ETIOLOGY AND CONTROL OF STEM ROT OF SUNFLOWER

(*Helianthus annuus* L.)

BY

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DECLARATION

I, Joel Bujulu, do hereby declare to the Senate of Sokoine University of Agriculture that this thesis is my own original work and that it has not been submitted, in whole or in part, for a degree award in any other University.

Signature : .....

Date : 26th March 1988

ABSTRACT

Surveys were carried out on the distribution in Tanzania of a new sunflower disease first observed in Morogoro Region. Experiments were conducted at the Tropical Pesticides Research Institute (T.P.R.I.) and Sokoine University of Agriculture (SUA) to identify the causal agents, assess their pathogenicity to sunflower varieties commonly grown in Tanzania and other crops normally grown in association with sunflower, and to determine biological activity of some selected fungicides to the disease causing organisms. Out of all eight surveyed regions, sunflower stem rot was confirmed only in Morogoro region. Fusarium moniliforme Sheld and F. graminearum Schwabe were identified as the causal organisms inciting the new sunflower disease which was later named "sunflower stem rot".

Sunflower varieties namely Record, Jupiter, Gor 104 and Dwarf comet were found less affected by both Fusarium spp. while Giant 549, IS 894, IS 894 amd 6F5 were more susceptible. Groundnuts, simsim, cowpeas and greengram did not show symptoms upon artificial inoculations with the two Fusarium spp. while maize, sorghum, beans and garden peas showed necrotic symptoms in the areas inoculated.

The fungicides Sisthane (fenapanil) and Ridomil (metalaxyl) were not effective against both fungi in laboratory culture tests while Benlate (benomyl) was effective. But Benlate proved inffective in controlling the disease in the field during field tests.

Severity of sunflower stem rot was more apparent in fields planted at very close spacing whether it was a monoculture or interplanted with other crops resulting in more plant deaths.

It is recommended that sunflower varieties which were found resistant be distributed to peasants in Morogoro Region and that only resistant crops like groundnuts, simsim, cowpeas [illegible] be used in intercropping. Wherever stem rot is endemic [illegible] lower and all other intercrops should be planted at wider spacing to avoid overcrowding which favour disease development. Further research is being suggested particularly on breeding for more resistant varieties.

The suitability of fungicide control of the disease in peasant grown sunflower appears remote and further investigations on this aspect should be on large farms.

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1. INTRODUCTION

Tanzania is an agricultural country with about 80% of her population dependent on agriculture. At present, both food and marketed cash crops form nearly 50% of the Gross Domestic Product (GDP) and 80% of the total export is accounted for by agricultural commodities (Anonymous, 1977). Hence agriculture is the main source of the nation's foreign exchange earnings.

Apart from the traditionally known crops like coffee, cotton, tea, tobacco, sisal and cashew, sunflower has of late gained a significant role in the Tanzania economy. As an oil crop which requires marginal soil fertility and is drought resistant, sunflower is grown extensively in more than 12 of the 20 regions in Tanzania mainland (Fig. 1, Appendix 3). Expanded crop production is most often accompanied by disease and insect pest incidence (Glasscock, 1947).

Yields of sunflower in Tanzania are still very low estimated at only 400 kg/ha while that of Europe averages at 1400 kg/ha and the world at above 1100 kg/ha (Appendix 2).

The low yields in Tanzania can be attributed to poor crop husbandry including irregular spacing,weeds and unavailability of improved seed varieties suitable for different agroecological zones.

All these bottlenecks coupled with frequent attack of existing sunflower varieiteis by insect pests and diseases have contributed in keeping yield figures pretty low.

Several insect pests have been reported on sunflower in Tanzania. Bohlen (1973) singled out two major pests: American Bollworm (Heliothis armigera) (Hb) Blue Bug (Calidea dregii) Germar. During the survey of major sunflower producing regions it was observed that mites (Tetranychus cinnabarinus) (Boisd) were causing extensive damage in many of the farms visited. Other insects found feeding on sunflower were : pollen beetles (Mylabria spp.), cluster bug (Agonoscelia pubescens) Thumb, green stink bug (Nezara viridula), cotton aphid (Aphis gossypii) and groundnut hopper (Hilda patruelis). Ismani Division of Iringa Region which is one of the most important sunflower growing areas was in the 1980 season heavily infested with Blue Bug and pollen beetles which caused considerable damage (Bujulu, 1980).

Diseases also exert a lot of influence on sunflower production. Riley (1960) reported the existence of mildew caused by Erysiphe cichoracearum DC and Leveillula taurica (Lev.) Arn., rust caused by Puccinia helianthi Schw. Sclerotinia wilt and head rot incited by Sclerotinia sclerotiorum (Lib.) de Bary (Whetzelinia sclerotiorum (Lib.) Korf and Domont), leaf spots caused by Alternaria helianthi (Hansf.) Tubaki and Nishihara, syn. Helminthosporium helianthi Hansf., and Septoria helianthi Ell. and Kell and sunflower brown rot caused by Pseudomonas solanacearum (E.F. Smith). Clinton (1961) also mentioned rust, mildew and leaf spots as diseases of sunflower in Tanzania but singled out sunflower rust as the most important at Mlingano, Tanga caused by Erwinia aroedeae. Allen (1974) isolated Phoma herbarum, Cochliobulus sp and Xanthomonas sp from leaf

spots. He also reported that marginal leaf blights were associated with Fusarium avenaccum, F. equiseti and F. semitectum. All these were new records for Tanzania on sunflower.

In 1975/76 season another serious disease was observed in Morogoro Region, the then largest sunflower producer (Appendix 3). The attack was causing severe rots on stems resulting in large scale loding. In 1980 a serious leaf blight outbreak caused by Alternaria sp. was observed in three divisions of Iringa Region (Bujulu, 1980). All these diseases appeared potentially threatening. Up till now not sufficient effort has been made to study the epidemiology of various disease pathogens of sunflower in Tanzania. This explains lack of recommended control measures for pests and diseases.

The new disease which was first noticed at the Dakawa sunflower farm was within a few weeks found to have spread in most fields in and around Morogoro township. It was attacking stems causing large necrotic areas which later expanded to cover lower parts of the stems. Such plants became prone to lodging and finally died.

The magnitude of this disease was alarming and it was believed that yield losses were likely to be very high and could badly affect the entire vegetable oil industry. This study project was therefore initiated at the Sokoine University of Agriculture with the following objectives:-

- to study the etiology of the disease
- to determine its distribution within all amjor sunflower producing regions and
- todevise possible control measures

2. LITERATURE REVIEW

2.1 The sunflower plant

Sunflower, Helianthus annuus L., belongs to the largest natural order of flowering plants, the Compositae. These are characterised by the crowding together individual flowers into heads surrounded by rosette of green bracts functioning like a calyx (Hurt, 1946). The plant is distinguished by its cylindrical, strong stalks which under normal crop densities grows to between 1.5 and 2 m tall. It bears 20 to 25 either toothed or untoothed and haired leaves growing alternatively from the stalk. Its terminal head is normally 10 to 15 cm across (Poole, 1975). The genus is subdivided into two groups; the annual which bears seeds and the perennial whose species are non seed bearers; like the Jerusalem artichoke (Helianthus tuberosus).

2.1.1 Origin and geographical distribution of sunflower

Th present commercial sunflower was first cultivated by North American Indians (Hurt, 1946). Its origin is believed to be Peru or Mexico (Hurt, 1946 and Glendon Hill, 1947). It was introduced to Europe in the sixteenth century by Spaniards (Hurt, 1946). From Spain, sunflower was then taken to Bavaria in 1625, to France in 1787, to Hungary and then to Russia and other parts of Europe (Hurt, 1946 ; Cramer, 1967 and Poole, 1975). Slowly sunflower then found its way to North America, Africa, Oceania and it is commercially grown in more than thirty countries (FAO Production Year Book, 1981).

The year of its introduction into East Africa is uncetain but Martens et al (1970) citing Holms (1925) acknowledged that sunflower had been grown in Kenya for many years.

Most of the world's sunflower is grown as a summer crop in temperate regions led by Russia which produces more than 50% of the total world production (Jensma, 1973). Appendix I gives the most important sunflower producers and Appendix 2 gives world regional total production.

2.1.2 Importance of sunflower production

Whenever sunflower is grown, whether on large or small scale, the main objective is the grain for the extraction of edible oil.

Other oil crops like groundnuts and soybeans are more expensive to grow and, actually, produce less oil per hectare than sunflower (Jensma, 1973). In addition to the above advantage, the sunflower is more resistant to drought and frost than related crops such as maize, sorghum and soybean (Jensma, 1973).

Demand for edible oil in Tanzania has risen very sharply in recent years. Monyo (1976) estimated that Tanzania would need 35000-40000 tons of edible oil by 1980. The rise is mainly attributed to the increase in population and to the fact that the populace have of late preferred refined oils to their traditional means of obtaining oils and fats. This phenomenon has aggravated the already acute shortage for oils and fats.

The most important oil crop, the groundnut (Arachis hypogaea) has been grown in Tanzania for many years but its prime use has not been the production of refined edible oil. Groundnuts have been and still are consumed whole after roasting or, pounded into powder for mixing in sauces and soups. It is also a common practice to use groundnuts for barter among neighbouring families. These practices are so common in Tanzania that very little crop is left for commercial sale. This trend is not only for groundnuts but also true for similar crops which are easily utilized before factory processing. These include simsim, coconuts, oil palm fruits and oyster nuts. The second most important oil crop, soybean, is not yet very popular in Tanzania.

Consequential to the fact stated above and the added sunflower quality of being very resistant to drought and not demanding high soil fertility, many regions have now embarked on expanded sunflower production (Appendix 3). In addition, the Government has also developed interest in sunflower growing, which is the most likely crop to alleviate edible oil shortages. Incentive to the prospective farmers has been created by price rises. Between 1976/77 and 1986/87 the official producer price for the mixed grade has risen by more than 900% (Appendix 4).

Production of sunflower in Tanzania, has, however, been fluctuating very considerably. Data so far available indicate that between 1980/1981 and 1984/1985 seasons yearly production has averaged at around 8,000 metric tons (Appendix 3). This indeed is not a great achievement considering the overall efforts being made politically and socially into the agricultural sector.

The tonnage has been very low and stagnant for many years. Unlike other related crops whose production figures are often greatly underquoted due to direct home consumption, the figures for sunflower grain are very close to real because very little of the grain is consumed without processing. That sunflower is difficult to use directly before sale has played a major part in keeping its hectarage low.

The only counter measure likely to increase production has rightly been the price increases. During the 1972/73 season the price for the mixed grade, which is the most common grade, among peasant farmers, was T.shs. 0.55 per kg and the production was 9464 tons but in the 1979/80 season when the price was raised to T.shs. 1.40 per kg of the same grade, production rose to approximately 15,560 tons (Appendices 3 and 4).

