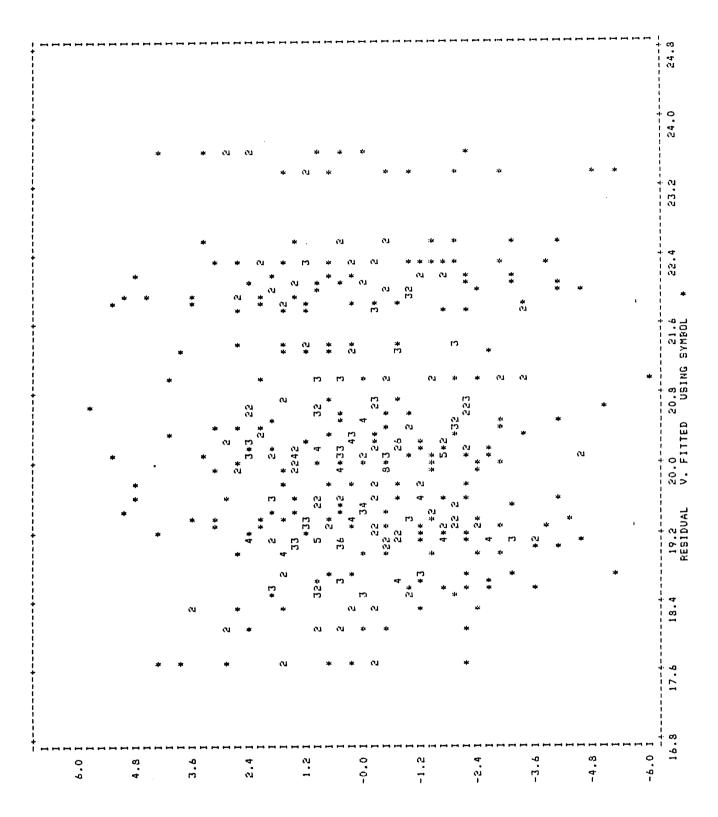


Figure 5 The residual values plotted against the fitted values among eighteen sunflower cultivars for the variant head diameter

Table of means of percentage filled seeds (seed set) Table 22 in eighteen sunflower cultivars at Towoomba, Warm Baths during the 1989/90 sunflower season

Cultivar	Mean		
AS 470	88.88		
AS 505	85.11		
AS 543	86.06		
CAR 1012	83.03		
CAR 1199	79.64		
PNR 7204	79.37		
PNR 7225	87.25		
PNR 7369	84.22		
PNR 7371	85.80		
SNK 22	84.46 85.22		
SNK 32 SNK 33	82.06		
SNK 33	86.90		
SNR 37 SO 222	82.70		
SO 242	85.38		
SO 306	82.96		
SO 323	85.74		
SUNKING	89.29		
Minimum	64.00		
Maximum	94.00		
Total values	540		
Missing values	0		
Mean replicate			
Mean replicate			
Mean replicate Grand mean	84.67		
Grand mean	04.07		
Effective stand	dard errors of	the mean	(ESE)
	Cultivars	Replicates	
Replicates	30	180	
ESE	0.857	0.35	50
Stratum standa	rd errors and	coefficient	ts of <sup>.</sup>

of variation

Stratum	df	SE	CV%
*units*	520	4.695	5.5

Table 23ANOVA table of percentage filled seeds (seed set) foreighteen sunflower cultivars at Warm Baths during the 1989/90 season

Source of variation	df	SS	SS%	MS	VR	F PR
Cultivar	17	3779.29	24.25	222.31	10.086	<.001
Replicates	2	341.06	2.19	170.53	7.737	<.001
Residual	520	11461.28	73.56	22.04		
Total	539	15581.63	100.00	28.91		
Grand Total	539	15581.63				
Grand mean		84	1.67			
Total number of observ	vations	540	)			

21.0	1 *
-21.019.5	0
-19.518.0	0
-18.016.5	1 *
-16.515.0	0
-15.013.5	3 **
-13.512.0	1 *
-12.010.5	12 *****
-10.5 9.0	7 ****
- 9.0 7.5	16 ******
- 7.5 6.0	22 *******
- 6.0 4.5	19 *******
- 4.5 3.0	35 *********
- 3.0 1.5	48 *******************
- 1.5 - 0.0	60 ******
0.0 - 1.5	89 ********************
1.5 - 3.0	92 **********************
3.0 - 4.5	62 ************************************
4.5 - 6.0	33 ********
6.0 - 7.5	27 *********
7.5 - 9.0	8 ****
9.0 - 10.5	4 ***
10.5 -	0

Missing values : 0 Asterisk represents 1.5 units

Figure 6 Histogram of residual for percentage filled seeds (seed set) in eighteen sunflower cultivars at Towoomba, Warm Baths during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 17 and 504 degrees of freedom: F = 4.07604

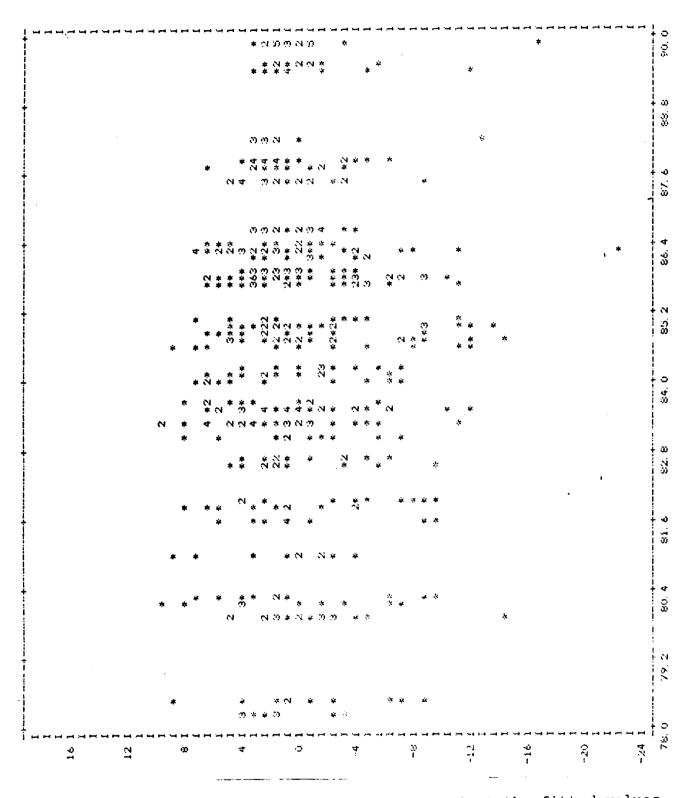


Figure 7 The residual values plotted against the fitted values among eighteen sunflower cultivars for the variant percentage seed set

Table 24Significance for all pairwise comparisons amongeighteen sunflower cultivars for the percentage filled seeds (seedset), using Dunn's method

	AS	AS	AS	CAR	CAR	PNR	PNR	PNR	PNR	SNK	SNK	SNK	SNK	so	SO	SO	so
	470	505	543	1012	1199	7204	7225	7369	7371	22	32	33	37	222	242	306	323
AS 50	5																
AS 54	3																
CAR	**																
1012																	
CAR	**		*														
1199																	
PNR	**	**	**														
7204																	
PNR					**	**											
7225																	
PNR	*					*											
7369																	
PNR					**	**											
7371																	
SNK 2	2					**											
SNK 3	2					**											
SNK 3	3 **						**										
SNK 3	7				**	**						**					
SO 22	2 **																
SO 24	2					**											
SO 30	6 **																
SO 32	3				**	**											
SUNKT	N	*		**	**	**		**		*	*	**		**	*	**	

Table 25	Kruska	al-Wallis a	analysis o	f va	ariance of	perce	entage	hollow
seeds in	eighteen	sunflower	cultivars	at	Towoomba,	Warm	Baths	during
the 1989,	/90 sunfl	ower sease	on					

Cultivar	Median	Inter o	quartile Class total
		range	
AS 470	2.50	0.55	4053.00
AS 505	3.70	0.70	7978.00
AS 543	3.00	0.85	5804.00
CAR 1012	4.25	1.65	8010.00
CAR 1199	10.30	3.70	14796.00
PNR 7204	7.15	1.00	12893.50
PNR 7225	3.10	1.00	6139.00
PNR 7369	3.95	2.25	8932.50
PNR 7371	3.05	1.95	6936.00
SNK 22	4.30	1.80	9760.00
SNK 32	3.40	1.45	7668.00
SNK 33	3.95	1.85	8870.50
SNK 37	3.15	0.50	5664.50
SO 222	4.05	0.95	8485.00
SO 242	4.15	0.85	7679.00
SO 306	4.30	1.55	9564.00
SO 323	2.90	2.15	6242.00
SUNKING	3.25	0.55	6595.00

Chi-Squared value = 154.7611

17 Degrees of freedom

Adjusted Chi-Squared value = 154.8338

17 Degrees of freedom

Table 26 Significance for all pairwise comparisons among eighteen sunflower cultivars for the percentage hollow seeds, using Dunn's method

	AS	AS	AS	CAR	CAR	PNR	PNR	PNR	PNR	SNK	SNK	SNK	SNK	SO	so	so	SO
	470	505	543	1012	1199	7204	7225	7369	7371	22	32	33	37	222	242	306	323
AS 505	5																
AS 543	3																
CAR																	
1012																	
CAR	**	**	**	**													
1199																	
PNR	**	**	**	**													
7204																	
PNR					**	**											
7225																	
PNR	**				**												
7369																	
PNR					**	**											
7371																	
SNK 22	2 * *				**												
SNK 32	2				**	**											
SNK 33	} *				**												
SNK 37	7				**	**											
SO 222					**	*											
SO 242					**	**											
SO 306					**												
SO 323	5				**	**											

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 17 and 504 degrees of freedom: F = 8.27758

\*\* \*\* \*\*

SUNKIN

Table 27 Table of means of percentage papery (dull) seed in eighteen sunflower cultivars at Towoomba, Warm Baths during the 1989/90 sunflower season

Cultivar	Mean	·	
AS 470	7.82		
AS 505	10.48		
AS 543	10.41		
CAR 1012	12.47		
CAR 1199	8.04		
PNR 7204	12.68		
PNR 7225	9.05		
PNR 7369	10.80		
PNR 7371	9.90		
SNK 22	11.52		
SNK 32	10.14		
SNK 33	12.99		
SNK 37	9.75		
SO 222	12.58		
SO 242	10.63		
SO 306	11.60		
SO 323	10.00		
SUNKING	6.89		
Minimum	2.30	)	
Maximum	48.00		
Total values	540		
Missing values			
Mean replicate		'9	
Mean replicate			
Mean replicate			
Grand mean	10.4		
Effective star	dard errors	of the mean	(ESE)
Popliastos	Cultivars 30	Replicates 180	
Replicates ESE	0.733	0.2	99
ц ц ц ц ц	0.755	0.2	
Stratum stands	ard arrors an	d coefficier	nts of variation
Stratum	df	SE	CV%
*units*	520	4.013	38.5

Table 28ANOVA table of percentage papery (dull) seed foreighteen sunflower cultivars at Warm Baths during the 1989/90 season

Source of variation	df	SS	SS%	MS	VR	F PR
Cultivar	17	1531.95	15.17	90.11	5.595	<.001
Replicates	2	192.99	1.91	96.50	5.991	0.003
Residual	520	8375.15	82.92	16.11		
Total	539	10100.09	100.00	18.74		
Grand Total	539	10100.09				
Grand mean		10	0.43			
Total number of observ	ations	540	)			

-	-12.5	0	
-12.5 -	-10.0	0	
-10.0 -	-7.5	1	*
-7.5 -	-5.0	21	****
-5.0 -	-2.5	112	* * * * * * * * * * * * * * * * * * * *
-2.5 -	0.0	174	**********
0.0 -	2.5	136	*******
2.5 -			****
5.0 -	7.5	27	****
7.5 -	10.0	13	****
10.0 -	12.5	1	*
12.5 -	15.0	4	**
15.0 -	17.5	0	
17.5 -	20.0	0	
20.0 -	22.5	0	
22.5 -	25.0	1	*
25.0 -	27.5	0	
27.5 -	30.0	0	
30.0 -	32.5	0	
32.5 -	35.0	0	
35.0 -	37.5	1	*
37.5 -	40.0	0	
40.0 -		0	

Missing values : 0 Asterisk represents 2 units

**Figure 8** Histogram of residual for percentage papery (dull) seed in eighteen sunflower cultivars at Towoomba, Warm Baths during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 17 and 504 degrees of freedom: F = 4.32629