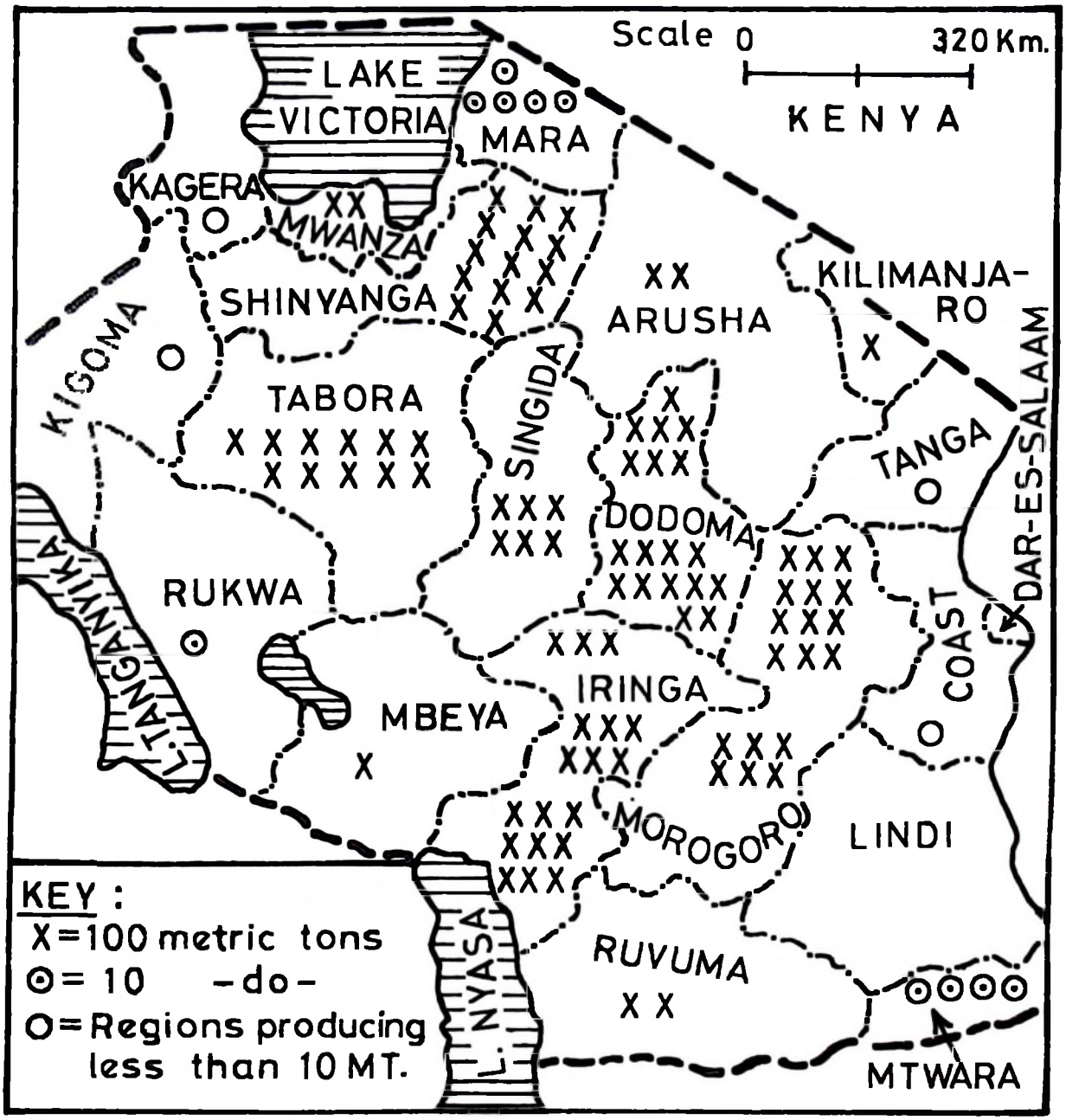
In the past almost all sunflower grain collected by GAPEX was exported to other countries. GAPEX itself had considerably a very minimal number of collecting centres and a small number of field staff. Therefore, sunflower was not bought from peasants immediately when grains were harvested. Storage facilities at village level in Tanzania are poor rendering crops which are vulnerable to pests and moulds to severe damage. Sunflower grain is not an exception, particularly considering the common pests like birds, rats, mice, chicken and others. Delayed purchasing therefore was one of the reasons which hindered increased sunflower production.

Presently, however, most of the sunflower produced within the country is urgently needed to feed the Multipurpose Oil Processing Company (MOPROCO) factory in Morogoro. In order to keep the factory working at full capacity sunflower is at present being collected promptly. Since farmers are not forced to store their produce, thus losing a great part of it through pest damage, their morale has been boosted.

2.1.3 Distribution of sunflower in Tanzania

A greater part of Tanzania, approximately 70% of total area (Philip, 1980) receives an average annual rainfall of less than 1000 mm and this comes within a single season lasting between 4 and 6 months. The rest of the year is fairly dry. The vegetation is either savanna, woodland savanna, grassland or to a lesser extent bushland and thicket (Fullard, 1965). All this land is dry with less fertile soils and hence, unsuitable for most of the common cash crops but otherwise good enough for sunflower.

Sunflower outpoints all related crops which are normally intercropped with it in drought resistance (Jensma 1973). With enough incentives to farmers sunflower can be grown on a large scale in drier parts throughout all regions in Tanzania. Regions which grow sunflower in an appreciable scale are given in Appendix 3 and Figure 1. In addition Lindi, Kagera, Rukwa and Tanga regions have developed interest in this crop.



5-YEAR MEANS OF REGIONAL SUNFLOWER PRODUCTION IN TANZANIA (1975/76 – 1979/80 March)

Fig. 1 : Map of Tanzania showing 5 year means of sunflower seed production (1975/76 to 1979/80, March)

2.1.4 Uses of sunflower

Sunflowers were first grown as ornamental and later North American Indians started using grains for food, oil, cosmetics, as a source of dye and for medicinal purposes (Putt and Saskston, 1957). In the nineteenth century, Russians initiated large scale sunflower farming particularly for the extraction of edible oil. Poole (1975) indicated that sunflower now ranks third as an oil crop in the world after soybeans and groundnuts. The grain contains about 40 - 45% oil (Poole, 1975). For human consumption, sunflower oil is as good as olive oil; however, it is better in keeping liquid at lower temperatures, 17 to 18^{o}C, while olive oil solidifies at 21^{o}C.

Sunflower oil consists mainly of glycerides, of oleic, linolic and palmitic acids and a little linolenic acid. The double refined grade has the highest qualities (Hurt, 1946).

After collecting edible oil the second pressing yields oil which is darker and this is normally used in industries for making fine paints, soaps and cosmetics (Hurt , 1946).

Sometimes also grains can be used as poultry feed particularly for layers. The high percentage of oil (40 to 45%) and protein (17 to 20%) makes the grains a well balanced diet for poultry and cage birds (Hurt, 1946).

Some countries grow sunflower as forage for cattle. It is either fed green or preserved as ensilage. In order to maximise soil coverage sunflower grown for forage is sown by broadcast and harvesting of crop is done while stems and leaves are still tender.

Other by-products include : stalk, heads and kernels. Stalks are used as firewood in rural areas but in industrialised countries they are used in glass industries and also for the production of cellulose (Hurt, 1946). The heads are processed into a feeding meal which is fed to poultry, cattle, and pigs. The kernels after oil extraction are processed into sunflower cakes also for animal feed.

Sunflower residues in the field can be utilized by burning and spreading the ash over the field or disk-ploughing them under the soil after harvest. In the tropics it is always found better to make compost out of the residues.

The hectarage under sunflower crop in Tanzania is rapidly increasing. It is likely therefore, that most of the crop will repeatedly be grown on the same fields from season to season hence facilitating the build up of diseases and other pests which is a common phenomenon to intensive cultivation of the same crops.

2.2 Major sunflower diseases

Sunflower rust caused by Puccinia helianthi Schw. was first described in 1822 from specimens collected from Southern United States (Putt and Sackstone 1957). They gave a geographical distribution of rust as United States of America, Canada, Mexico, Bermuda, Cuba, Dominican Republic, Guatemala, some 16 countries in Europe, China, Israel, Japan, Korea, U.S.S.R. (Asia); several countries in Africa, some States in Australia, Argentina, Peru, Chile and Uruguay.

Such an alarming spread forced agricultural researchers to look into the possibilities of combating this disease. Sunflower rust resistance was therefore investigated (Putt and Sackston, 1970 and 1971; Siddiqui, 1972; Hoes and Putt, 1966; Sackstone and Miah 1963; etc.). Hand in hand with breeding for resistance, chemical control was also tried (Szoko and Szabo, 1965; Bruni, 1971).

Verticilium wilt incited by Verticillium alboatrum was first recognized in Manitoba, Canada in 1948 (Sackston, McDonald and Martens, 1957). It is known to reduce yields by more than 50% (Zimmer et al 1973). Studies on the control of this disease have also been carried out (Zimmer et al 1973; Sackston et al 1977; Hoes and Putt, 1966; Putt, 1958).

Sclerotinia wilt and head rot caused by Sclerotinia sclerotiorum is a widely spread disease and causes heavy losses. Kolte and Mukhopadhyay (1973) reported that the disease caused an average

plant loss of 12% in India. Putt (1958) studied the difference in susceptibility of sunflower to Sclerotinia wilt which was further studied by Melihova (1967). Attempts to control the disease have been studied by Crisan, 1962.

In East Africa, Hansford (1937) appears to be the first to mention Alternaria tenuis Bolle f.a pathogenic fungus attacking sunflower in Uganda. Nattrass (1950, 1951, 1952, 1953) working in Kenya reported serious infections by Puccinia helianthi Schw. and Algugo tragopogonis (D.C.)S.F. Gray; and also losses caused by Sclerotinia minor Jagga and S. sclerotiorum (Lib.) de Bary respectively. In 1949 Nattrass noted the presence of a leaf spot caused by Ascochyta or Diplodia helianthi, Septoria helianthi and Alternaria zinniae. In 1970 Martens working in Kenya reported that leaf necrosis wilt (unknown cause) affected about 5% of two fields but warned that since this disease was spreading fast, it could turn out to be a serious disease. He further reported that rugose mosaic was observed in three fields. Verticillium wilt caused by Verticillium alboatrum Reinke and Berth was found in a number of sunflower fields in Kenya and its spread was very much alarming since it was a distructive disease in Russia and North America (Martens 1970). In Tanzania a sunflower disease known as 'Krommnek disease' was first identified by Wallace (1947). Other sunflower diseases occuring in Tanganyika (Tanzania Mainland) were listed by Riley (1960). Allen (1974) reported all diseases attacking sunflower in Tanga Region.

Previously in East Africa, stem rots had been associated with Sclerotinia sclerotiorum and regarded as a minor disease (Martens et al 1960). However by 1977 the stem and head rots had gained so much

ground that Ongoma and Njoroge (Pers. comm. 1977) then working on sunflower project at Njoro in Kenya, classified this disease among the three most dangerous diseases, after rust and Verticillium wilt. As mentioned by Allen (1974), Wienk (1968) reported Erwinia aroideae as a causative bacterium to the pith soft rot of sunflower in Tanga Region.

Banihashemi (1975) observed a pathogen causing black stem rot, a serious sunflower disease in Iran which was later identified as Phytophthora drechsleri Tucker. Symptoms described by Banihashemi (1975) somehow resemble those of a disease observed in Morogoro (Keswani, 1976).

Several workers have isolated Fusarium sp. from diseased sunflower plants and sometimes from grains either still in the field or after harvest. Sackston (1957), isolated Fusarium sp. from sunflower but this was from mottled leaves. In India, Gupta et al (1980) reported that Fusarium solani had between 1976 and 1978 caused a serious colar rot to sunflower seedlings at Agra. Fusarium sp. was also isolated from lower stems and roots of sunflower by Orellana (1971), but this was associated with vascular wilt. Ganacharya et al (1978)found that sunflower seedlings of up to six weeks of age were highly susceptible to Fusarium oxysporum but plants over ten weeks were not attacked. Between 1978 and 1980 Vijayalakshmi and Rao (1986) also reportedly isolated Fusarium spp. from developing sunflower grains. A new disease on sunflower stems has just been reported by Zazzerini and Tosi (1987); caused by Fusarium tabacinum but its symptoms are external necrotic streaks and a pale pinkish red discoloration of the pith.

Fusarium has also been found on stored sunflower grains by other workers like Singh and Pradat (1986) who have just reported that Fusarium moniliforme (Gibberella fujikuroi) decreases cholesterol contents in stored sunflower grains. However, so far no Fusarium spp. have ever been associated with a sunflower stem rot similar to the one found in Morogoro.

3. MATERIALS AND METHODS

3.1 Collection of diseased specimens

Since the disease was first noticed at the then Faculty Farm, all initial specimens were collected from fields in Morogoro. Later during the survey, some additional specimens were collected from Mtakenini, Kitange, Kibedya, Chakwale and Kiloti, all villages within Morogoro Region.

Diseased stems, some with early and some with advanced symptoms were cut above ground level using a sharp pocket knife. Smaller fresh pieces of between 30 and 60 cm with necrotic areas of different sizes were placed in paper bags and sent to the laboratory.

3.2 Sunflower disease survey

Survey for the incidence of stem rot of sunflower was done in regions selected on the basis of their importance in sunflower production. These regions included: Tabora, Shinyanga, Dodoma, Singida, Morogoro, Iringa, Mbeya, Kilimanjaro and Arusha. Most fields covered were along main and feeder roads, particularly those belonging to schools, prisons, Ujamaa villages and occassionally private individuals; all were carefully surveyed. Members of the team normally consisted of the author, representative of GAPEX and sometimes an official from the local office of the Ministry of Agriculture. On arrival at the farm the team walked across the field diagonally counting diseased plants out of every group of twenty. Where the field was very large we counted diseased plants as above, walked fourty meters

then counted again for a total of 10 to 12 counts. The disease incidence was then calculated (Streets, 1972).

At most district headquarters Meteorological offices were visited to collect data on rainfall, temperature, relative humidity; and relevant literature on the 'Monthly Farming Weather' were studied to extract recommended sunflower planting dates.

3.3 Isolation of microorganism(s)

Diseased samples were washed with tap water and small sections from the edge of lesions cut, sterilized in 0.1% mercuric chloride, washed several times (generally three times) in sterile distilled water and then quickly transferred to 9 cm petri dishes containing previously prepared either Potato Dextrose Agar (PDA) or Malt Extract Agar (MEA) (Ainsworth, 1968). Some stem pieces were incubated in luch boxes with moistened filter papers for direct examination of mycelial growth.

3.4 Identification of Microorganisms

Slides were prepared by using cotton blue-lacto-phenol from seven to fifteen days' old cultures and also from the diseased pieces incubated in lunch boxes for up to ten days. From both petri plates and incubated samples a total of three slides were made each time and examined under a compound microscope at different magnifications. Some cultures were sent in MacCartney bottles to the Commonwealth Mycological Institute (CMI), Kew UK to confirm the identification.

Cultures which did not sporulate readily were transferred to another chamber with utraviolet 'Black Light' on the fourth day. They remained under a Black Light florescent tube for up to two weeks (Ainsworth, 1968).

3.5 Slide culture technique

In order to facilitate easy identification the slide culture technique according to Ridell (1950) was also employed. About 10 ml of PDA or MEA were melted and poured into a sterile 9 cm petri dish to form a layer 2 mm deep. After the medium solidified 1 cm squares were ruled out over the whole plate using a sterile scapel and a flamed glass rod. Using sterile forceps flamed microscopic slides were placed under cover of a petri dish lid. A flamed cover slip was placed upon this slide to overlap at one end. An agar square was then lifted out of the petri dish carefully and was transferred rapidly to centre of the cooled slide. Small bits of mycelium collected from clean previously prepared culture were placed at the centre of each edge of the agar blocks. With sterile forceps the cover slips were then placed centrally upon the upper surface of the agar squares and the slides were transferred to moist chambers and left to grow undisturbed.

When the cultures were fully grown up to about 1 cm from the cover slip, each cover slip was lifted vertically from the agar block carefully and placed aside, culture uppermost. The

agar blocks were also lifted and discarded leaving both slide and cover slip with areas of fungal growth joined by scanty lines of mycelium. A drop of 95% ethyl alcohol was applied to the centre of each of the rings of growth and was allowed to flow outwards to wet the fungus. Just before it evaporated a drop of lactophenol blue was applied to the centre as for the alcohol. A clean cover slip was gently lowered upon the slide culture to spread the dye outwards over the damp mycelium. Similarly a clean slide was applied to the cover slip preparation and was then quickly inverted. These were left overnight and examined under the compound microscope the following day.

During identification, observations were made regarding rate of growth, colony colour on both sides of petri dishes, texture of surface, shape and size of conidia, septation and presence or absence of clamydospores. Measurements were made using a compound microscope fitted with an ocular micrometer.

3.6 Pathogenicity tests

Sunflower seedlings were raised in polythene bags at TPRI for one to two months for inoculations. The soils used were collected from a nearby field and could be described as volcanic ash sandy loam, dark greyish brown in colour. According to FAO legend they could be called 'mollic andosol'. Such soils have low bulk density and T.P.R.I. soils have 0.9 g/cc which makes them easily erodable especially by winds. However, they had a good natural fertility and therefore no additional

fertilizers were added. There was deficiency of manganese whose concentration in a normal soil is normally more than 100 ppm but the concentration of the rest of nutrients were sufficient for normal plant growth (Table 1.).

For field tests sunflower plants were raised in a field within the T.P.R.I. compound. Soils were the same as those used in glasshouse trials.

Table 1 : Composition of soils used in a T.P.R.I. glasshouse for raising test sunflower plants

Element	Amount	Method
K	740 ppm	
Ca	660 ppm	Extraction with dilute HCl + H_2SO_4
Mg	129 ppm	
Zn	4.4 ppm	
Cu	5.3	Extraction with aq. 1% Na EDTA
Mn	5.0	

3.6.1 Glasshouse inoculations

Cultures to be used for inoculations were grown in 9 cm petri dishes. Each petri dish culture was divided into four equal blocks. Each culture block was removed and fastened onto the uninjured stem, according to Banihashemi (1975) and Ainsworth (1978). The inoculum was then covered with aluminium foil (alaf Kitchen foil) and to prevent drying before infection, a layer of cotton wood was added on to the foil; this was watered every morning. This same procedure was followed for the control plants using agar without inoculum. In each case plants were inoculated at the age of between 30 and 40 days. Inoculations were carried out between September, 1981 and February, 1982. Mean monthly temperatures at 0900 hours and 1300 hours for the relevant months are given in Table 2. Some of the infected plants were used for reisolation to check if the symptoms were caused by the same fungus. The cultures obtained were examined under the microscope.

Table 2 : Mean monthly temperatures at 0900 and 1300 hours in the T.P.R.I. glasshouse between September, 1981 and February, 1982

Month	0900 hours °C	1300 hours °C
September	23.4	33.0
October	28.0	32.0
November	26.8	35.0
December	28.0	32.0
January	28.0	34.5
February	28.6	34.5

3.6.2 Field inoculations

The above procedure was followed except that all plants were manured with Farm Yarm Manure (FYM) and fertilised with Sulphate of ammonia (AS) at the rate of 15 g/planting hole. The older plants between the ages of 50 and 70 days were also inoculated to study the effect of physiological maturity of the plants on infection by the pathogens. In all these trials a variety 'Record' was used and no plant was injured prior to inoculations. Inocula were fastened anywhere between 30 and 50 cm above the soil level.

3.6.3 Glasshouse inoculations with *F moniliforme* and *F. graminearum*

During pathogen isolation from naturally infected plants both *F. moniliforme* and *F. graminearum* grew together in culture differing in texture and later in colour. It was therefore necessary to carry out inoculations using a mixture of both species to see if there was differences in the mode of infection between the mixture and the individual species inoculated singly.

3.6.4 Soil inoculations with *F. moniliforme* and *F. graminearum*

Soil inoculations were carried out both in the glass house and in the field to check the possibility of the pathogen infecting sunflower from the soil. Two weeks old cultures collected from 9 cm petri dishes were burried around each plant and watered every day. Ten plants were treated with each fungal species and ten plants were only watered as controls. This procedure was repeated four times in the glasshouse and four times in the field. Observations for any symptoms were made every day till plant attained maturity.

3.7 Growth of the pathogens

3.7.1 Growth of pathogen(s) at different temperatures

Six plates of each species of the fungus were placed in incubators (type "Incubaril Neuherger") regulated to 40, 35, 30, 25, 20 and 18°C then to a refrigerator (type Frigidaire) in chambers kept at 10 and 8°C. Linear growth was measured every day till the plates were covered by mycelial growth or at the end of the third week. Three weeks were considered the longest waiting period after which cultures were too old and could not grow any more. This procedure was repeated four times.

3.7.2 Rate of growth on different media

All four readily available media were tried to find the most suitable for optimum growth of both Fusaria species under study. These included Potato Dextrose Agar (PDA), Malt Extract Agar (MEA), Czapek Dox Agar (DOX) and Malt Extract mixed with Agar No. 3 in the laboratory (MEA - 3).

The media were boiled, sterilized in a Baird and Tatlock (London) Ltd. outoclave poured into 9 cm petri dishes and left to cool for twenty four hours. Using a sterilized needle, pieces of mycelium picked from mature cultures were inoculated to the centre of each plate and left to grow at 19 to 20^{o}C which was the room temperature in the TPRI culture room. Linear measurements of all 18 plates made from each medium were taken every morning until a petri dish was fully covered by mycelial growth. The culture room was lighted with a 40W day light fluorescent tube continuously.

3.8 Varietal screening for resistance

3.8.1 Varietal screening in glasshouse

Seven varieties were sown in polythene bags as described under previous glasshouse trials. Inoculations were carried out as described under section 3.6 (Pathogenicity tests). Tested varieties are given in Table 3.

3.8.2 Varietal field screening

Two season trials were conducted at the Tropical Pesticides Research Institute (TPRI) Farm in 1979 and 1980.

A complete randomised block design was used with four replications.

Twenty plants were spaced at 30 cm from each other and 90 cm between rows. Each plant had three rows. In 1979 the field was planted on 15th April and in 1980 on 20th April. Varieties are given in Table 3.