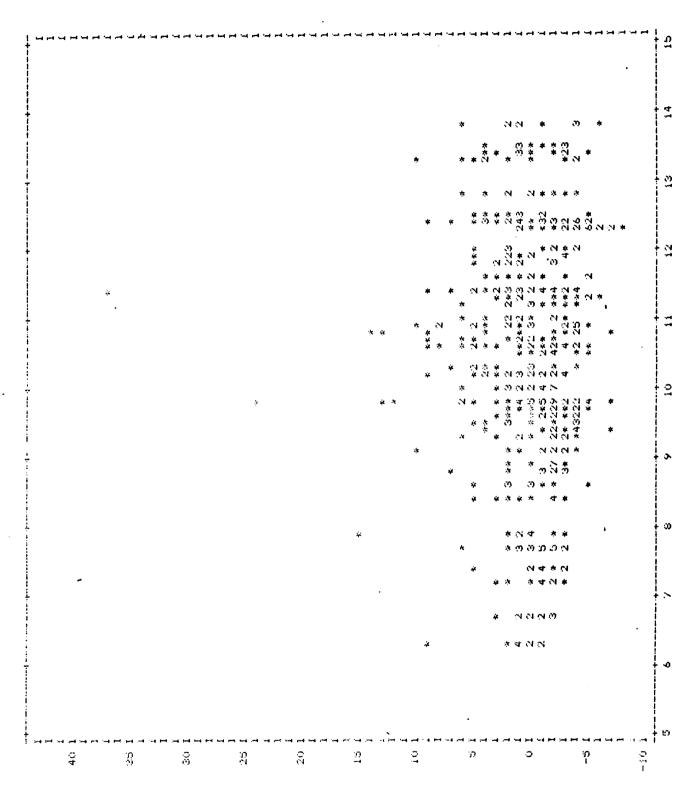


Figure 9 The residual values plotted against the fitted values among eighteen sunflower cultivars for the variant percentage dull seeds

	AS 470	<b>AS</b> 505	AS 543	CAR 1012	CAR 1199	PNR 7204	PNR 7225	<b>PNR</b> 7369	PNR 7371	SNK 22	SNK 32	SNK 33	SNK 37		SO 242	SO 306	SO 323
AS 505																	
AS 543																	
CAR 1012	**																
CAR 1199				**													
PNR 7204	**				**												
PNR 7225						**											
PNR 7369																	
PNR 7371																	
SNK 22																	
SNK 32																	
SNK 33	**				**		**										
SNK 37																	
SO 222	**				**		*										
SO 242																	
SO 306	**				**												
SO 323				*		**						**		**			
SUNKIN		**	**	**		**		**	*	*	**	**		**	**	**	

Table 29 Significance for all pairwise comparisons among eighteen sunflower cultivars for the percentage papery (dull) seeds, using Dunn's method

Table 30Table of means of the 1000 seed mass in eighteensunflower cultivars at Towoomba, Warm Baths during the 1989/90sunflower season

Cultivar	Mean				
AS 470	51.75				
AS 505	51.20				
AS 543	63.11				
CAR 1012	65.25				
CAR 1199	65.90				
PNR 7204	54.45				
PNR 7225	68.09				
PNR 7369	69.91				
PNR 7371	67.58				
SNK 22	60.78				
SNK 32	58.13				
SNK 33	71.29				
SNK 37	79.01				
SO 222	81.84				
SO 242	69.65				
SO 306	66.93				
SO 323	58.30				
SUNKING	69.40				
	1.0	<b>2</b> 0			
Minimum	10.				
Maximum	99.	90			
Total values	540				
Missing values		22			
Mean replicate					
Mean replicate					
Mean replicate					
Grand mean	65.	14			
Effective stan	dard errors	of the	mean	(ESE)	
	Cultivars				
Replicates	30	керт	icates 180		
ESE	2.270		0.92	7	
	2.2/0		0.72	,	
Stratum standa	rd errors a	nd coef	ficient	s of '	variation

Stratum	df	SE	CV%
*units*	520	12.431	19.1

			· · · · · ·			
Source of variation	df	SS	SS%	MS	VR	F PR
Cultivar	173	35871.1	29.56	2110.1	13.655	<.001
Replicates	2	5126.9	4.22	2563.4	16.589	<.001
Residual	520	80355.6	66.22	154.5		
Total	539	121353.5	100.00	225.1		
Grand Total	539	121353.5				
Grand mean		65	.14			
Total number of observa	tions	540				

Table 31ANOVA table of the 1000 seed mass for eighteen sunflowercultivars at Warm Baths during the 1989/90 season

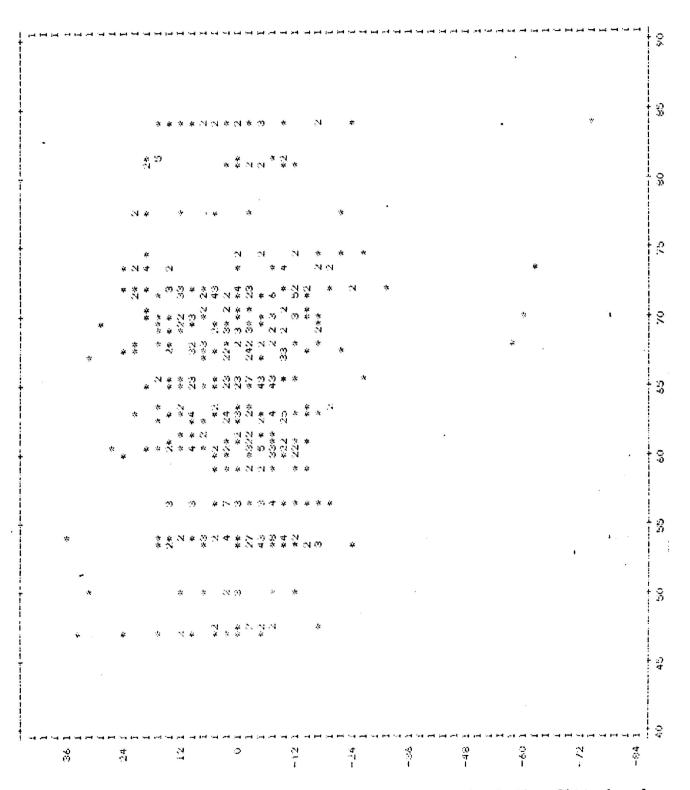
				_	
	-70	-	-65	0	
	-65	-	-60	1	*
	-60	-	-55	0	
	-55	-	-50	0	
	-50	-	-45	0	
	-45	-	-40	0	
-	-40	-	-35	0	
	-35	-	-30	1	*
	-30	-	-25	3	**
	-25	-	-20	9	****
-	-20	-	-15	21	****
-	-15	_	-10	52	***************************************
	-10	_	-5	93	************************
-	-5	-	0	101	***************************************
	0	-	5	92	***********************
	5	-	10	50	*****
	10	-	20	37	* * * * * * * * * * * * * * * * * * * *
2	20	_	25	17	* * * * * * * *
2	25	-	30	2	*
	30	-	35	3	**
	35	-		1	*

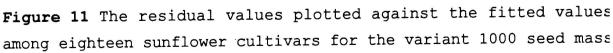
Missing values : 0 Asterisk represents 1.5 units

- -70 1 \*

Figure 10 Histogram of residual for 1000 seed mass in eighteen sunflower cultivars at Towoomba, Warm Baths during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 17 and 504 degrees of freedom: F = 2.38337





### CHAPTER 4

## INVESTIGATION INTO SEED SET OF TWO SUNFLOWER CULTIVARS AT DIFFERENT PLANT DENSITIES.

#### 4.1. Introduction

Cultivar and plant density both play an important role in the growth and plant development of sunflower. These influences are manifested in plant characteristics such as plant height, stem diameter and head diameter. The end result is that this is carried through to the potential number of seeds that develop and the potential yield to be obtained. Yield in sunflower is determined by the number of heads per hectare, the number of achenes per head, and the mean weight of seed (Bonari, Vannozzi, Benvenuti & Baldini, 1992; Robinson, 1978). Sunflowers produce only one capitulum per plant, which makes the number of plants area the most important production parameter. per unit Consequently the number of plants per hectare strongly influences seeds per head, and their mean weight. the number of Environmental factors such as climate, soil type, diseases and pests also play a role. Smaller heads have a smaller achene number than those of larger heads. The effect of plant density

on the potential yield is not only masked by the environmental factors listed above, but also by management techniques such as irrigation and mineral nutrition (Bonari <u>et al.</u>, 1992). In South Africa sunflower is generally planted at a density of 35000 plants per hectare (Van der Merwe, 1988). Row spacing ranges from 0.75 m to 1.0 m.

An important characteristic of sunflower is its ability to compensate for low plant densities by increasing head size. This results in more seeds per head as well as larger (heavier) seed (Robinson, 1978).

The aim of this investigation was to quantify the effect of plant density on the seed set and the occurrence of hollow seededness in both a dwarf and an intermediate sunflower cultivar at a single locality.

#### 4.2. Methods

#### 4.2.1. Description of trial

Permission was obtained from the Director, Highveld Region, Potchefstroom to make use of the plant density trials under his supervision (Project H5213/30/1/3) to conduct the study. The object of these trials was to supply the producer with reliable information on optimum plant density in dwarf cultivars as opposed to intermediary cultivars. Dwarf sunflower cultivars flower within 70-80 days from planting and therefore have a longer growing season, while intermediate cultivars flower from 60-70 days from planting. The old open-pollinated varieties had a very long growing season of between 90-100 days before full bloom. The extent of these trials therefore ideally suited the present investigations as all climatic and cultivar data would have been available for analysis as well.

The trial was planted by hand on 5 February 1990 at Vermaas, North-West Province on Avalon soils. The trial was maintained by personnel of the Highveld Region. Soil analysis was performed prior to fertilizing and planting. The soil characteristics are presented in Table 32. Fertilizer was applied one week prior to planting. 180 kg 320(25) which represents 82 kg N, and 50 kg KAN(28) which represents 36 kg P per hectare, was applied mechanically. Plant nutrition was therefore equal for the different plant densities investigated.

The different plant density treatments are presented in Table 33. The trial was planted in a randomized block design with six different plant densities, which were repeated three times. The nett plot size remained constant at 36 m<sup>2</sup>, i.e. 12 m x 3 m, inter- and intra-row distances decreased as plant density increased.

Rainfall was monitored and is presented as the total rainfall for each month during the period October 1989 to April 1990 (Table 34). The accumulated rainfall of 562.0 mm measured over the

period is equal to the long term average of 560 mm for this locality.

Plots were thinned after germination to the required plant density. Plant density was again calculated prior to harvesting (Table 35). Although it was difficult to maintain the high population densities, no significant differences in density were found between the two cultivars at each treatment. Table 32 Soil characteristics at the plant density trial site, Vermaas, North-West Province

Characteristic	Description
Soil type	Avalon 36
Effective depth	550 mm
pH (H <sub>2</sub> O)	6.0
P	44 ppm
K	0.55 me 100 g <sup>-1</sup>
Ca	1.83 me 100 g <sup>-1</sup>
Mg	0.61 me 100 g-1
Percentage clay	10%
Texture	Sandy loam

Table 33 Plant density treatments investigated for each of the cultivars PNR 7460 (dwarf type) and SNK 32 (intermediary type)

Plant density	Designation	
15000 plants ha <sup>-1</sup>	POP 15	<u> </u>
30000 plants ha <sup>-1</sup> 45000 plants ha <sup>-1</sup>	POP 30 POP 45	
60000 plants ha <sup>-1</sup>	POP 60	
75000 plants $ha^{-1}$	POP 75 POP 90	
90000 plants ha <sup>-1</sup>	FOF 90	

Table 34 Monthly rainfall (mm) recorded at the plant density trial site, Vermaas, North-West Province, for the period October 1989 to April 1990

Oct	Nov	Dec	Jan	Feb	March	Apr	Total
19.0	97.0	115.0	51.0	59.0	133.0	88.0	562.0

Table 35 Calculated plant density of the different treatments at time of harvesting

Treatment designation	PNR 7460	SNK 32	
POP 15	15129	15925	
POP 30	30826	29120	
POP 45	45839	43225	
POP 60	62221	59150	
POP 75	76326	72325	
POP 90	86190	86905	

# 4.2.2. Determination of seed set and hollow seededness

The occurrence and extent of hollow seededness was investigated for both the dwarf sunflower cultivar (PNR 7460) and the intermediate cultivar (SNK 32) at six plant densities. Ten mature sunflower heads with a diameter of not less than 120 mm were harvested by hand from each of the three replicates for each Threshing was done by hand and after drying the cultivar. achenes were separated by hand into filled, hollow and underdeveloped (dull or papery) achenes. Filled achenes consisted of the oil-bearing seed or kernel and the surrounding pericarp. Hollow achenes developed a normal pericarp, which occasionally was smaller but contained no seed. These achenes also had a much lower mass as filled achenes and were usually blown out with the chaff during mechanical harvesting. Under-developed achenes consisted of a papery under-developed pericarp only, with no The percentage filled, hollow and papery seeds were seed. expressed as a function of the total number of seeds.

#### 4.2.3. Statistical analysis

A two-way analysis of variance (ANOVA), including plant densities and replicates as sources of variation, was performed on the following variables: percentage filled seeds (seed set), percentage papery (dull) seeds and the 1000 seed mass for both dwarf and intermediate sunflower cultivars.

The following results and graphical output were also displayed by the computer program for each of the above-mentioned variables:

- (a) The effective standard errors of the means for different plant densities and replicates
- (b) The standard error among plants after removing the variation due to plant densities and replicates, as well as the corresponding coefficient of variation.
- (c) A histogram of the residuals for each of the variables, the main purpose being to see if the normality assumption is acceptable. Normality of the residuals was reasonable in all cases except for % hollow seeds.
- (d) A plot of the residuals against the fitted values, as well as the results of different tests to check for the validity of the assumption of homogeneous variances among different cultivars.

For the variable percentage hollow seeds the non-parametric Kruskall-Wallis analysis of variance with random replication was used to analyse the data. This was carried out for both the dwarf and intermediate sunflower cultivars. The median, inter quartile range and class total were calculated. The Chi-Squared value and adjusted Chi-Squared value were calculated to test whether differences existed among treatments.

Finally, pairwise comparisons between the different plant densities for the two cultivars were carried out using the nonparametric method of Dunn.

Statistical analysis was performed using the GENSTAT statistical analysis programme (Alvey, Galvey & Lane, 1982) on a Burroughs computer.

#### 4.3. Results

The number of days to 50 percent flowering, the mean plant height and the mean stem diameter differed between the dwarf and the intermediary cultivar (Table 36). A positive interaction was found between the increase in plant density and plant height in both cultivars, i.e. as plant density increased, the internodes became longer, resulting in longer and more slender plants (Table 36). This phenomenon is well known in flowering plants. It resulted in a negative correlation between plant density and the parameters head diameter and the number of days to 50% flowering (Tables 37 + 38).

Insect pollination was adequate as mean numbers of 314 honeybees, 309 spotted maize beetles and 20 other insect pollinators were observed per 100 open capitula. This enhanced seed set.

Tables 37 and 38 contain the means of the primary data which were used for further analyses for the dwarf sunflower cultivar PNR 7460 and the intermediate sunflower cultivar SNK 32, respectively. The results obtained for each cultivar are presented separately in relation to seed set (filled seeds), hollow seededness, unfilled (papery or dull) seeds and the 1000

seed mass.

#### 4.3.1. Filled seeds

Significant inter plant density variation showed up when the percentage filled seeds (equal to seed set) was analysed for the dwarf sunflower cultivar PNR 7460 (Table 39). The highest seed set was found at a plant density of 15,000 plants per hectare. Seed set then decreased with an increase in plant density. The calculated F-value in analysing percentage seed set was 14.016 (P < .001) for plant density (Table 40), which points to significant The effective standard errors of the mean were variance. calculated at 1.686 for plant density (Table 39). Standard error of the mean (SE) was calculated at 9.235 with a coefficient of variation of 13.7% (Table 39). A histogram of the residual values showed an even or symmetrical distribution (Figure 12). A plot of residual values against the fitted values is presented in Figure 13. The Miller-Levene test for homogeneity of variance was calculated as F = 2.49874 indicating significant variance between the different plant densities.

The percentage filled seeds (seed set) showed significant inter plant density variation in the intermediate sunflower cultivar SNK 32 (Table 41). The highest seed set was found at a plant density of 15,000 plants per hectare, which decreased as plant density increased. The calculated F-value for seed set was

64.354 (P < .001) for plant density (Table 42). This value indicated highly significant variance for the parameter plant density. The effective standard errors of the mean was calculated at 1.042 for plant density (Table 41). The standard error of the mean (SE) was calculated at 5.709 with a coefficient of variation of 7.7% (Table 41). A histogram of the residual values showed a symmetrical distribution (Figure 14). A plot of the residual values against the fitted values is presented in Figure 15. The Miller-Levene test for homogeneity of variance was calculated as F = 2.72227 indicating significant variance among the different plant densities.

#### 4.3.2. Hollow seededness

Heterogeneous variation for population density was found at the lower population densities (between 15,000 and 30,000 plants ha<sup>-1</sup>) and then at the higher population densities (between 60,000 and 90,000 plants ha<sup>-1</sup>) in the dwarf sunflower cultivar PNR 7460 for hollow seededness (Table 43). A Chi-Squared value of 37.0710 was calculated (Table 43). A comparison in pairs of the level of significance among the plant densities is illustrated in Table 44. Surprisingly the lowest level of hollow seededness (2.50%) was found at a density of 30,000 plants ha<sup>-1</sup>. The Miller-Levene test for homogeneity of variance indicated highly significant variance with a calculated F-variate of 3.60603.

The intermediate sunflower cultivar SNK 32 showed significant variation for hollow seededness between the 30,000 plant density and the other planting densities (Table 46). Hollow seededness was significantly lower at this density (2.75%) compared to other plant densities (Table 45). The calculated Chi Square value of 34.3135 indicates significant variance between the different plant densities (Table 45). The Miller-Levene test for homogeneity of variance was calculated as F = 1.61472 indicating insignificant variance among plant density for the parameter hollow seededness.

#### 4.3.3. Papery seeds

Significant inter plant density variation was shown when the percentage papery (dull) seeds was analysed in the dwarf sunflower cultivar PNR 7460. The lowest percentage of papery seeds was recorded at a plant density of 15,000 plants ha<sup>-1</sup> with increasing percentages of papery seeds as plant density increased (Table 47). Analysis of variance showed a calculated F-value of 13.22 (P < .001) for plant density (Table 48). The effective standard errors of the means were calculated as 1.708 for plant density (Table 47). The standard error of the mean (SE) was calculated at 9.357 with a coefficient of variation of 33.3% (Table 47). A histogram of the residual values showed an even distribution (Figure 16). The residual values was plotted against the fitted values and presented graphically in Figure 17. The Miller-Levene test for homogeneity of variance was calculated

as F = 1.53229 indicating insignificant variance between the different plant densities.

Inter plant density variation differed significantly when the percentages of papery (dull) seeds were analysed in the intermediate sunflower cultivar SNK 32. The lowest percentage papery seeds was recorded at a plant density of 15,000 plants ha <sup>1</sup>. The number of papery seeds then increased with an increase in plant density (Table 49). Analysis of variance showed a calculated F-value of 61.029 (P < .001) for plant density, which was highly significant (Table 50). The effective standard errors of the means were calculated at 0.999 for plant density (Table 49). The standard error of the mean (SE) was calculated at 5.472 with a coefficient of variation of 25.3% (Table 49). A histogram of the residual values showed a symmetrical distribution (Figure 18). The residual values against the fitted values were plotted graphically in Figure 19. The Miller-Levene test for homogeneity of variance was calculated as F = 5.26916 indicating highly significant variance among the different plant densities.

#### 4.3.4. Seed mass

The 1000 seed mass factor was used as parameter to ascertain variation between the different plant densities with regard to the potential yield in the two cultivars under investigation.

The highest 1000 seed mass was found at 15,000 plants  $ha^{-1}$  (83.78 g). This differed significantly from that found in other plant densities in the dwarf sunflower cultivar PNR 7460 (Table 51). Analysis of variance revealed a calculated F-value of 75.250 (P < .001) for plant densities (Table 52). This value points to a highly significant variance. The effective standard errors of the means were calculated as 1.668 for plant density (Table 51). The standard error of the mean (SE) was calculated at 9.138 with a coefficient of variation of 16.7% (Table 51). A histogram of the residual values is presented in Figure 20 and shows an even distribution. The plot of residual values against the fitted values is illustrated graphically in Figure 21. The Miller-Levene test for homogeneity of variance was calculated as F = 5.93406 indicating highly significant variance.

Analysis of variance showed highly significant differences in 1000 seed mass for the different plant densities in the intermediate sunflower cultivar SNK 32. The F-value was calculated at 29.603 (P < .001) for plant density (Table 54). The 1000 seed mass did decrease in weight as the plant density The effective standard errors of the means were increased. calculated at 1.493 for plant density (Table 53). The standard error of the mean (SE) was calculated at 8.176 with a coefficient of variation of 16.1% (Table 53). A histogram of the residual values showed a symmetrical distribution (Figure 22). The residual values against the fitted values are presented graphically in Figure 23. The Miller-Levene test for homogeneity of variance was calculated as F = 3.71786 indicating highly

significant variance among the plant densities investigated.

#### 4.4. Discussion

Various researchers have investigated the influence of planting density on the number of seeds per head and the average seed mass in sunflower (Holt & Campbell, 1984; Putt & Urnau, 1943; Steer, Coaldrake, Pearson & Canty, 1986; Vijayalakshmi, Sanghi, Pelton & Anderson, 1975). They found a negative correlation between plant density, the number of seed per heads and the average seed mass. As plant density increased, the number of seeds head and average seed mass decreased (l.c). This negative correlation was confirmed by the present study.

The present detailed analysis of the seeds' developmental path into filled, hollow and unfilled seed, furthermore, demonstrated that a head of a specific diameter can support only a specific number of seeds, nourishing them to reach maturity (Table 41 and 42). Not only did the total number of seeds in both the dwarf and intermediate cultivars decrease but the number of unfilled (dull) seeds increase simultaneously. The level of hollow seededness did differ significantly for both the dwarf and intermediate cultivars at the various plant densities. Of major significance for the potential yield was the high percentage unfilled (dull) seeds, which increased with an increase in plant density. In the dwarf cultivar this level of hollow seededness increased from 20% to 39%. The percentage of unfilled seeds

increased in the intermediate cultivar from 11% at 15,000 plants ha<sup>-1</sup> to 33% at 90,000 plants ha<sup>-1</sup>. According to Herring (1981), this phenomenon of unfilled seeds in the centre of the sunflower head is a major reason for poor yield in sunflower. Seed mass showed the same negative correlation. As plant density increased, the seed mass decreased from 83.78 g 1000<sup>-1</sup> seeds at 15,000 plants ha<sup>-1</sup> to 44.98 g 1000<sup>-1</sup> at 75,000 plants ha<sup>-1</sup> in the dwarf cultivar PNR 7460. A similar decrease in seed mass and potential yield was observed in the intermediate cultivar SNK 32.

Khalifa (1984) postulated that few nutrients are transferred from the vegetative growth to the flower head because of the high competition for nutrients in the vegetative plant parts at higher plant densities. This results in inferior heads with low numbers of seed per head. Shading is a further possible restricting factor at higher plant densities (Rawson & Turner, 1982). High plant densities can exhaust the available soil moisture more quickly under dryland conditions, which is the general practice for sunflower production in South Africa.

#### 4.5. Conclusions

- 1. Plants of the dwarf cultivar PNR 7460 were shorter than those of the intermediate cultivar SNK 32.
- Head diameter was larger in PNR 7460 than in SNK 32. The measurements of this parameter decreased with increased plant density in both cultivars.

- 3. The percentage seed set (filled seeds) decreased in both cultivars as the planting density increased.
- 4. A negative relationship was found between hollow seededness and plant density. The percentage hollow seeds decreased in both cultivars as plant density increased. This confirms previous speculation that hollow seededness is associated with head size.
- 5. A positive relationship was found between unfilled (papery or dull seeds) and plant density. The number of unfilled seeds increased in both PNR 7460 and SNK 32 as the plant population increased. This supports reports that increasing competition for nutrients results in a decrease in yield potential.
- 6. The negative correlation between hollow seededness and plant density and positive correlation between unfilled seed and plant density require a compromise between a high and low plant density for optimum yield.
- 7. The 1000 seed mass decreased in both dwarf and intermediate cultivars as the plant density increased. This again points to the increase in demand for nutrients at higher plant populations. It also confirms sunflower's ability to compensate by increasing seed mass at lower planting densities.