The plots were weeded twice gapped two weeks after sowing and no fertilizers were applied. Assessment for stem rot was conducted every 14 days.

3.8.3 Varietal field screening at Morogoro Faculty Farm

Two trials, screening varieties for resistance to stem rot, were conducted at the Faculty Farm in 1980 and 1981. Design and spacing were the same as in section 3.8.2. Planting was carried out on 21st April, 1980 and on 18th March, 1981 using varieties shown in Table 3.

Disease assessment was carried out once in 1980 and twice in 1981 by counting all and diseased plants in each plot. Incidence of stem rot was calculated as follows:-

$$\% \text{ stem rot} = \frac{\text{No. of diseased plants/plot}}{\text{No. of all plants/plot}} \times 100$$

This was then subjected to analysis of variance.

Table 3 : Sunflower varieties and their country of origin screened against stem rot both at TPRI and Sokoine University of Agriculture Farm., 1979 - 1981

Varieties	Country of origin
Record	Kenya
Jupiter	U.K.
Gor 104	South Africa
Dwarf comet	Kenya
NS - P - 20	Yugoslavia
Giant 549	USA
1S 894	USA
1S 893	USA
1S 891	USA
1S 849	USA
1S 56F5	USA
6F5	USA
NS-P-62	Yugoslavia
NS-P-61	Yugoslavia

3.9 Host range

In Tanzania sunflower is normally intercropped with maize, beans or peas (Anon, 1979). It was also observed in many parts of Tanzania that sunflower is further planted with sorghum, cowpeas and in a few places with groundnuts. During the nation-wide survey none of these interplanted crops showed similar disease symptoms as those on sunflower. Also many of these crops were left scattered in the field after harvest till the following season hence increased chances of carrying over the inoculum to the following crop. It was necessary therefore, to test susceptibility of some of the crops intercropped with sunflower so that they could be avoided or encouraged depending on their reaction to the pathogens. All work was conducted in a glasshouse.

Potted plants which included maize (Zea mays) Var. Katumani, sorghum (Sorghum bicolor) Var. Serena, beans (Phaseolus vulgaris) Var. Canadian wonder, groundnuts (Arachis hypogaea), simsim (Sesamum indicum), cow peas (Vigna unguiculata) , green gram (Phaseolus aureus)and garden peas (Pisum sativum) were inoculated with one quarter of a 9 cm culture, covered with aluminium foil (Alaf Kitchen Foil), and then proceeded as described under pathogenicity tests. Number of days from inoculation to appearance of first symptoms was assessed and then the appearance of the necrotic areas was monitored throughout the entire growing period to see if necrosis was expanding changing color or remaining unchanged.

3.10 Chemical control

As one of the possible control measure, fungicide reportedly effective on some Fusarium spp. were screened to test if they could be used in controlling the sunflower stem rot. These included benomyl (Benlate 50% W.P.); Methyl 1-(Butylcarbamoyl) - 2 Benzimidazole carbonate (dupont de Nemours & Co. Inc.), fenapanil (Sisthane 24% W.P.) ∝ - n - butyl - ∝ phenyl-H-imidazole-1-propanenitrile (Rohm and Hass) and metalaxyl (Ridomil 25% W.P.); Methyl D, L-N (2, 6 - Dimethyl-phenyl) - N - (2-Methoxy acetyl) - alaminate (Ciba Geigy).

Two methods of testing were employed

i) Fungitoxicity Test - 1 ml of a given percentage was pipetted into 25 ml of previously prepared malt extract agar, mixed thoroughly poured into petri dishes and then left to cool over night.

ii) Difusibility Test - Malt extract agar was prepared as described earlier poured into 9 cm petri dishes and left to cool. Four filter dics papers previously sterilized and dipped into a relevant fungicide solution were placed at equal distances from the centre of the petri dish. All chemicals were tested at 0.1, 0.5, 1 and 1.5 a.i..

Linear growth was measured daily. For each concentration three petri dishes were prepared and three for the control with a total of 15 plates. Growth curves are shown in Figs. 22, 23, 24 and 25.

3.10.1 Glasshouse fungicide screening

Plants of sunflower variety Record were raised in polythene bags as previously described using unfertilized TPRI soils. Spraying was effected using 0.5, 1.0 and 1.5% a.i. of only Benlate. The plants were between 30 and 48 days old but each test batch of the same age. Each concentration was sprayed to five plants 24 hours before or after inoculation using Devilbis hand atomizers. Disease assessment which continued till head formation included days to first appearance of symptoms and later the total number of plants killed if any.

3.10.2 Field fungicide screening

Seedlings were raised in TPRI field for two months. The variety Record was selected because of its excellent viability. Seedlings were inoculated as described under pathogenicity testing. Fungicide suspensions were prepared at 1% concentration found effective in the laboratory tests and then applied to the inoculated areas by using the same hand atomizer. Ten plants

were sprayed with each concentration up to run-off and the other ten sprayed with distilled water as controls.

Spraying was effected as a protectant, twenty four hours before inoculation or curatively. The curative sprays were carried out twenty four hours after inoculation and another batch was sprayed on the second, fifth and tenth day after inoculation.

Control - inoculated but not sprayed at all.

These trials were repeated five times using five plants for each batch. The number of infected, healed and dead plants were recorded.

4. RESULTS

4.1 Symptoms

In the field, symptoms started with very small spots dark brown in colour developing on stems usually somewhere between 10 to 30 cm above ground. As they increased in number, the older lesions enlarged longitudinally but more markedly radially. They became darker as they enlarged and coalesced. Under favourable condisions many spots developed rapidly progressing upwards. When these spots coalesced, dry black rots developed (Fig. 2a, 2b, 3). Under humid conditions and particularly when plant population was very high, which was normally the case following sowing by the broadcast method affected plants became weaker and prone to lodging. Wind, weight of the forming head and termite attack were the common agents which accelerated such lodging long before maturity and many of the severely infected plants collapsed even before flowering.

4.2 Disease survey

Stem rot was found serious all over Morogoro Region. Severity of disease increased with increasing plant density particularly in villages like Kibedya, Kitange, Mtakenini, Majawanga, Kiloli, Mkalama and Chakwale where farmers planted their seeds by a traditional broadcasting method (Fig.4). It was observed that plant size and height differed greatly and due to high plant population weaker and shorter plants were the most susceptible to stem rot (Fig. 2c). Plants which had a wider spacing were less attacked and the disease was less severe to the infected few plants.