Table 36 The mean number of days to 50% flowering, mean plant height and mean stem diameter in the dwarf sunflower cultivar PNR 7460 and intermediate sunflower cultivar SNK 32 at Vermaas during the 1989/90 sunflower season

	PNR 60	SNK 32	
mean number of days to	72	63	
50% flowering			
mean plant height (m)	1.18	1.70	
mean stem diameter (mm)	28.99	27.15	

Table 37 Mean head diameter (mm), number of filled, hollow and papery seeds, 1000 seed mass (g), % seed set and % hollow seeds among different planting densities in the dwarf sunflower cultivar PNR 7460 at Vermaas during the 1989/90 sunflower season

Planting	Head	Filled	Hollow	Papery	1 000	Seed	Seed	Hollow
density	dia-	seeds	seeds	seeds	seed	set	Yield	seeds
	meter				mass	(%)	kg/ha	(%)
	(mm)				(g)			
POP 15	254	1615	114	435	83.78	74	1542	6
POP 30	192	1454	67	526	52.88	71	1712	3
POP 45	168	1129	77	479	51.03	67	1451	5
POP 60	160	1019	50	386	47.97	69	1142	4
POP 75	144	869	71	611	44.98	56	1380	5
POP 90	148	1003	72	424	48.00	65	1320	6

**Table 38** Mean head diameter (mm), number of filled, hollow and papery seeds, 1000 seed mass (g), % seed set and % hollow seeds at different planting densities in the intermediate sunflower cultivar SNK 32 at Vermaas during the 1989/90 sunflower season

Planting	Head	Filled	Hollow	Papery	1 000	Seed	Seed	Hollow
density	diameter	seeds	seeds	seeds	seed	set	Yield	seeds
	(mm)				mass	(%)	kg/ha	(%)
					(g)			
POP 15	227	2056	109	280	66.68	84	1557	4
POP 30	186	1737	66	365	51.55	80	1836	3
POP 45	165	1453	86	356	48.27	76	1719	5
POP 60	153	1243	69	352	46.58	75	1600	4
POP 75	140	981	74	419	46.72	67	1567	5
POP 90	139	880	73	462	44.79	62	1462	5

**Table 39** Table of means of percentage filled seeds (seed set) in the dwarf sunflower cultivar PNR 7460 at different planting densities at Vermaas during the 1989/90 sunflower season

POP 15       74.37         POP 30       71.42         POP 45       66.84         POP 60       69.39         POP 75       56.18         POP 90       65.30         Minimum       34.30         Maximum       90.90         Total values       180         Missing values       0         Mean replicate 1       67.51         Mean replicate 2       67.05	Plant density	Mean
POP 45       66.84         POP 60       69.39         POP 75       56.18         POP 90       65.30         Minimum       34.30         Maximum       90.90         Total values       180         Missing values       0         Mean replicate 1       67.51         Mean replicate 2       67.05	POP 15	74.37
POP 60       69.39         POP 75       56.18         POP 90       65.30         Minimum       34.30         Maximum       90.90         Total values       180         Missing values       0         Mean replicate 1       67.51         Mean replicate 2       67.05	POP 30	71.42
POP 75       56.18         POP 90       65.30         Minimum       34.30         Maximum       90.90         Total values       180         Missing values       0         Mean replicate 1       67.51         Mean replicate 2       67.05	POP 45	66.84
POP 9065.30Minimum34.30Maximum90.90Total values180Missing values0Mean replicate 167.51Mean replicate 267.05	POP 60	69.39
Minimum34.30Maximum90.90Total values180Missing values0Mean replicate 167.51Mean replicate 267.05	POP 75	56.18
Maximum90.90Total values180Missing values0Mean replicate 167.51Mean replicate 267.05		65.30
Total values180Missing values0Mean replicate 167.51Mean replicate 267.05	Minimum	34.30
Missing values0Mean replicate 167.51Mean replicate 267.05	Maximum	90.90
Mean replicate 167.51Mean replicate 267.05		180
Mean replicate 2 67.05	Missing values	0
-	Mean replicate	1 67.51
	Mean replicate	2 67.05
Mean replicate 3 67.19	Mean replicate	3 67.19
Grand mean 67.25	Grand mean	67.25
LSDt (0.05) = 14.79	LSDt (0.05) =	14.79

LSDt (0.01) = 19.45

Effective standard errors of the mean (ESE) Planting density Replicates Replicates 30 60 ESE 1.686 1.192

Stratum	standard	errors	and	coefficients	of	variation
Strat *units		df 172		SE 9.235		CV%
~units	<u>^</u>	1/2		9.235	13	• /

**Table 40** ANOVA table of percentage filled seeds (seed set) in the dwarf sunflower cultivar PNR 7460 at different planting densities at Vermaas during the 1989/90 sunflower season

Source of variation	df	SS	SS%	MS	VR	F PR
Plant density	5	5976.47	28.94	1195.2	14.016	<.001
Replicates	2	6.57	0.03	3.28	0.038	0.962
Residual	172	14667.88	71.03	85.28		
Total	179	20650.91	100.00	115.37		
Grand Total	179	20650.91	100.00			
Grand mean		67	.25			
Total number of observ	vations	180	)			

	-	-35	1	*
-35	-	-30	0	
-30	-	-25	0	
-25	-	-20	2	**
-20	-	-15	6	****
-15	-	-10	16	****
-10	-	- 5	25	****
- 5	-	0	35	*****
0	-	5	37	******
5	-	10	37	********
10	-	15	14	****
15		20	6	****
20	-		1	*

Missing values : 0 Asterisk represents 1 unit

Figure 12 Histogram of residual for percentage filled seeds (seed set) in the dwarf sunflower cultivar PNR 7460 at different plant densities at Vermaas during the 1989/90 sunflower season.

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Miller-Levene test for homogeneity of variance.
This is distributed as a F-variate with 5 and 168 degrees of freedom: F = 2.49874
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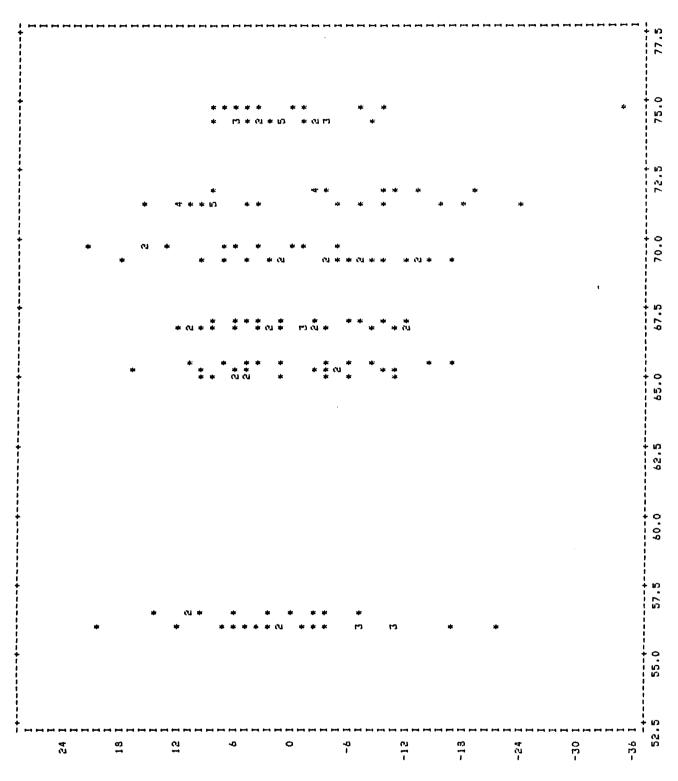


Figure 13 The residual values plotted against the fitted values in the dwarf sunflower cultivar PNR 7460 for the variant percentage seed set

Table 41 Table of means of percentage filled seeds (seed set) in the intermediate sunflower cultivar SNK 32 at different planting densities at Vermaas during the 1989/90 sunflower season

Plant density	Mean
POP 15	84.31
POP 30	80.13
POP 45	76.23
POP 60	74.55
POP 75	66.52
POP 90	61.97
Minimum	48.10
Maximum	91.40
Total values	180
Missing values	0
Mean replicate	1 73.64
Mean replicate	2 74.02
Mean replicate	3 74.19
Grand mean	73.95
LSDt (0.05) =	9.13
LSDt (0.01) =	12.02

Effective standard errors of the mean (ESE) Planting density Replicates Replicates 30 60 ESE 1.042 0.737

Stratum standard	errors	and	coefficients	of	variation
Stratum	df		SE		CV%
*units*	172		5.709	7	.7

Table 42 ANOVA table of percentage filled seeds (seed set) in the intermediate sunflower cultivar SNK 32 at different plant densities at Vermaas during the 1989/90 sunflower season

Source of variation	df	SS	SS%	MS	VR	F PR
Plant density	5	10487.64	65.13	2097.5	64.354	<.001
Replicates	2	9.39	0.06	4.70	0.144	0.866
Residual	172	5606.13	34.81	32.59		
Total	179	16103.17	100.00	89.96		
Grand Total	179	16103.17	100.00			
Grand mean		73	8.95			
Total number of observ	vations	180	)			

	-	-18	1	*
-18	-	-15	0	
-15	-	-12	4	***
-12	-	- 9	5	****
- 9	-	- 6	14	****
- 6	-	- 3	26	****
- 3	-	0	39	*****
0	-	3	41	*************
3	-	6	21	****
6	-	9	21	****
9	-	12	7	****
12	-	15	0	
15	-		1	*

Missing values : 0 Asterisk represents 1 unit

Figure 14 Histogram of residual for percentage filled seeds (seed set) in the intermediate sunflower cultivar SNK 32 at different plant densities at Vermaas during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 5 and 168 degrees of freedom: F = 2.72227

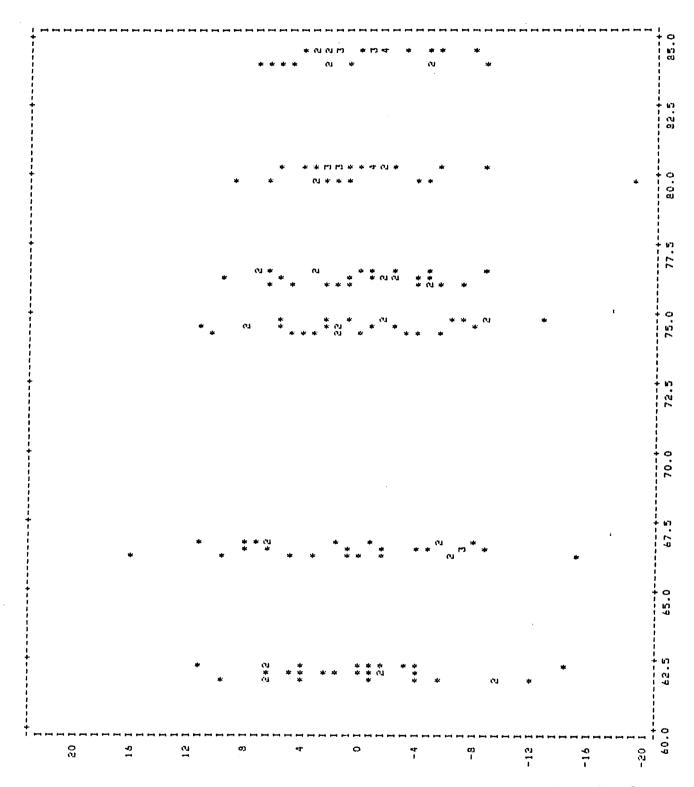


Figure 15 The residual values plotted against the fitted values in the intermediate sunflower cultivar SNK 32 for the variant percentage seed set **TABLE 43** Kruskal-Wallis analysis of variance of percentage hollow seeds in the dwarf sunflower cultivar PNR 7460 at different plant densities at Vermaas during the 1989/90 sunflower season

Plant density	Median	Inter qua	artile Class total
		range	
POP 15	4.70	2.25	3247.00
POP 30	2.50	1.05	1630.50
POP 45	4.15	0.85	2909.00
POP 60	3.10	0.80	2014.00
POP 75	4.15	1.85	2760.00
POP 90	6.00	1.40	3729.50

Chi-Squared value = 37.0710 5 Degrees of freedom Adjusted Chi-Squared value = 37.0941 5 Degrees of freedom

Table 44 Significance for all pairwise comparisons between different plant densities for the percentage hollow seeds in the dwarf sunflower cultivar PNR 7460, using Dunn's method

 POP 15
 POP 30
 POP 45
 POP 60
 POP 75

 POP 45
 \*
 \*
 \*
 \*

 POP 60
 \*
 \*
 \*
 \*

 POP 75
 \*
 \*
 \*
 \*

 POP 90
 \*\*
 \*
 \*
 \*

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 5 and 168 degrees of freedom: F = 3.60603

**TABLE 45** Kruskal-Wallis analysis of variance of percentage hollow seeds in the intermediate sunflower cultivar SNK 32 at different plant densities at Vermaas during the 1989/90 sunflower season

Plant density	Median	Inter qua	rtile Class total
		range	
POP 15	4.05	1.20	2630.00
POP 30	2.75	0.70	1374.50
POP 45	4.30	0.75	2913.50
POP 60	3.90	0.80	2577.00
POP 75	4.80	1.25	3380.00
POP 90	4.40	0.95	3415.00

Chi-Squared value = 34.3135 5 Degrees of freedom Adjusted Chi-Squared value = 34.3394 5 Degrees of freedom

Table 46 Significance for all pairwise comparisons between different plant densities for the percentage hollow seeds in the intermediate sunflower cultivar SNK 32, using Dunn's method

 POP 15
 POP 30
 POP 45
 POP 60
 POP 75

 POP 30
 \*
 \*\*
 \*\*
 \*\*

 POP 60
 \*
 \*\*
 \*\*
 \*\*

 POP 75
 \*\*
 \*\*
 \*\*
 \*\*

 POP 90
 \*\*
 \*\*
 \*\*
 \*\*

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 5 and 168 degrees of freedom: F = 1.61472 **Table 47** Table of means of percentage papery (dull) seeds in the dwarf sunflower cultivar PNR 7460 at different plant densities at Vermaas during the 1989/90 sunflower season

Plant density	Mean
POP 15	20.17
POP 30	25.21
POP 45	28.45
POP 60	27.06
POP 75	39.05
POP 90	28.87
Minimum	7.10
Maximum	62.70
Total values	180
Missing values	0
Mean replicate	1 27.94
Mean replicate	2 28.61
Mean replicate	3 27.86
Grand mean	28.14
LSDt (0.05) =	14.97
LSDt (0.01) =	19.71

Effective standard errors of the mean (ESE) Plant density Replicates Replicates 30 60 ESE 1.708 1.208

Stratum	standard	errors	and	coefficients	of	variation
Strat *units		df 172		SE 9.357	33	CV% .3

**Table 48** ANOVA table of percentage papery (dull) seeds in the dwarf sunflower cultivar PNR 7460 at different plant densities at Vermaas during the 1989/90 sunflower season

Source of variation	df	SS	SS%	MS	VR	F PR
Plant density	5	5788.08	27.74	1157.6	13.22	<.001
Replicates	2	20.37	0.10	10.18	0.116	0.890
Residual	172	15059.88	72.17	87.56		
Total	179	20868.34	100.00	116.58		
Grand Total	179	20868.34	100.00			
Grand mean		28.	14			
Total number of observ	vations	180	)			

	-	-16	6	****
-16	-	-12	7	****
-12	-	- 8	19	****
- 8	-	- 4	31	*****
- 4	-	0	28	*****
0	-	4	32	****
4	-	8	23	****
8	-	12	13	* * * * * * * * * * * *
12	_	16	13	****
16	-	20	4	* * * *
20	-	24	2	* *
24	_	28	1	*
28	_		1	*

Missing values : 0 Asterisk represents 1 unit

Figure 16 Histogram of residual for percentage papery (dull) seeds in the dwarf sunflower cultivar PNR 7460 at different plant densities at Vermaas during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 5 and 168 degrees of freedom: F = 1.53229

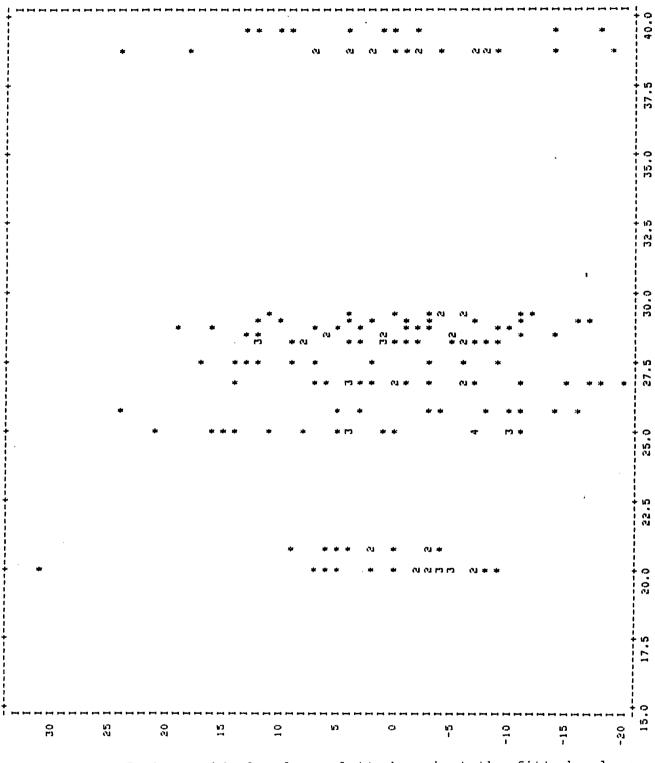


Figure 17 The residual values plotted against the fitted values in the dwarf sunflower cultivar PNR 7460 for the variant percentage dull seeds

**Table 49** Table of means of percentage papery (dull) seeds in the intermediate sunflower cultivar SNK 32 at different plant densities at Vermaas during the 1989/90 sunflower season

Plant density	Mean			
POP 15	11.35		-	
POP 30	16.96			
POP 45	19.21			
POP 60	21.26			
POP 75	28.30			
POP 90	32.87		_	
Minimum		5.60		
Maximum Tatal waluar		47.10		
Total values Missing values	-	180 0		
Mean replicate	1	21.64		
Mean replicate		21.84		
Mean replicate		21.49		
Grand mean		21.66		
LSDt (0.05) =	8.76			
LSDt (0.01) =	11 53			
	11.00			
Effective stand	lard err	ors of	the mean	(ESE)
Plan	it densi	ty R	eplicates	
Replicates	30		60	
ESE	0.999		0.70	06

Stratum	standard	errors	and	coefficients	of	variation
Strat	um	df		SE		CV%
*units	5*	172		5.472	25	.3

**Table 50** ANOVA table of percentage papery (dull) seeds in the intermediate sunflower cultivar SNK 32 at different plant densities at Vermaas during the 1989/90 sunflower season

Source of variation	df	SS	SS%	MS	VR	F PR
Plant density	5	9136.13	63.94	1827.2	61.029	<.001
Replicates	2	3.76	0.03	1.88	0.063	0.939
Residual	172	5149.72	36.04	29.94		
Total	179	14289.62	100.00	79.83		
Grand Total	179	14289.62	100.00			
Grand mean		21	.66			
Total number of observ	vations	180	)			

	12.5 1	*
-12.5	10.0 4	***
-10.0	7.5 7	*****
- 7.5	5.0 21	****
- 5.0	2.5 23	****
- 2.5 -	0 41	*****
0 –	2.5 31	*****
2.5 -	5.0 18	****
5.0 -	7.5 19	*****
7.5 -	10.0 8	*****
10.0 -	12.5 3	***
12.5 -	15.5 3	***
15.5 -	1	*

Missing values : 0 Asterisk represents 1 unit

Figure 18 Histogram of residual for percentage papery (dull) seeds in the intermediate sunflower cultivar SNK 32 at different plant densities at Vermaas during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 5 and 168 degrees of freedom: F = 5.26916

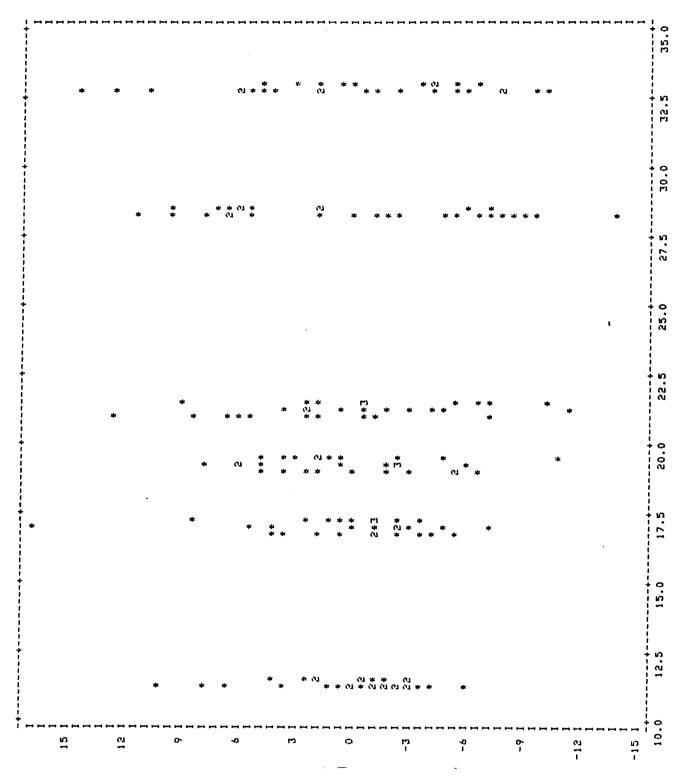


Figure 19 The residual values plotted against the fitted values in the intermediate sunflower cultivar SNK 32 for the variant percentage dull seeds

Table 51Table of means of the 1000 seed mass in the dwarfsunflower cultivar PNR 7460 at different plant densities at Vermaasduring the 1989/90 sunflower season

Plant density	Mean
POP 15	83.78
POP 30	52.88
POP 45	51.03
POP 60	47.97
POP 75	44.98
POP 90	48.00
Minimum	30.80
Maximum	128.80
Total values	180
Missing values	0
Mean replicate	1 56.59
Mean replicate	2 53.72
Mean replicate	3 54.01
Grand mean	54.77
LSDt (0.05) =	14.62
LSDt (0.01) =	19.25

Effective standard errors of the mean (ESE)

	Plant density	Replicates
Replicates	30	60
ESE	1.668	1.180

Stratum standard errors and coefficients of variation

Stratum	df	SE	CV%
*units*	172	9.138	16.7

**Table 52** ANOVA table of the 1000 seed mass in the dwarf sunflower cultivar PNR 7460 at different plant densities at Vermaas during the 1989/90 sunflower season

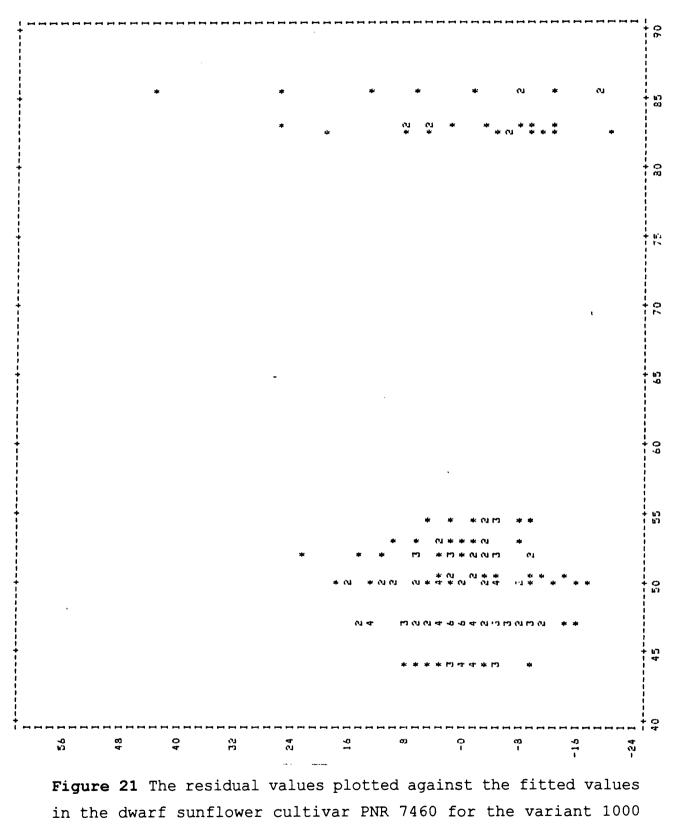
Source of variation	df	SS	SS%	MS	VR	F PR
Density	5	31416.52	68.18	6283.3	75.250	<.001
Replicates	2	298.10	0.65	149.05	1.785	0.171
Residual	172	14361.77	31.17	83.50		
Total	179	46076.38	100.00	257.41		
Grand Total	179	46076.38	100.00			
Grand mean		54.	.77			
Total number of obser	vations	180	)			

	-	-18	3	***
-18	-	-12	9	****
-12	-	- 6	27	*******
- 6	-	0	54	***************************************
0	-	6	50	**********
6	-	12	20	****
12	-	18	12	****
18	-	24	2	**
24	-	30	2	**
30	-	36	0	
36	-	42	0	
42	-	48	1	*
48	-		0	

Missing values : 0 Asterisk represents 1 unit

Figure 20 Histogram of residual of the 1000 seed mass in the dwarf sunflower cultivar PNR 7460 at different plant densities at Vermaas during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 5 and 168 degrees of freedom: F = 5.93406



seed mass

Table 53 Table of means of the 1000 seed mass in the intermediate sunflower cultivar SNK 32 at different plant densities at Vermaas during the 1989/90 sunflower season

Plant density	Mean	
POP 15	66.68	—
POP 30	51.55	
POP 45	48.27	
POP 60	46.58	
POP 75	46.72	
POP 90	44.79	<u> </u>
Minimum	27.90	
Maximum	91.50	
Total values	180	
Missing values	0	
Mean replicate		
Mean replicate		
Mean replicate		
Grand mean	50.77	_
LSDt (0.05) =	13.07	
LSDt (0.01) =	17.21	
Effective stand	lard errors of	the mean (ESE)
Plan	t density R	Replicates
Replicates	30	60
ESĒ	1.493	1.055