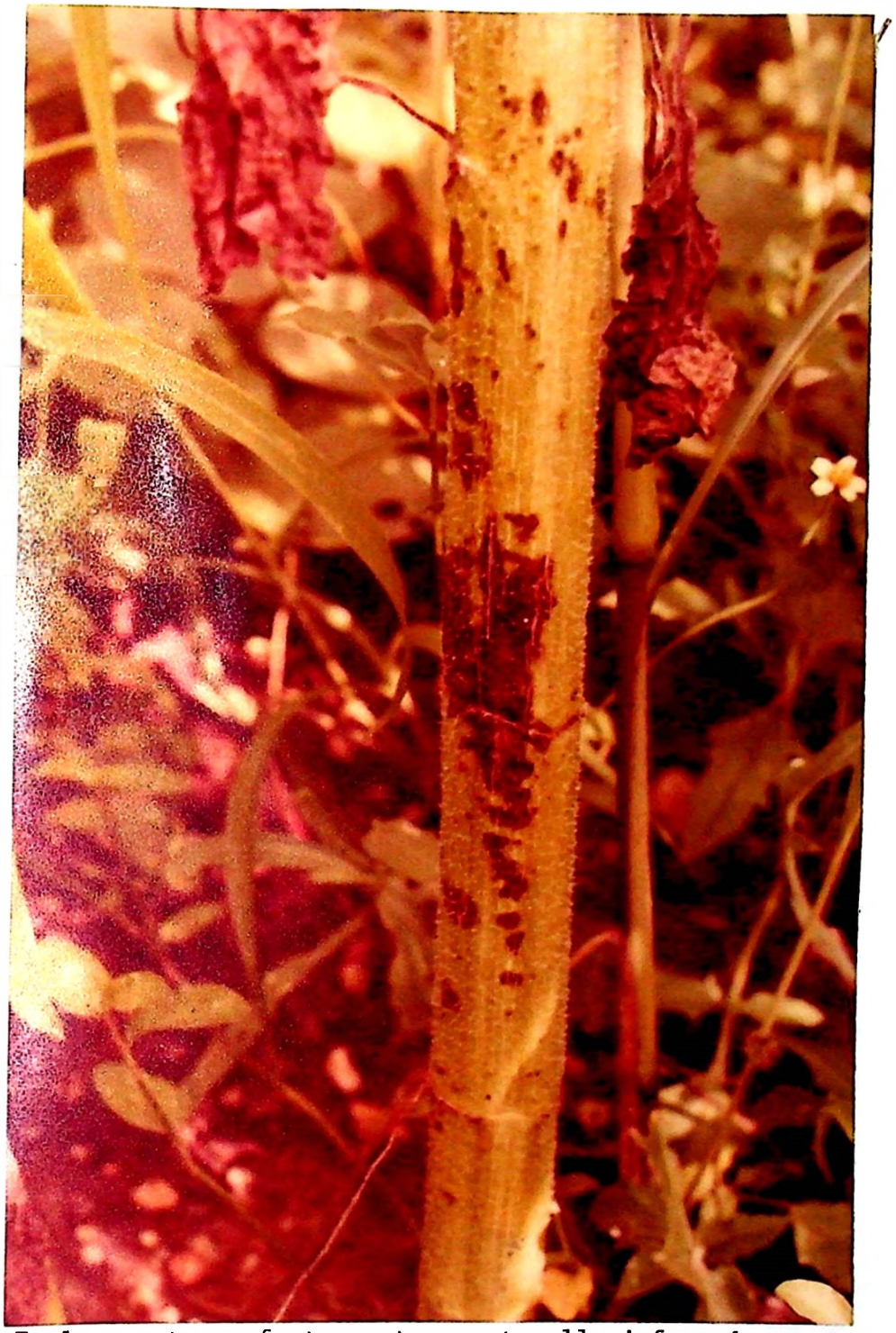


Fig. 2a : Early symptoms of stem rot on naturally infected sunflower

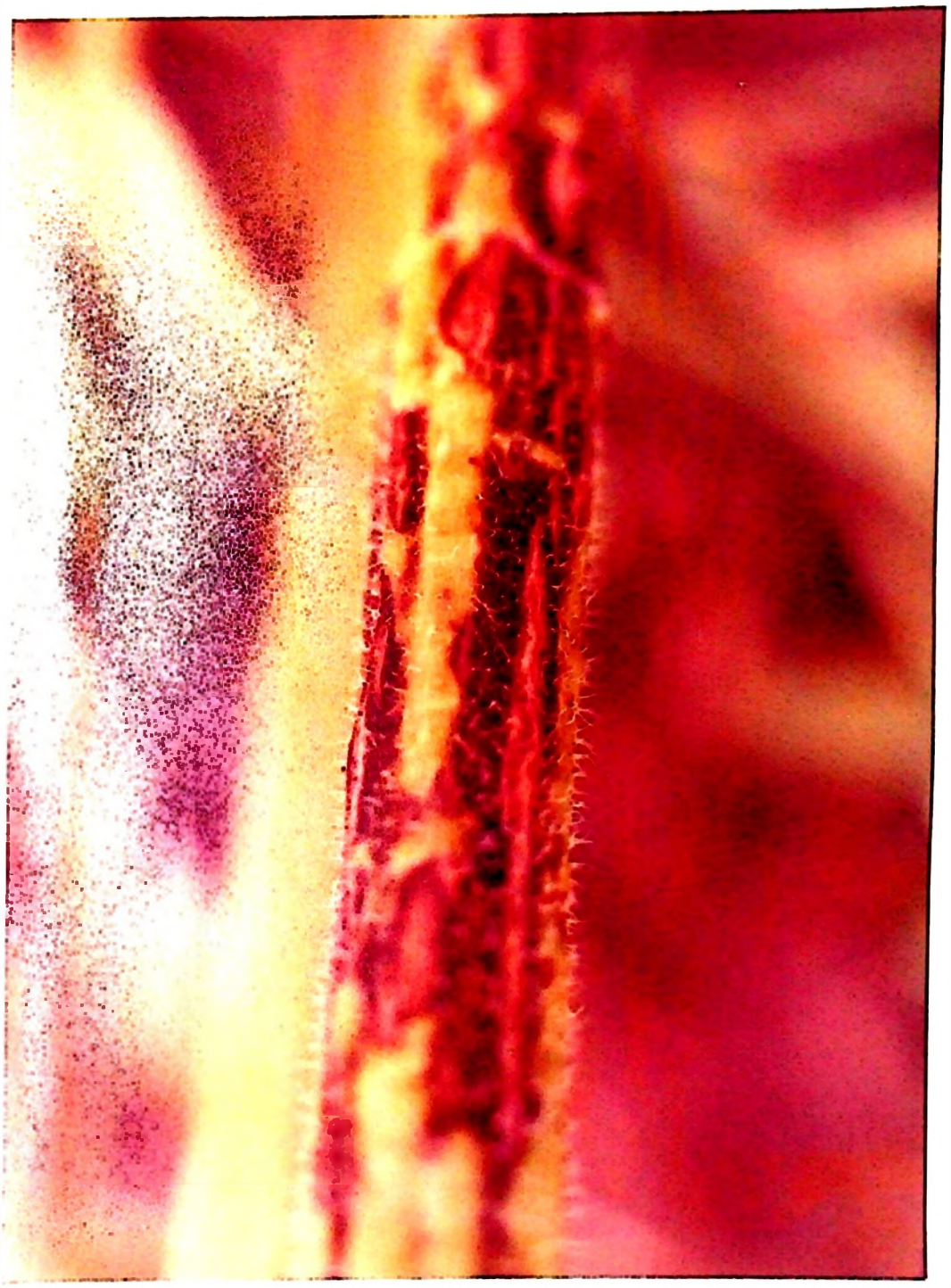


Fig. 2b: Advanced symptoms of stem rot on naturally infected sunflower

Fig. 2c : Diseased sunflower stems showing disease severity to weak stems while the strong stems survive till maturity

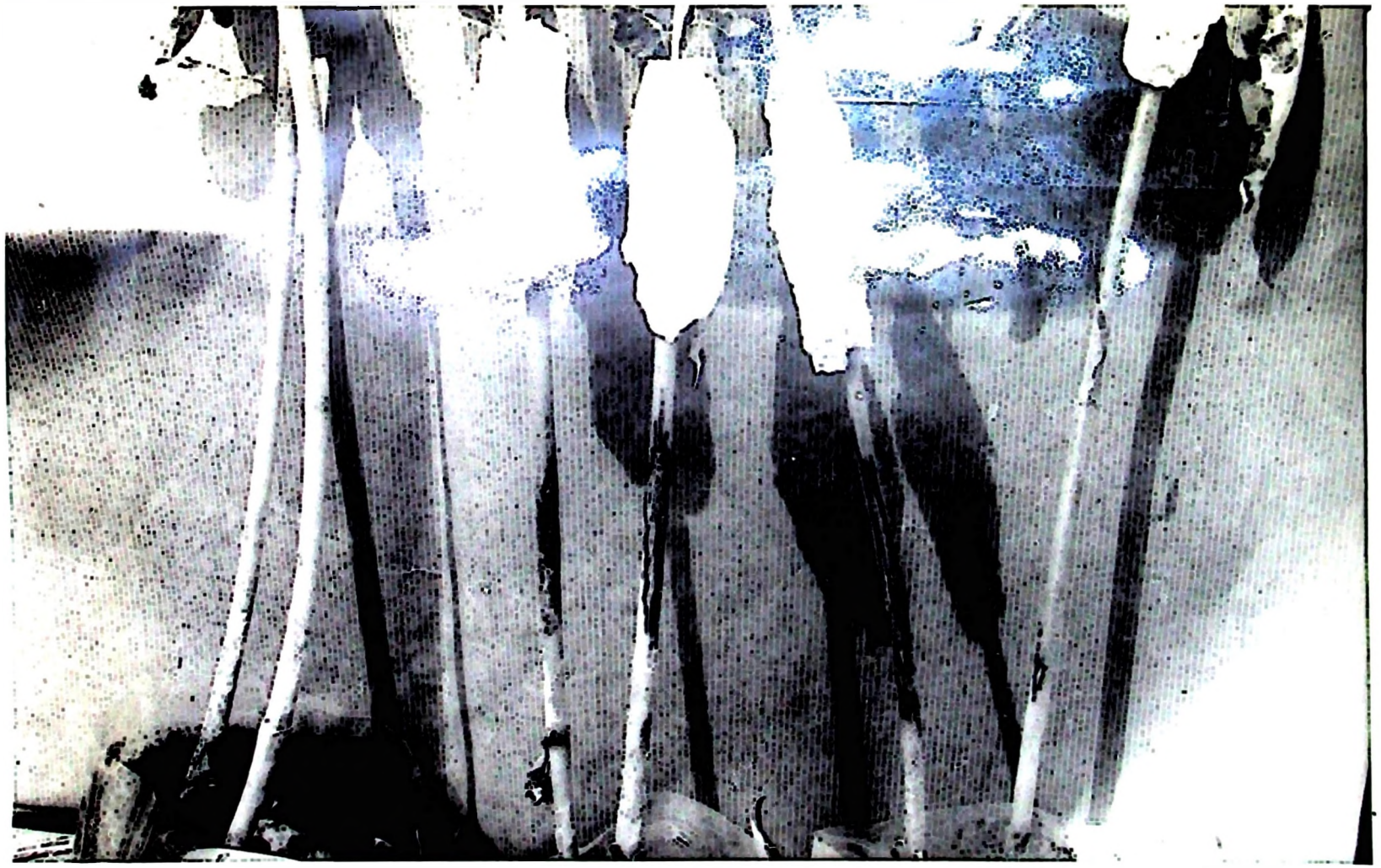


Fig. 3 : Symptoms of stem rot on artificially inoculated sunflowers with controls on the exteme left.

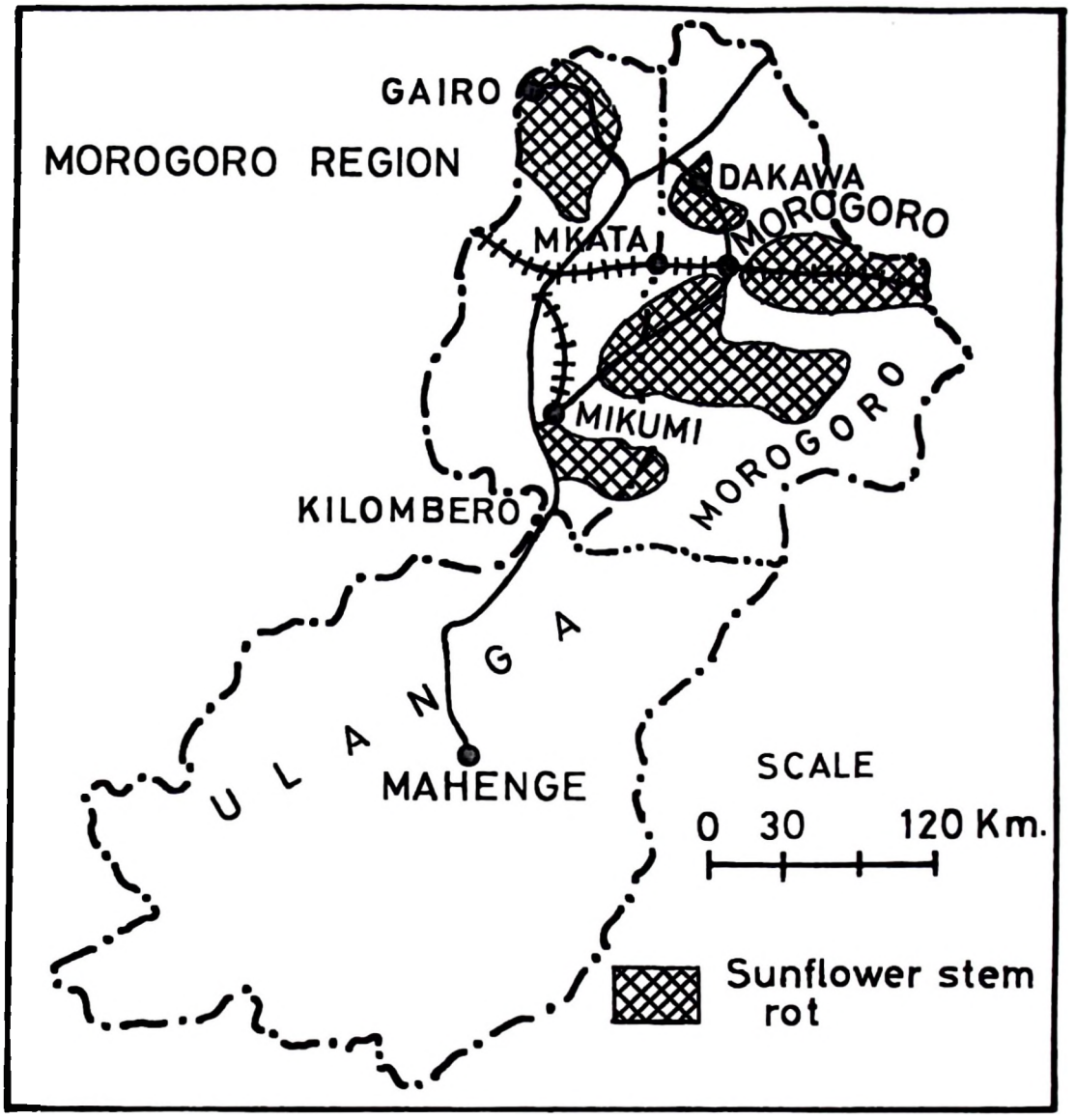


Fig. 4 : Map of Morogoro Region showing areas with infected sunflower

All other regions surveyed were found free of stem rot.