Stratum standard errors and coefficients of variation

Stratum	df	SE	CV%
*units*	172	8.176	16.1

Table 54ANOVA table of the 1000 seed mass in the intermediatesunflower cultivar SNK 32 at different plant densities at Vermaasduring the 1989/90 sunflower season

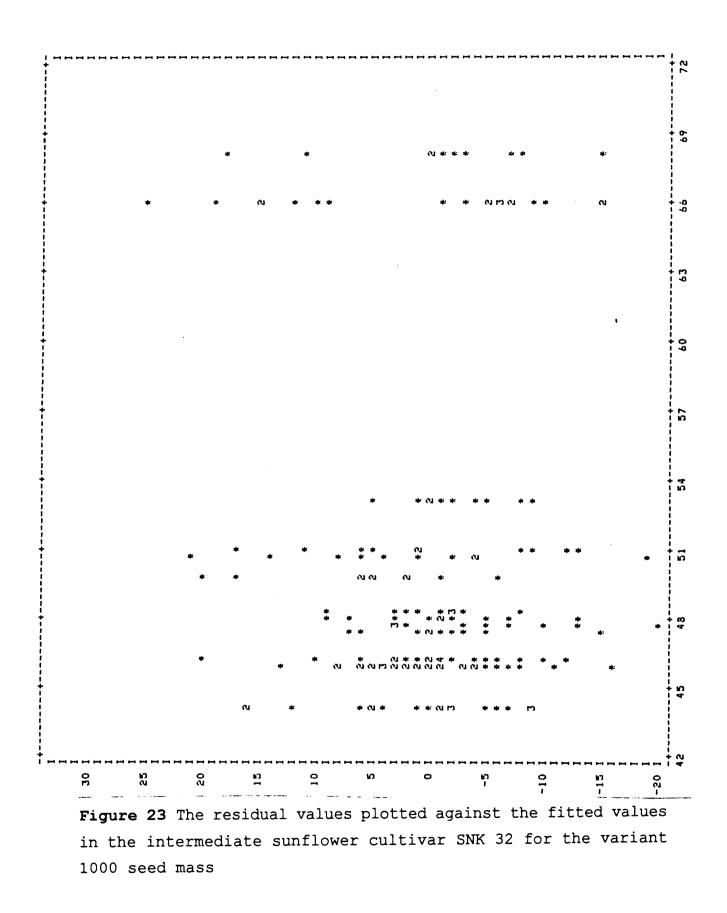
Source of variation	df	SS	SS%	MS	VR	F PR
Density	5	9893.78	45.85	1978.7	29.603	<.001
Replicates	2	185.86	0.86	92.93	1.390	0.252
Residual	172	11496.94	53.28	66.84		
Total	179	21576.58	100.00	120.54		
Grand Total	179	21576.58	100.00			
Grand mean		50	.77			
Total number of observ	ations	180	)			

	-	-16	2	**
-16	-	-12	9	****
-12	-	- 8	16	****
- 8	-	- 4	29	*****
- 4	-	0	41	******
0	-	4	37	******
4	-	8	22	****
8	-	12	9	****
12	-	16	7	****
16	-	20	5	****
20	-	24	2	**
24	-	28	1	*
28	-		0	

Missing values : 0 Asterisk represents 1 unit

Figure 22 Histogram of residual of the 1000 seed mass in the intermediate sunflower cultivar SNK 32 at different plant densities at Vermaas during the 1989/90 sunflower season.

Miller-Levene test for homogeneity of variance. This is distributed as a F-variate with 5 and 168 degrees of freedom: F = 3.71786



## CHAPTER 5

# INVESTIGATION INTO POLLEN QUALITY IN SUNFLOWER

### 5.1. Introduction

### 5.1.1. Sunflower pollination

The influence of environmental conditions, agronomical practices and cultivar morphological characteristics on the potential yield in sunflower is well researched (Bonari, Vannozzi, Benvenuti & Baldini, 1992; Knowles, 1978; Robinson, 1978; Fick, 1978). This was also the subject of recent studies on sunflower production in South Africa (Loubser, 1983; Loubser, 1991; Loubser, Grimbeek & Bronkhorst, 1988; Loubser, Grimbeek & Bronkhorst, 1990; Van der Merwe, 1988). Self-compatibility and self-fertilization has long been recognised as a genetically inherited characteristic of sunflowers (Luciano, Kinman & Smith, 1965; Olivieri, Lucchin & Parrini, 1988; Schuster, 1985; Vranceanu, Stoenescu & Scarlet, 1978). Insects are the agents of pollination and their effect on the potential yield has been reported on in South Africa (Birch, Van der Sandt, Herrmann & Johannsmeier, 1985; Du Toit, 1990; Du Toit & Holm, 1992 a+b). Engelbrecht (1996) commented on climatic and agronomical factors that influenced sunflower pollination in

South Africa. Researchers agree that xenogamy (crossachieving pollination) is more effective in successful fertilization than geitonogamy (selfing). According to Schuster (1985) anther and pollen development in sunflower is much more vulnerable to environmental conditions than embryogenesis. This "weakness" in pollen results in male sterility. Male sterility was therefore studied intensively by geneticists, as it provided a unique tool in sunflower breeding (Knowles, 1978; Fick, 1978). Environmental stresses during pollen development, germination and pollen tube growth also affected the functioning of the pollen and eventually seed set (Shivanna, Linskens & Cresti, 1991). Surprisingly little attention has been given to the physiology of sunflower pollen, as this could be an important limiting factor in the potential yield for commercial sunflowers. A possible reason for this neglect could be because of sunflowers' ability to partially compensate in seed weight where fewer seeds are produced, therefore masking the effect of poor pollen quality. A further reason for this lack in sunflower pollen research is It is well that the pollen is released in the trinucleate state. known that the physiology of trinucleate pollen is much more complex than that of binucleate pollen (Brewbaker, 1967; Hoekstra & Bruinsma, 1980).

#### 5.1.2. Trinucleate pollen

Most flowering plants shed their pollen, prior to sperm cell formation, in the binucleate stage. According to Brewbaker

(1967), however, some 30% of all flowering plants shed their pollen in the trinucleate stage. This follows after the mitotic production of the second sperm cell. Therefore, the trinucleate pollen incorporates the twin sperm cells and the vegetative nucleus (Brewbaker, 1967). It is further suggested that the condition is phylogenetically advanced. trinucleate All investigated species of the Asteraceae have trinucleate pollen (Brewbaker, 1967). Trinucleate pollen respires at much higher rates in humid air than the binucleate type. This is associated with rapid loss of pollen vitality (Hoekstra & Bruinsma, 1980). Trinucleate pollen has fully developed mitochondria at dehiscence, enabling it to germinate more rapidly on the stigma. In contrast, plant species with binucleate pollen show various stages of mitochondrial development which result in delayed pollen tube emergence.

In vitro germination of pollen provides a useful tool for studying pollen physiology, which in turn can be used to obtain reliable viability assays (Leduc, Monnier & Douglas, 1990). The classical Brewbaker and Kwack medium (Brewbaker & Kwack, 1963) is simple and is used extensively for the study of binucleate At the same time it has demonstrated the essential pollen. nature of calcium, boric acid and sucrose in pollen germination. This medium, however, is not suitable for germinating trinucleate Pollen tubes are abnormally short and reduced to pollen. (Leduc <u>et al</u>., 1990). Various media with protuberances supplementing components have been formulated since, to enhance germination of trinucleate pollen. Supplementing the media with

casein hydrolysate, an amino acid, improved germination of tobacco pollen (Tupy, Hrabetova & Capkova, 1983). Polyethylene glycol (PEG) improved pollen tube growth in <u>Petunia</u> (Zhang & Croes, 1982). Leduc <u>et</u>. <u>al</u>. (1990) found the presence of vitamins B1 and B6 to be stimulatory to the in vitro germination of trinucleate pollen. Micro-elements such as calcium, magnesium and boron also have an essential function in pollen germination (Brewbaker & Kwack, 1963; Leduc <u>et al</u>., 1990).

#### 5.1.3. Pollen viability and time

Dehiscence and release of sunflower pollen occur normally within 24 hours after anthesis (Knowles, 1978). Pollen is shed inside the anther tube formed by the fused anthers. The growing style pushes the pollen through the anther tube. As the sunflower is protandrous, the freshly released pollen is available to pollinate the receptive stigmatic lobes of neighbouring florets where anthesis had occurred the previous day. A carrier agent needed, however. The process of pollination and was fertilization can be completed within 24 hours. Researchers have where pollination and fertilization was found that, not successful, the stigmatic lobes could remain receptive for pollen germination for several days. Failure to successfully pollinate the stigmas, which otherwise would result in fertilization of the ovules, can be caused by a lack of pollinating agents or adverse weather conditions. Although it is known that pollen can be successfully stored for 20-25 days at 4-5°C and a relative

humidity of less than 40% (Frank, Barnabas, Gal & Farkas, 1982), it is uncertain for what period sunflower pollen remains viable under field conditions.

The aim of this study was to investigate different germination mediums for their suitability to understand the germination physiology of sunflower pollen. Secondly the viability of sunflower pollen was studied using different cultivars, and thirdly, the longevity of sunflower pollen was determined.

#### 5.2. Methods

#### 5.2.1. Pollen germination

The standard Brewbaker and Kwack medium (Brewbaker & Kwack, 1963) was used. This basal medium in distilled water consists of:

- 10% sucrose
- 100 ppm H<sub>3</sub>BO<sub>3</sub>
- $300 \text{ ppm Ca}(NO_3)_2.4H_2O$
- 200 ppm  $MgSO_4.7H_2O$
- 100 ppm KNO<sub>3</sub>

The following variations of the basal medium was prepared:

- 1. varying sucrose concentrations from 10 40%;
- 2. varying sucrose concentrations plus 1.5% gelatine, pH = 4.3;
- 3. varying sucrose concentrations plus 1.5% gelatine, pH = 6.0;
- 4. varying sucrose concentrations plus 1.5% gelatine. Pollen was dehydrated for 24 hours in a desiccator, pH = 6.0;

- 5. varying sucrose concentrations plus 1.5% gelatine. Pollen was soaked in distilled water for five minutes, pH = 6.0;
- 6. varying sucrose concentrations plus 1.5% gelatine. Pollen was soaked in 8N NaOH for five minutes, pH = 14.0;
- 7. varying sucrose concentrations plus 1.5% gelatine. Pollen
  was soaked in diluted dish washing soap for five minutes, pH
  = 8.0;
- 8. varying sucrose concentrations plus 1.5% gelatine. Pollen
  was soaked in acetone for five minutes, pH = 7.0;
- 9. 10% sucrose plus varying PEG4000 concentrations, pH =7.0;
- 30% sucrose plus 1.5% gelatine and varying concentrations of boron.
- All treatments were replicated six times.

Fresh florets was collected daily from sunflower plants at 07:00 and transferred in a cooler bag with ice to the laboratory where it was stored in a fridge at  $2-4^{\circ}C$  for further use on the same The hanging drop technique of Stanley & Linskens (1974) was dav. used for all pollen germination tests. Using a small paint brush, the pollen was transferred from the anthers to a single drop of the prepared germination medium on a glass microscope cover slip. The cover slip was then turned upside-down and placed on a 15 mm open ended glass tube of 20 mm diameter. The glass tube was smeared evenly with vaseline (petroleum jelly) on both cut ends and placed on a glass microscope slide, creating a glass "chamber". A drop or two of the germination medium was placed at the bottom of the glass well. The vaseline was used to seal off the hanging drop chambers, preventing evaporation of the

germination medium during the period of incubation of 36 hours. After the required incubation period, the cover slip was transferred to a clean microscope slide and studied with an optical microscope to determine pollen germination and pollen tube growth.

#### 5.2.2. Pollen viability

Pollen viability in sunflower was studied as follows:

1. Pollen viability in the following 14 sunflower cultivars: SNK 32; SNK 37; SNK 22; SNK 33 AS 470; AS 543; AS 505 SO 222; SO 323; SO 306; SO 242 PNR 7204; PNR 7369 CAR 1012

- Pollen sampled from one single head but on consecutive days of flowering representing the developing of the capitulum (day 1 to day 7) (cultivar SO 323)
- 3. Pollen tested after different periods of storage in an airtight container in a fridge at approximately 5°C (cultivar SO 323):
  - 2 hours storage
  - 1 week storage
  - 1 year storage
  - 2 years storage.

Alexander's stain was used to determine viability and longevity

of pollen. The general procedure used was as follows:

- Pollen was collected from the different cultivars or from consecutive rings on the same head, and transferred in a cooler bag with ice to the laboratory.
- A stock solution of the stain was prepared as described by Alexander (1969)
- 3. A drop of the stock solution was applied to a glass slide
- 4. Pollen was transferred from the anthers to the solution on the glass slide
- 5. The drop was then covered with a cover slip and heated slightly over an alcohol flame
- 6. The prepared slide was then studied with an optical microscope to determine pollen viability
- 7. Each treatment was replicated five times.

As in all other Asteraceae, the sunflower pollen is dimorphic and we distinguish between small and large grains based on size. The viability in large and small grains were scored separately.

#### 5.3. Results

No success was achieved in germinating sunflower pollen <u>in vitro</u> with any of the ten treatments described in the methods (Tables 70 to 83). Protuberances were visible but they were not scored as germinated. To be scored as germinated, a pollen tube of at least twice the diameter of the pollen grain itself had to have emerged. Plasmolysis of pollen grains did however occur in 42% of pollen grains (n = 1569). Plasmolysed pollen grains could be distinguished by the absence of nuclei in the tubes, and more than one tube from a single grain.

Pollen viability among the fourteen sunflower cultivars showed a figure of 100% in two cultivars (AS 505 and SO 242) and above 95% in all other cultivars with the lowest recorded percentage that of CAR 1012 (95.6%) (Table 81). Pollen viability was higher in the larger grains than in the smaller grains.

Pollen viability from different stages of the developing capitulum were determined (Table 82). No large differences occurred between viability among large and small grains. Pollen viability did not differ between the different days after flowering commenced. The ratio between large and small pollen grains ranged between 32% and 42% large pollen grains for the different cultivars (Table 82).

Sunflower pollen could be stored successfully for one season. Viability was 90.3% after one week's storage at 5°C and decreased only to 85% after one year's storage (Table 83). Viability did, however, decreased markedly after two year's storage. The viability of the small pollen grains decreased drastically. It had decreased to 84% after only one week's storage. This further decreased to 76.0% after one year's storage and only 11.0% remained viable after two years (Table 83). In contrast the large grains maintained reasonably good viability, 98% after one week's storage, 95% after one year and 77% after two years.

#### 5.4. Discussion

This study confirmed the difficulty to germinate and study sunflower (trinucleate) pollen in vitro. Hoekstra & Bruinsma (1980) explained the advantages of soaking pollen in organic solvents without any effect on vitality. Soaking had the purpose of removing the sticky lipophilic material from the pollen sporoderm. This was also performed during this study for in sunflower pollen, without any success improving its germination. Brewbaker & Kwack (1963) postulated that the pollen population of a test sample could have an effect on its germination. Small populations of less than 20 pollen grains germinated poorly. This can well be true for sunflowers as it is known that a large number of pollen grains must be transferred to the stigma before fertilization takes place. This indicates some unidentified "pollen growth factor" necessary for sunflower pollen to germinate. Such a "pollen growth factor" was presumably absent from the prepared growth media. The importance of boron in pollen germination and pollen tube growth was demonstrated by Vasil (1964) and Schmucker (1935),as satisfactory levels of boron are needed in the stigma exudate as well as in the stigma, style and ovary. These conclusions are supported by Lewis (1954) who found that pollen germination in the Asteraceae is regulated by the stigmatic surface, as germination was suspended in the stigmatic surface. Frankel & Galum (1977) concluded that trinucleate pollen was probably more sensitive to dehydration, radiation and long storage compared to

binucleate pollen. They attribute this to the less pronounced exine, and the exhaustion of the pollen grain of its reserves because of the second mitotic division. Pollen tube growth would then be dependent upon the style to support its growth. Pollen germination and pollen tube penetration in the style are the firsts steps to be achieved before successful fertilization can occur. It is suggested that future studies be concentrated on <u>semi in vivo</u> germination of sunflower pollen on a severed stigma and style implanted on different agar media to study the pollen physiology. Xanthopoulus (1991), having studied the pollen germination rate <u>in situ</u> on sunflower heads, expressed pollen germination in terms of the percentage seed set that was achieved.

The physiology of trinucleate pollen is still a new field of study. Various researchers have studied different aspects of trinucleate pollen. Shivanna <u>et</u>. <u>al</u>. (1991) studied the environmental stress conditions of Relative Humidity and high temperature in Nicotiana tabacum. Though none of these stress factors affected pollen viability, pollen vigour was markedly affected. Pollen grains subjected to high RH at 38°C took longer to germinate and pollen tubes emerged after a longer time. A decrease in seed set when high temperatures and low RH prevailed during pollination, had often been reported by sunflower growers in South Africa. This could well be because of the inability to produce adequate amounts of viable pollen. The same phenomenon was reported in maize where temperatures above 38°C greatly reduced pollen viability (Schoper, Lambert & Vasilas, 1987).

Staining with Alexander's stain indicated that sunflower had high viability percentages among cultivars investigated. Short term storage in a protected environment did not adversely effect viability. Long term storage indicated the rapid breakdown in viability. This was more pronounced in the small grains. It is postulated here that the thicker exine of the larger pollen grains is an evolutionary mechanism to ensure fertilization. The small pollen grains are thus more susceptible to harsh environmental conditions. The influence of radiation and ultraviolet rays will require further investigation. Furthermore, it is postulated that the "pollen growth factor" is present in the stigma and that the small pollen grains carry the unlocking mechanism, as they would benefit most by this to secure successful pollination. The quantity of pollen produced by the different cultivars and by different florets on the same capitulum also require further investigation.

#### 5.5. Conclusions

- No method and / or medium could be found to successfully germinate the trinucleate sunflower pollen in the absence of the stigma and style.
- The presence of a "pollen growth factor" is postulated as it is known that a large number of pollen grains are required for successful pollination.

- 3. It is here suggested that such a "pollen growth factor" would be present in the stigma as pollen could not be germinated in the absence of the stigma.
- 4. It is further postulated that the small pollen grains carry the ability to "unlock" or "activate" the pollen growth factor, as they would benefit most by this to secure successful pollination.
- 5. Viability of the dimorphic sunflower pollen is high within all cultivars investigated.
- Smaller pollen grains are more susceptible to long term storage, i.e. in their vitality.

**Table 55** The influence of sucrose on germination of sunflower pollen after two hours using the standard Brewbaker medium, incubated at 28°C and artificial illumination

010	Slide 1		Slide 2		Slide 3		Slide 4		Slide 5		Slide 6		Total		010
su	G	NG	G	NG	G										
10%	0	47	0	98	0	92	0	92	0	59	0	79	0	467	0
20%	0	29	0	57	0	20	0	23	0	17	0	25	0	171	0
30%	0	23	0	18	0	28	0	31	0	37	0	57	0	194	0
40%	0	30	0	20	0	23	0	18	0	36	0	48	0	175	0

G = germinated

NG = non-germinated

**Table 56** The influence of sucrose on germination of sunflower pollen after four hours using the standard Brewbaker medium, incubated at 28°C and artificial illumination

010	Slide 1		Slic	Slide 2		Slide 3		Slide 4		Slide 5		Slide 6		Total	
su	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G
10%	0	26	0	27	0	18	0	26	0	28	0	28	0	153	0
20%	0	29	0	32	0	42	0	42	0	47	0	25	0	217	0
30%	0	35	0	17	0	35	0	37	0	23	0	38	0	185	0
40%	0	17	0	24	0	19	0	19	0	11	0	13	0	103	0

G = germinated

Table 57	The influence of sucrose + 1.5% gelatine on germination of sunflower pollen after 3 hours
using the s	standard Brewbaker medium, pH 4.3 and incubated at 28°C and artificial illumination. The
number of	small pollen grain are presented in parentheses

0	Slid	e 1	Slic	de 2	Slic	de 3	Slid	de 4	Slic	le 5	Sli	de 6	То	tal	olo
su	G	NG	G	NG	G	NG	G								
20%	0	39 (14)	0	50 (10)	0	48 (15)	0	21 (7)	0	23 (10)	0	27 (8)	0	208	0
22%	0	35 (12)	0	33 (6)	0	29 (10)	0	19 (10)	0	17 (5)	0	26 (6)	0	159	0
25%	0	16 (7)	0	21 (11)	0	27 (7)	0	19 (4)	0	18 (9)	0	15 (4)	0	116	0
28%	0	14 (7)	0	15 (3)	0	25 (9)	0	22 (3)	0	17 (4)	0	28 (11)	0	121	0
308	0	14 (9)	0	19 (12)	0	30 (12)	0	40 (16)	0	33 (16)	0	26 (8)	0	162	0
32%	0	41 (21)	0	37 (19)	0	32 (16)	0	65 (31)	0	38 (16)	0	38 (19)	0	251	0
35%	0	32 (18)	0	24 (13)	0	27 (10)	0	21 (8)	0	19 (8)	0	33 (24)	0	156	0
38%	0	24 (14)	0	27 (11)	0	27 (10)	0	20 (11)	0	19 (12)	0	34 (14)	0	151	0
40%	0	28 (12)	0	35 (10)	0	40 (22)	0	55 (29)	0	50 (26)	0	37 (18)	0	245	0

G = germinated NG = non-germinated

010	Slic	de 1	Slic	le 2	Slid	le 3	Slid	le 4	Slic	le 5	Slic	le 6	Tot	cal	0 <sup>0</sup>
su	G	NG	G	NG	G										
20%	0	46	0	55	0	55	0	21	0	23	0	31	0	231	0
22%	0	35	0	36	0	34	0	19	0	17	0	29	0	170	0
25%	0	22	0	26	0	29	0	21	0	22	0	17	0	137	0
28%	0	18	0	18	0	28	0	23	0	19	0	34	0	140	0

Table 58The influence of sucrose + 1.5% gelatine on germination of sunflower pollen after 3 hoursusing the standard Brewbaker medium, pH 6.0 and incubated at 28°C and artificial illumination

G = germinated

NG = non-germinated

30%

32%

35%

38%

40%

**Table 59** The influence of sucrose + 1.5% gelatine on germination of sunflower pollen after 3 hours using the standard Brewbaker medium, pH 6.0 and incubated at 28°C and artificial illumination. Pollen was dehydrated for 24 hours prior to the germination test

010	Slide 1		Slide 2		Slide 3		Slide 4		Slid	Slide 5		Slide 6		Total	
su	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G
20%	0	32	0	45	0	40	0	21	0	23	0	23	0	184	0
22%	0	32	0	30	0	24	0	18	0	17	0	23	0	144	0
25%	0	15	0	15	0	22	0	17	0	27	0	34	0	130	0
288	0	11	0	17	0	19	0	20	0	15	0	17	0	99	0
30%	0	18	0	13	0	24	0	32	0	25	0	22	0	134	0
32%	0	30	0	27	0	25	0	49	0	30	0	28	0	189	0
35%	0	32	0	24	0	23	0	17	0	23	0	24	0	143	0
38%	0	17	0	22	0	21	0	13	0	15	0	27	0	115	0
40%	0	22	0	34	0	29	0	40	0	63	0	46	0	234	0

G = germinated

**Table 60** The influence of sucrose + 1.5% gelatine on germination of sunflower pollen after 3 hours using the standard Brewbaker medium, pH 6.0 and incubated at 28°C and artificial illumination. Pollen soaked in distilled water for five minutes prior to the germination test

0 0	Slide 1		Slide 2		Slide 3		Slide 4		Slid	Slide 5		Slide 6		Total	
su	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G
20%	0	41	0	36	0	13	0	40	0	32	0	21	0	183	0
22%	0	26	0	11	0	47	0	17	0	27	0	48	0	176	0
25%	0	32	0	45	0	32	0	19	0	14	0	18	0	160	0
28%	0	37	0	14	0	19	0	15	0	40	0	45	0	170	0
308	0	17	0	32	0	37	0	28	0	40	0	12	0	166	0
32%	0	27	0	16	0	22	0	22	0	19	0	16	0	122	0
35%	0	24	0	27	0	15	0	46	0	50	0	42	0	204	0
388	0	32	0	17	0	38	0	41	0	15	0	27	0	170	0
40%	0	31	0	17	0	27	0	25	0	21	0	27	0	148	0

G = germinated

**Table 61** The influence of sucrose + 1.5% gelatine on germination of sunflower pollen after 3 hours using the standard Brewbaker medium and incubated at 28°C and artificial illumination. Pollen soaked in 8N NaOH, pH 14, for five minutes prior to the germination test

010	Slic	le 1	Slic	le 2	Slid	е З	Slid	.e 4	Slid	e 5	Slid	e 6	Tot	al	010
su	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G
20%	0	55	0	26	0	28	0	40	0	32	0	19	0	200	0
22%	0	38	0	17	0	57	0	17	0	27	0	54	0	210	0
25%	0	39	0	56	0	39	0	23	0	23	0	22	0	202	0
28%	0	44	0	17	0	28	0	18	0	44	0	56	0	207	0
30%	0	26	0	44	0	49	0	44	0	56	0	20	0	239	0
32%	0	48	0	35	0	38	0	53	0	35	0	35	0	244	0
35%	0	42	0	40	0	25	0	56	0	58	0	66	0	287	0
38%	0	46	0	28	0	48	0	52	0	27	0	41	0	242	0
40%	0	43	0	27	0	49	0	54	0	47	0	46	0	266	0