Monthly rainfall data which were collected from meteorological offices during the sruvey showed that during the sunflower growing months the amount of rainfall in Morogoro Region was not significantly different from that in Singida (Iramba and Singida Districts), Dodoma (Mpwapwa District), Shinyanga (Kahama District) and Mbeya (Fig. 6).

Months during which farmers planted and harvested their sunflower in different regions are given in Appendix 10. The Western sunflower zone (Tabora & Shinyanga) planted their sunflower in November, the central zone (Singida & Dodoma) planted in January and the rest of the surveyed regions (Arusha, Kilimanjaro, Morogoro, Iringa and Mbeya) planted between March and April (Appendix 10).

Air temperatures in the surveyed regions were ranging between 17 and 24^{o}C during the sunflower growing season, Morogoro inclusive(Appendix 11).

Relative humidity at 0900 hours was found to lie between 76 and 89% during the sunflower growing months (Appendix 12) and that taken at 1500 hours was between 44 and 70% (Appendix 11).

Soils where sunflower was being extensively grown were similar throughout Tanzania. The Regional Agricultural Development Officers described them as sandy loam, volcanic red loam, clay loam or volcanic ash sandy loam (Appendix 10).

Table 4 : Regions and their districts where sunflower stem rot survey was conducted between March and August, 1980

Region	Districts (villages)
Tabora	Tabora - (Tumbi, Puge and Sikonge), Nzega, Igunga.
Shinyanga	Kahama (Sabasabini, Iboja, Kinamaruka, Lowa, Kakebe, Igunda, Nyandekwa and Mpunze).
Dodoma	Mpwapwa, (Chamkoroma, Iduo)
Singida	Singida and Iramba (Mtipa, Ilongero), Mpamba, Iguguno and Kyengege)
Morogoro	Morogoro and Kilosa (Maharaka, Mtakenini, Kitange, Kibedya, Chakwale, Kiloli, Mkalama, Majawanga, Msimba, Dakawa)
Iringa	Iringa, Mufindi, Njombe (Ikengeza, Chamdindi, Nyang'oro, Kihorogota, Igumbiro, Image, Mafinga, Makambaku, Ikingula, Igwechanya)
Arusha	Arumeru, Babati, Monduli, Karatu
Kilimanjaro	Moshi and Hai (Kahe, TPC, Sanya Juu)

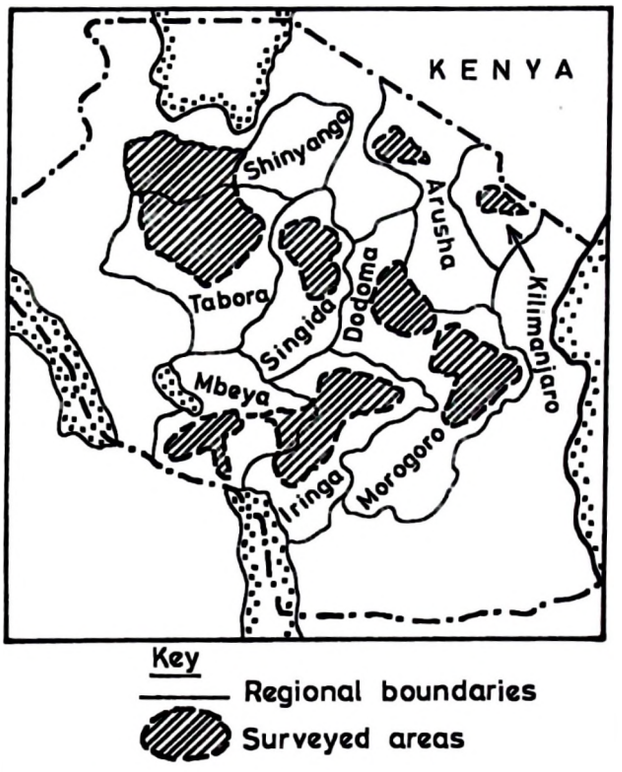


Fig. 5. Map of Tanzania showing surveyed regions for stem rot

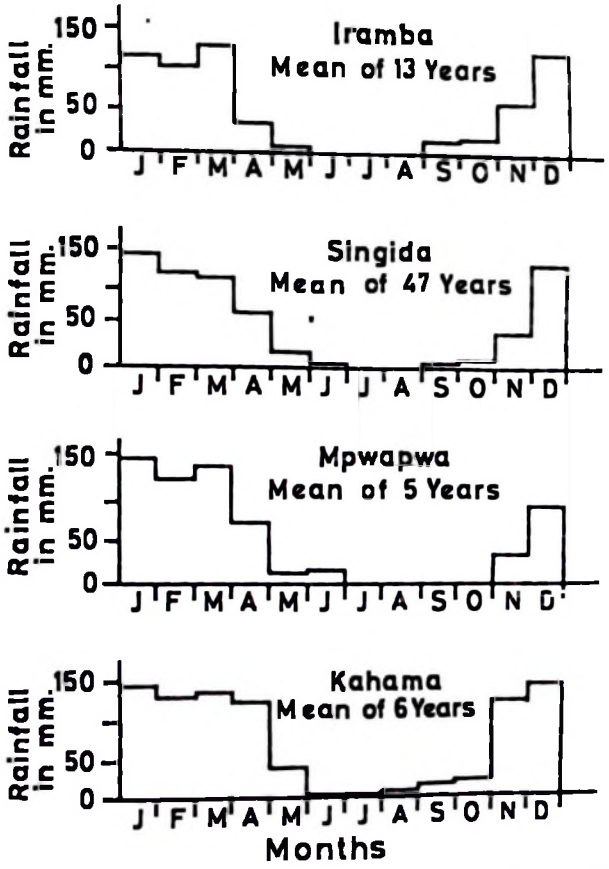


Fig. 6 : Mean monthly rainfall recorded at Iramba, Singida Mpwapwa and Kahama

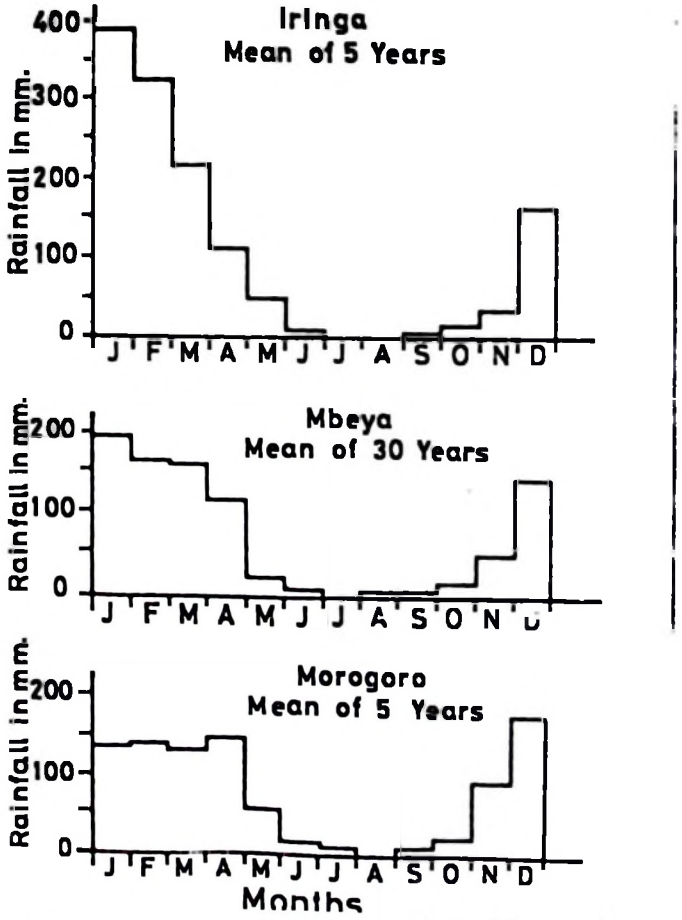


Fig. 7 : Mean monthly rainfall recorded at Iringa, Mbeya and Morogoro

4. 3 Isolation and identification of microorganisms

Cultures started to grow within the first two days beginning 24 hours following inoculation. The cultures were left to grow until each 9 cm petri dish was fully covered. This was achieved by the end of the third week.

All isolates either grown on PDA and MEA or isolated directly from diseased incubated plant tissues produced two types of culture which always appeared different both in colour and texture. It was therefore necessary to subculture each type of culture individually to make identification possible. These were named as isolate number one and number two.

4.3.1 Isolate number one

This isolate grew fairly well at 20°C. Its initial rate of growth was moderately rapid and mycelium was colourless not readily distinguishable from the colour of the media on which it was growing. As the fungus grew older the colour turned whitish, then light grey and later light violet the under side of the plates being dark violet than the upper. Texture of mycelium looked compact with some concentric rings (Fig. 8a). Aerial mycelium was pale turning pink-violet. Older cultures appeared powdery. Subcultured plates kept 60 cm under the laboratory day-light tube produced spores within four days while those kept in a special chamber lighted with a U.V. black light tube produced spores within two to three days. A week

Fig. 8a : Cultural characteristics of F. moniliforme, turned up-side down

Fig. 8b Microconidia of F. moniliforme

following subculturing, slides prepared from plates kept both under the day light and black light tubes had abundant conidia most of which (about 80%) were microconidia. All microconidia were non septate and measured between 5.4 to 13.5 μm by 2.1 to 2.7 μm. The few macroconidia which were present had between 4 and 6 septations and measured from 26.4 to 50.4 by 2.8 to 4 μm. All these measurements were taken from thirty spores of each group.

Using slides prepared from the Ridell slide cultures, it was observed that conidia were produced laterally on short hyphal branches crowded in sporodochia and many of them were in chains on phialides.

The description of spores, colour and texture of culture, measurements of both macroconidia, and microconidia and the manner in which the microconidia were born on phialides fully resembled those of Fusarium moniliforme Sheld. (Booth, 1971). Same cultures sent to the Commonwealth Mycological Institute (CMI) were later identified also as Fusarium moniliforme Sheld. under their Herb. IMI numbers 224083, 224107, 239480 and 239483.

4.3.2 Isolate number two

The second type grew very rapidly at 18°C - 25°C starting with cotton white mycelim which soon turned violet-red on both sides of the petri dishes. Then these cultures grew older the upper side turned deep red while the under side turned black (Fig. 9a). The mycelium was less compact with longer hyphae than those of isolate number one.