G = germinated

**Table 62** The influence of sucrose + 1.5% gelatine on germination of sunflower pollen after 3 hours using the standard Brewbaker medium and incubated at 28°C and artificial illumination. Pollen soaked in dish washing soap, pH 8, for five minutes prior to the germination test

0 <sup>1</sup> 0	Slic	le 1	Slid	le 2	Slid	le 3	Slid	e 4	Slic	le 5	Slid	e 6	Tot	al	010
su	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G
20%	0	23	0	50	0	36	0	39	0	42	0	53	0	243	0
22%	0	25	0	20	0	32	0	24	0	92	0	44	0	237	0
25%	0	36	0	33	0	27	0	23	0	39	0	25	0	183	0
28%	0.	25	0	27	0	10	0	25	0	14	0	57	0	158	0
30%	0	30	0	23	0	23	0	25	0	70	0	13	0	184	0
32%	0	13	0	13	0	27	0	17	0	43	0	46	0	159	0
35%	0	14	0	18	0	33	0	71	0	35	0	46	0	217	0
38%	0	24	0	23	0	23	0	60	0	21	0	31	0	182	0
40%	0	17	0	18	0	12	0	12	0	37	0	21	0	117	0

G = germinated

**Table 63** The influence of sucrose + 1.5% gelatine on germination of sunflower pollen after 3 hours using the standard Brewbaker medium and incubated at 28°C and artificial illumination. Pollen soaked in acetone, pH 7, for five minutes prior to the germination test

010	Slic	de 1	Slid	le 2	Slid	le 3	Slid	e 4	Slic	le 5	Slid	e 6	Tot	al	olo
su	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G
208	0	19	0	29	0	24	0	26	0	22	0	59	0	179	0
228	0	21	0	45	0	39	0	40	0	42	0	24	0	211	0
25%	0	39	0	54	0	45	0	41	0	41	0	33	0	253	0
28%	0	56	0	40	0	43	0	22	0	36	0	45	0	242	0
30%	0	21	0	53	0	66	0	31	0	20	0	43	0	234	0
32%	0	55	0	47	0	33	0	24	0	55	0	49	0	263	0
35%	0	44	0	42	0	58	0	33	0	56	0	49	0	282	0
38%	0	29	0	18	0	34	0	27	0	48	0	33	0	189	0
40%	0	43	0	37	0	25	0	38	0	32	0	36	0	211	0

G = germinated

Table 64	The influence	e of PEG 4000 on	germination of	sunflower pollen after	3 hours using the
standard Br	ewbaker medium	with 10% sucros	e and incubated	at 28°C and artificial	illumination, pH 7

0 <sup>1</sup> 0	Slic	le 1	Slic	le 2	Slid	le 3	Slic	le 4	Slid	e 5	Slid	e 6	Tot	cal	0 <sup>j</sup> 0
PEG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G
08	0	26	0	17	0	33	0	25	0	35	0	59	0	195	0
5%	0	20	0	40	0	45	0	19	0	26	0	24	0	174	0
10%	0	44	0	19	0	16	0	29	0	18	0	33	0	159	0
15%	0	28	0	36	0	26	0	29	0	33	0	45	0	197	0
20%	0	19	0	24	0	20	0	17	0	29	0	31	0	140	0
25%	0	37	0	24	0	21	0	72	0	30	0	35	0	219	0
30%	0	25	0	36	0	31	0	22	0	36	0	47	0	197	0
35%	0	26	0	15	0	30	0	76	0	39	0	53	0	239	0
40%	0	28	0	33	0	47	0	38	0	45	0	20	0	211	0

G = germinated
NG = non-germinated

# **Table 65** The influence of boron on germination of sunflower pollen after 3 hours using the standard Brewbaker medium with 30% sucrose plus 1.5% gelatine and incubated at 28°C and artificial illumination

В	Slic	le 1	Slid	e 2	Slid	.e 3	Slid	e 4	Slid	.e 5	Slid	e 6	Tot	al	00
ppm	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G	NG	G
0	0	13	0	30	0	20	0	26	0	20	0	16	0	125	0
25	0	19	0	41	0	38	0	37	0	24	0	64	0	223	0
50	0	33	0	34	0	29	0	26	0	21	0	28	0	171	0
75	0	52	0	40	0	56	0	27	0	30	0	27	0	232	0
100	0	21	0	49	0	29	0	24	0	23	0	57	0	203	0
250	0	41	0	31	0	39	0	36	0	45	0	39	0	231	0
500	0	30	0	28	0	20	0	29	0	40	0	45	0	192	0
750	0	21	0	17	0	23	0	18	0	18	0	29	0	126	0
1000	0	21	0	10	0	10	0	71	0	14	0	17	0	143	0
2500	0	25	0	18	0	14	0	24	0	24	0	35	0	140	0
5000	0	15	0	26	0	32	0	16	0	13	0	41	0	143	0

G = germinated

Table 66Percentage pollen viability determined with Alexander's vital stain in fourteen differentsunflower cultivars

Cultivars	% large viable pollen grains	% small viable pollen grains	% Total viability	n
SNK 32	98.5	97.5	96.9	414
SNK 37	100.0	96.4	97.5	371
SNK 22	97.8	98.7	98.4	533
SNK 33	100.0	97.8	98.6	653
AS 470	97.7	96.8	97.2	366
AS 543	100.0	99.2	99.6	342
AS 505	100.0	100.0	100.0	435
SO 222	97.2	97.0	97.1	442
SO 323	98.4	98.3	98.3	470
SO 306	100.0	98.7	99.2	394
SO 242	100.0	100.0	100.0	417
PNR 7204	98.6	98.7	98.6	422
PNR 7369	96.9	97.3	97.1	376
CAR 1012	91.8	97.5	95.6	429

Table 67Percentage pollen viability determined with Alexander's vital stain in florets from thesame head but on consecutive days of flowering

Days	% large viable pollen grains	% small viable pollen grains	% Total viability	n
Day 1	99.2 $(n = 132)$	96.6	97.9	399
Day 2	98.3 (n = 129)	97.3	97.8	372
Day 3	100.0 (n = 139)	96.8	98.4	394
Day 4	99.2 $(n = 125)$	98.0	98.6	385
Day 5	98.2 (n = 156)	98.7	98.4	410
Day 6	99.3 $(n = 167)$	97.6	98.3	407
Day 7	98.3 (n = 177)	96.2	97.2	419

Table 68Percentage pollen viability determined with Alexander's vital stain in the sunflowercultivar SO 323 after different periods of storage

Storage Period	% large viable pollen grains	% small viable pollen grains	% Total viability	n
2 hours	99.2	96.6	97.9	399
1 week	98.3	84.9	90.3	382
1 year	95.1	76.0	85.5	279
2 years	77.1	11.0	44.0	210

### RECOMMENDATIONS

Sunflower breeders and agronomists should take note of hollow seededness and unfilled seeds as a yield determining factor. This will vary between different localities, cultivars and plant density, while plant physiological characteristics such as pollen quality will further affect seed set.

- Breeders and agronomists need to identify cultivars that will perform constantly above average over a wide range of localities (climatic conditions and soil type). Maintaining specific cultivars for a narrow set of climatic conditions will not be cost effective, and limits the competitive edge.
- 2. Breeders and agronomists need to identify those characteristics in a cultivar that will provide a competitive edge, at an early stage of the developmental phase. Poor characteristics such as an infertile head centre and a high percentage of hollow seeds will reduce company profits as well as damage the company credibility.
- 3. Extension workers and agronomists should make growers aware of the higher demand in nutrients when planting density is increased. Fertilizer programmes need to be adjusted to higher planting densities.

### SUMMARY

Continuing problems with poor to average seed set in the sunflower producing areas in South Africa led to prior investigations into the role of honeybees in the pollination of commercial sunflower. Guidelines from these studies were implemented by farmers. It was realised that pollen transfer by honeybees and other insects, was just one process in the pollination ecology of sunflower. The present investigation was launched to better the knowledge and understanding of seed set problems by focussing on aspects such as hollow seededness, unfilled seeds and on pollen quality.

Hollow seededness could not be correlated to specific cultivar characteristics in the eighteen cultivars investigated. No significant differences could be found when the same cultivar was tested at seventeen localities in South Africa. It was confirmed that hollow seededness could be associated with plant density, because the percentage hollow seeds decreased when plant density increased. It is concluded that slow pollen movement, which results in self-pollination, is the main contributing factor in the phenomenon of hollow seededness.

There was no significant difference in the number of unfilled seeds between the eighteen cultivars investigated. However, the

number of unfilled seeds was unsatisfyingly high, between 5% and 14%. The percentage unfilled seeds differed significantly between the various localities investigated. A positive correlation was found between plant density and the number of unfilled seeds: the higher the plant density the higher the number of unfilled seeds. This confirmed reports that the increased competition for nutrients with increased plant density, resulted in a decrease in yield.

Boron levels differed significantly in soil, leaf and floret It was difficult to interpret these results, as samples. knowledge on the action of boron in sunflower is still lacking. Boron affects the reproductive processes of pollen germination, pollen tube growth, and seed set and fruit set. Boron thus has an indirect influence on hollow seededness and unfilled seed. Tt is important to investigate the role of boron in the various physiological processes associated with anthesis and fertilization further.

The presence of a "pollen growth factor" in the stigma is postulated, as no suitable method and / or medium could be identified to successfully germinate the trinucleate sunflower pollen, using conventional techniques. Pollen viability is high within the cultivars investigated. The dimorphic sunflower pollen is an adaptation to environmental conditions as the large pollen grain has a longer vitality.

## OPSOMMING

Voortslepende probleme met lae tot gemiddelde sonneblom opbrengste in die verskillende produksiegebiede van Suid-Afrika het gelei tot vorige ondersoeke na die rol van heuningbye in die bestuiwing van kommersiële sonneblom. Die aangewese riglyne is suksesvol deur boere geïmplementeer. Daar is egter besef dat die oordraging van stuifmeel deur heuningbye en ander insekte, slegs een van die prosesse in die bestuiwingskompleks van sonneblom is. Die huidige ondersoek is geloods om die probleme rondom saadset beter te verstaan. Daar is veral gefokus op aspekte soos holsadigheid, ongevulde sade en stuifmeelkwaliteit.

Agtien sonneblomkultivars is ondersoek en holsadigheid kon nie met spesifieke kultivar eienskappe gekorreleer word nie. Geen betekenisvolle verskille is by 'n enkele kultivar waargeneem waar dit by sewentien verskillende lokaliteite ondersoek is nie. Die studie het bevestig dat holsadigheid geassosieer kan word met plantdigtheid by beide die kultivars wat ondersoek was: soos plantdigtheid toeneem het die persentasie holsade afgeneem. Daar is tot die gevolgtrekking gekom dat stadige stuifmeel beweging en stuifmeelbuisgroei, wat tot selfbestuiwing lei, die hoof bydraende faktor is by die verskynsel van holsadigheid.

Daar was geen betekenisvolle verskil tussen die agtien kultivars

met betrekking tot holsade nie. Die getal ongevulde sade, wat gewissel het van 5 tot 14%, was egter onbevredigend hoog. Die persentasie ongevulde sade het betekenisvol verskil tussen die onderskeie lokaliteite. 'n Positiewe korrelasie is gevind tussen plantdigtheid en die aantal ongevulde sade: hoe hoër die plantdigtheid, hoe hoër die aantal ongevulde sade. Dit het verslae bevestig dat 'n toename in kompetisie vir voedingstowwe met 'n toename in plantdigtheid, tot gevolg het dat die oesopbrengs afneem.

Die boorvlakke in grond-, blaaren blomontledings het betekenisvol verskil. Dit was egter nie moontlik om hierdie resultate te ontleed nie, aangesien kennis van die interaksie van boor in sonneblom nog gebrekkig is. Boor affekteer verskeie van die reproduktiewe prosesse soos stuifmeelbuiskieming, stuifmeelbuisgroei en saadset en vrugset. Boor het dus wel 'n indirekte invloed op beide holsadigheid en ongevulde sade. Dit is belangrik dat die rol van boor in hierdie verskillende fisiologiese prosesse wat met blomontwikkeling en bevrugting geassosieer word, in toekomstige navorsing ondersoek sal word.

Die aanwesigheid van 'n "stuifmeel groeifaktor" word voorgestel, aangesien geen geskikte metode of medium, onder die konvensionele tegnieke, gevind kon word om die tri-nukluêre sonneblomstuifmeel te kiem nie. Stuifmeelkiemkragtigheid was hoog by al die kultivars wat ondersoek is. Die dimorfiese sonneblomstuifmeel is wel by omgewingstoestande aangepas aangesien die groot stuifmeelkorrels 'n langer lewenskragtigheid het.

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