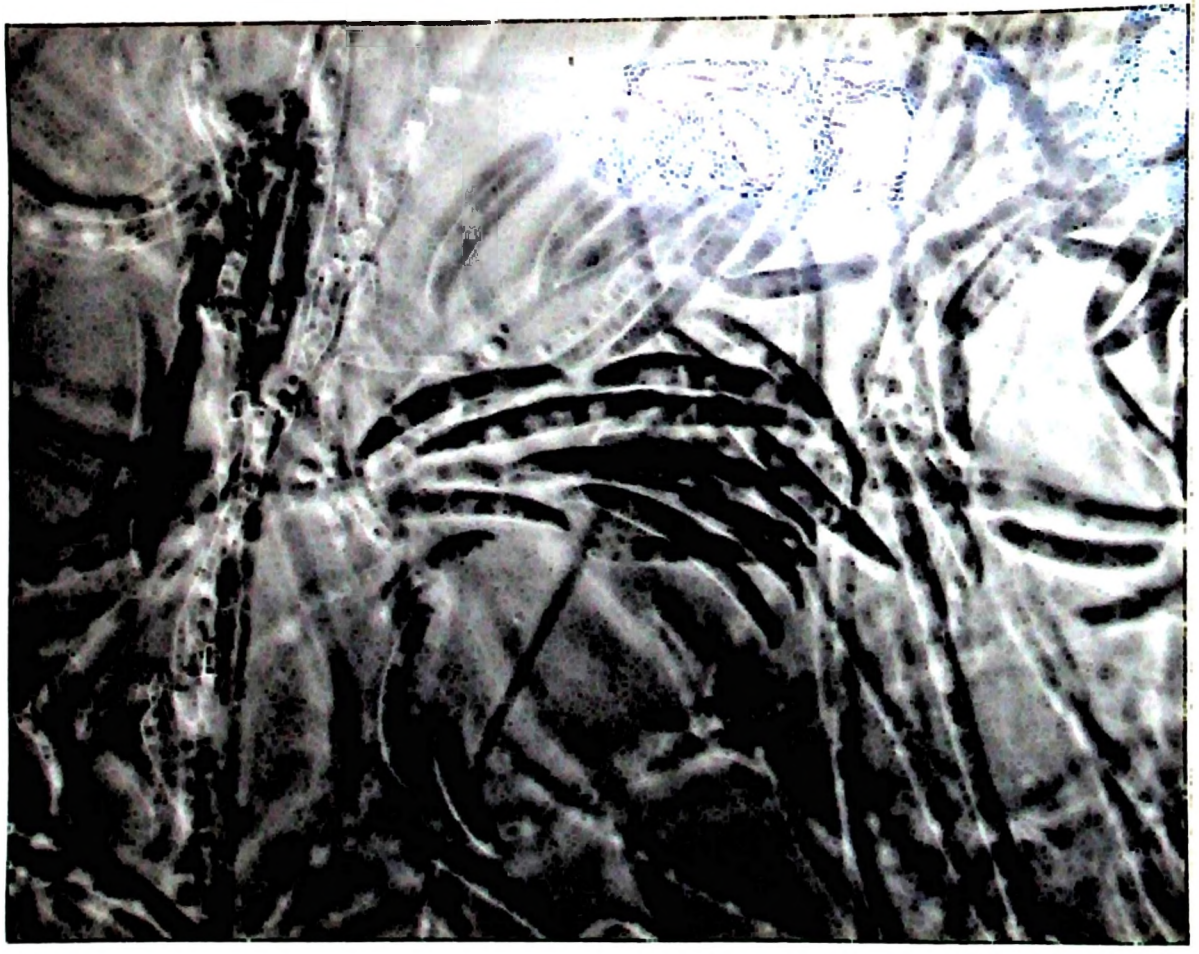


Fig. 9b : Macroconidia of F. graminearum

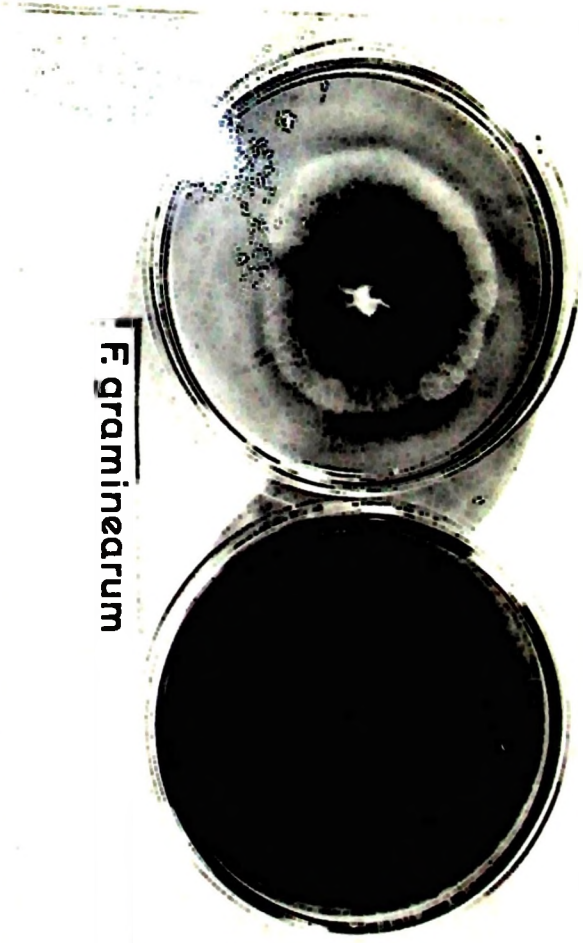


Fig. 9a Cultural characteristics of F. graminearum turned upside down

Spores were formed two days after inoculations when culture were kept under the U.V. black light while under the normal day light tubes spores were formed around the sixth day. Only macroconidia were found with 3 - 7 fine septa. They measured between 24.3 to 54 by 2.7 to 5.7 μm. Chlamydospores were always abundant.

This type of culture was identified as Fusarium graminearum Schwabe (Booth, 1971) and later confirmed by CMI under their Herb. IMI number 2306628 and IMI number 239481.

Pathogenicity of both cultures differed greatly. F. moniliforme and F. graminearum both produced symptoms on one month old sunflower plants two days after inoculation. While most of F. moniliforme inoculated plants continued to grow well over 15 days following inoculations all plants inoculated with F. graminearum collapsed within the first six to fifteen days after inoculation (Figs. 10 , 11).

4.4 Pathogenicity tests to sunflower plants

4.4.1 Glasshouse inoculations with F. moniliforme and F graminearum

F. moniliforme produced symptoms on one month old sunflower plants two days after inoculation. Symptoms on two months or older plants first appeared five clays after inoculation. Symptoms started as small specks on incoulated areas which enlarged and coalesced covering the entire stem circumference in two weeks (Fig. 10).

The same fungus was re-isolated from inoculated plants.

F. graminearum produced symptoms on the second or third day following inoculations to plants up to two months old. Lesions grew more rapidly than those formed by F. moniliforme, coalescing and forming a black dry rot which covered the entire stem circumference within four days. Plants started collapsing on the fifth day and within ten days more than 80% of all plants had died (Fig. 11). Re-isolation from such plants always yielded cultures of F. graminearum with the same characteristics.

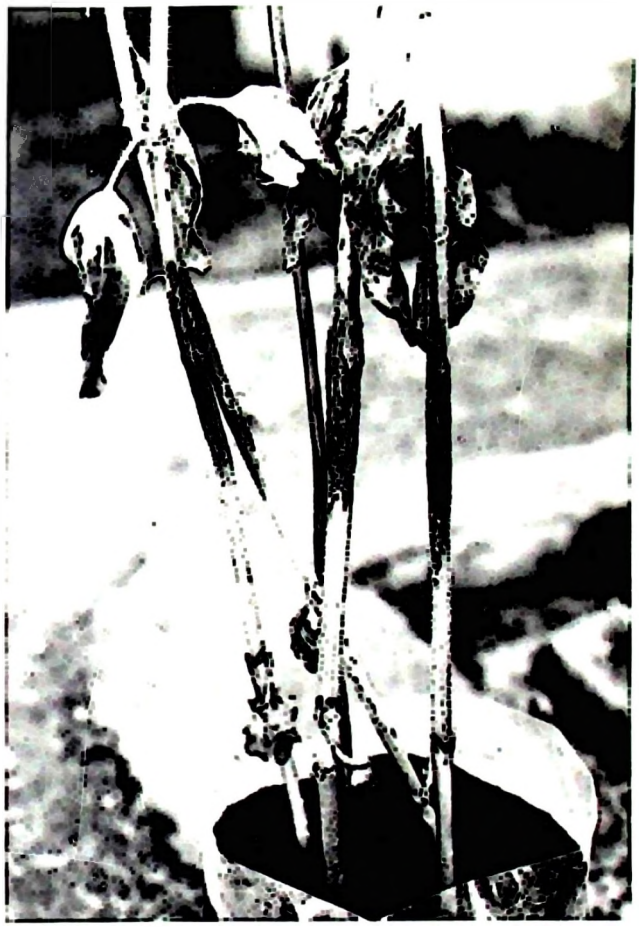


Fig. 10 : Reaction of sunflower inoculated with F. moniliforme

Inoculations made using a mixture of F. moniliforme and F. graminearum produced symptoms on the second day which consisted of specks which enlarged and coalesced to form the same dry black rot. Some of the inoculated plants died on the sixth day after inoculation; 40% of all inoculated plants died after one month. F. graminearum had a synergistic effect on plants inoculated with F. moniliforme while F. moniliforme had an anergistic effect on plants inoculated with F. graminearum.

4.4.2 Soil inoculations with F. moniliforme and F. graminearum in the glasshouse and in the field

All attempts to inoculate sunflower plants of different ages, mainly between one month and three months of age in the glasshouse and in the field through the soil failed to produce any symptoms.

4.4.3 Field inoculations with F. moniliforme and F. graminearum

First symptoms appeared on the fourth day and the lesions enlarged steadily. The roting was dry and in many cases it was accompanied with cracks and hypertrophy (Fig. 12 and 13). The un-inoculated control plants remained healthy till harvest.

Fig. 12 : Sunflower stem rot symptoms caused by F. moniliforme inoculated in the field

Fig. 13 : Sunflower stem rot symptoms caused by F graminearum inoculated in the field

4.5 Growth studies

4.5.1 Growth rate under different temperatures

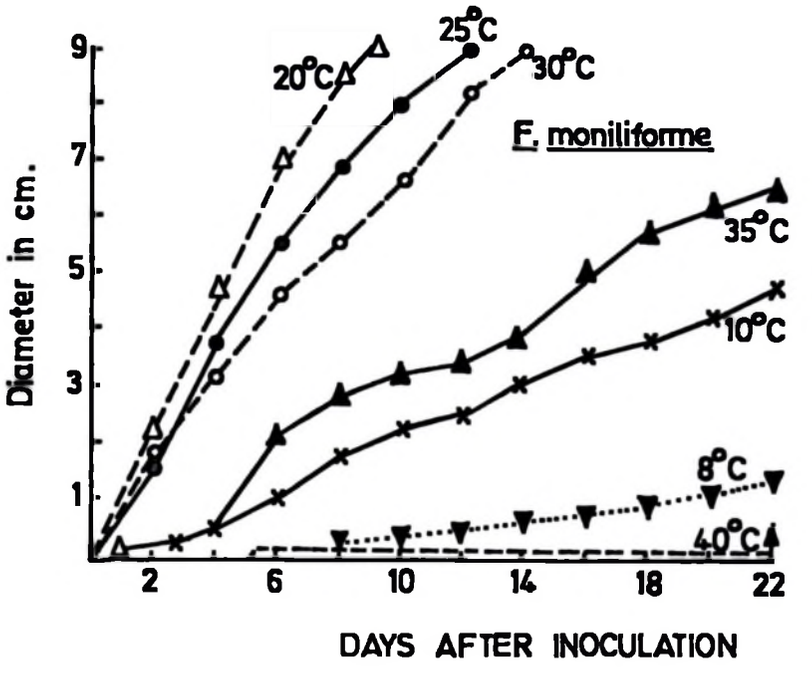
For both species growth rate was measured at 40°, 30°, 35°, 23°, 10° and 8°C. Six plates for each fungus species were measured for each temperature for a maximum period of three weeks and means were calculated.

F. moniliforme grew fastest at 20°C but generally temperatures ranging from 18° to 30°C were found suitable for normal growth. The fungus grew very poorly at 8°, 10° and 35°C. There was no growth at 40°C (Fig. 14).

F. graminearum grew well at temperature ranging between 18° to 25°C where it covered the petri dishes within 11 days. At 8° and 30°C growth of this fungus was very slow and it did not grow at all between 35° and 40°C.

4.5.2 Growth rate on different types of media

Linear growth was measured twenty four hours after inoculation and this was continued daily until 9 cm petri dishes were covered by the mycelim. Cultures were kept in a TPRI culture room lighted continuously with a 40W day light florescent tube with temperatures ranging between 19° and 20°C. F. moniliforme grew



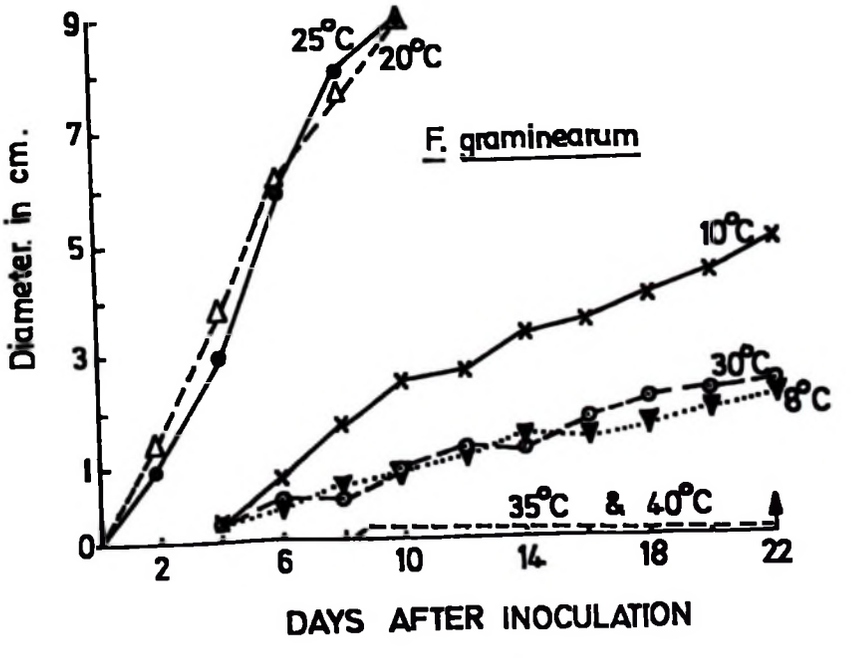


Fig. 14 : Growth of F. moniliforme and F. graminearum under different temperature

fastest in MEA - 3 covering the plate within 8 days. The slowest growth was recorded on PDA where the fungus took 12 days to cover the petri dishes. All media used in these experiments were rated as very good for F. moniliforme (Fig. 15).

F. graminearum grew faster on Czapek Dox Agar (Dox) covering the plate in 6 days while it grew slowest on Malt Extract Agar (MEA) covering the plate in 16 days. Generally with the exception of MEA, the rest of the media were found suitable for culturing F. graminearum (Fig. 15).

4.6 Varietal resistance

4.6.1 Glasshouse trials at TPRI

Plants of all varieties became infected on the fourth day by both fungal species. Varieties inoculated with F. graminearum died one after the other and were all dead within three weeks following inoculation (Fig. 11) while those inoculated with F. moniliforme, although all plants got infected, none was over killed (Fig. 10).

4.6.2 Field trials at TPRI 1979 - 1980

A continuous disease assessment was conducted at two weekly intervals till maturity. Not a single plant was infected till harvest during both season (1979-1980).

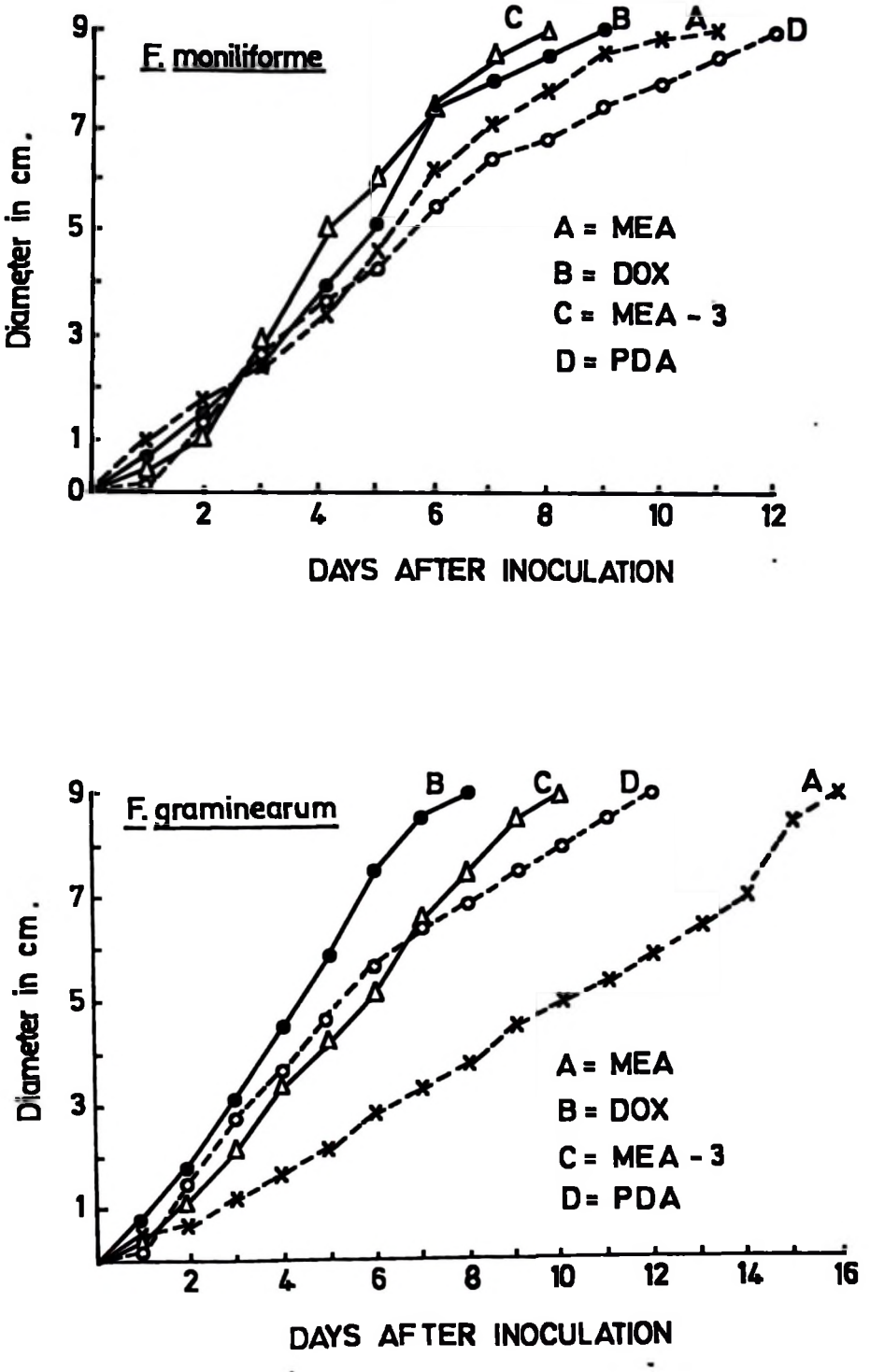


Fig. 15: Growth of F. moniliforme and F. graminearum on different media kept at between 20°C and 25°C

4.6.3 Field varietal screening at Morogoro Farm 1980 and 1981

Results of field screening of varieties at the Morogoro Farm for the seasons of 1980 and 1981 are given in Table 5. In 1980 varieties Record, Jupiter, Gor 104, Dwarf comet, Giant 549, IS 894, IS 893 and IS 849 were significantly different at 5% level compared to variety 6F5. Jupiter was the least diseased. Data collected in 1981 do not give any significant differences between varieties in percentages of infected plants.

Overall means for three assessments, however (one 1980 and two 1981) indicate that Record, Jupiter, Gor 104 and Dwarf comet had comparatively less diseased plants than other varieties (Table 5).

Table 5 : Incidence of stem rot at Morogoro Farm, 1980 - 1981

Variety	% diseased plants			Overall means
	20.6.1980	8.6.1981	26.6.1981	
1. Record	10.1	13.8	29.4	17.76
2. Jupiter	3.8	17.0	20.5	13.78
3. Gor 104	14.2	14.7	24.3	17.74
4. Dwarf comet	15.7	13.1	22.9	17.25
5. Giant 549	30.2	20.3	20.6	23.71
6. IS 894	39.8	14.3	29.9	28.01
7. IS 893	27.4	16.3	22.9	22.20
8. IS 849	35.1	16.9	30.6	27.50
9. 6/FS	74.1	16.6	26.2	38.95
10. 56 F5	26.9	17.4	n.a.	22.15
S.E.	8.94	4.67	5.20	
LSD (P = 5%)	18.3	NS	NS	

n.a. = not available

NS = not significant

4.7 Host range

Each of the tested crops reacted differently to the two *Fusarium* spp. and results are given in Table 6.

Maize (*Zea mays*) showed symptoms between the fifth and seventh day when inoculated with *F. moniliforme* and seven days when inoculated with *F. graminearum*. Symptoms were always similar, starting with small specks which later enlarged longitudinally and radially coalescing to form large clearly visible black lesions (Fig. 16). All lesions were dry and did not cause death of plants. Reisolation from these lesions always yielded *F. moniliforme* and *F. graminearum* as originally inoculated. Plants which were not inoculated (controls) remained without symptoms(Fig. 17).

Table 6 : Crops tested for susceptibility to *F. moniliforme* and *F. graminearum*

	Susceptibility		Remarks
	Fm	Fg	
Maize (*Zea mays*)	+	+	
Sorghum (*Sorghum bicolor*)	++	++	100% killed
Beans (*Phaseolus vulgaris*)	+	+	Weak plants
Groundnuts (*Arachis hypogaea*)	-	-	Very small specks
Simsim (*Sesamum indicum*)	-	-	Superficial specks
Cowpea (*Vigna unguiculata*)	-	-	Superficial specks
Greengram (*Phaseolus aureus*)	+	-	No effect
Garden peas (*Pisum sativum*)	-	-	One plant killed. Pods did not fill properly

Key : Fm.=*Fusarium moniliforme*

Fg.=*F. graminearum*

\+ = Susceptible

\- = not infected or very superficial

Fig. 16 : Reaction of maize plants to F. graminearum

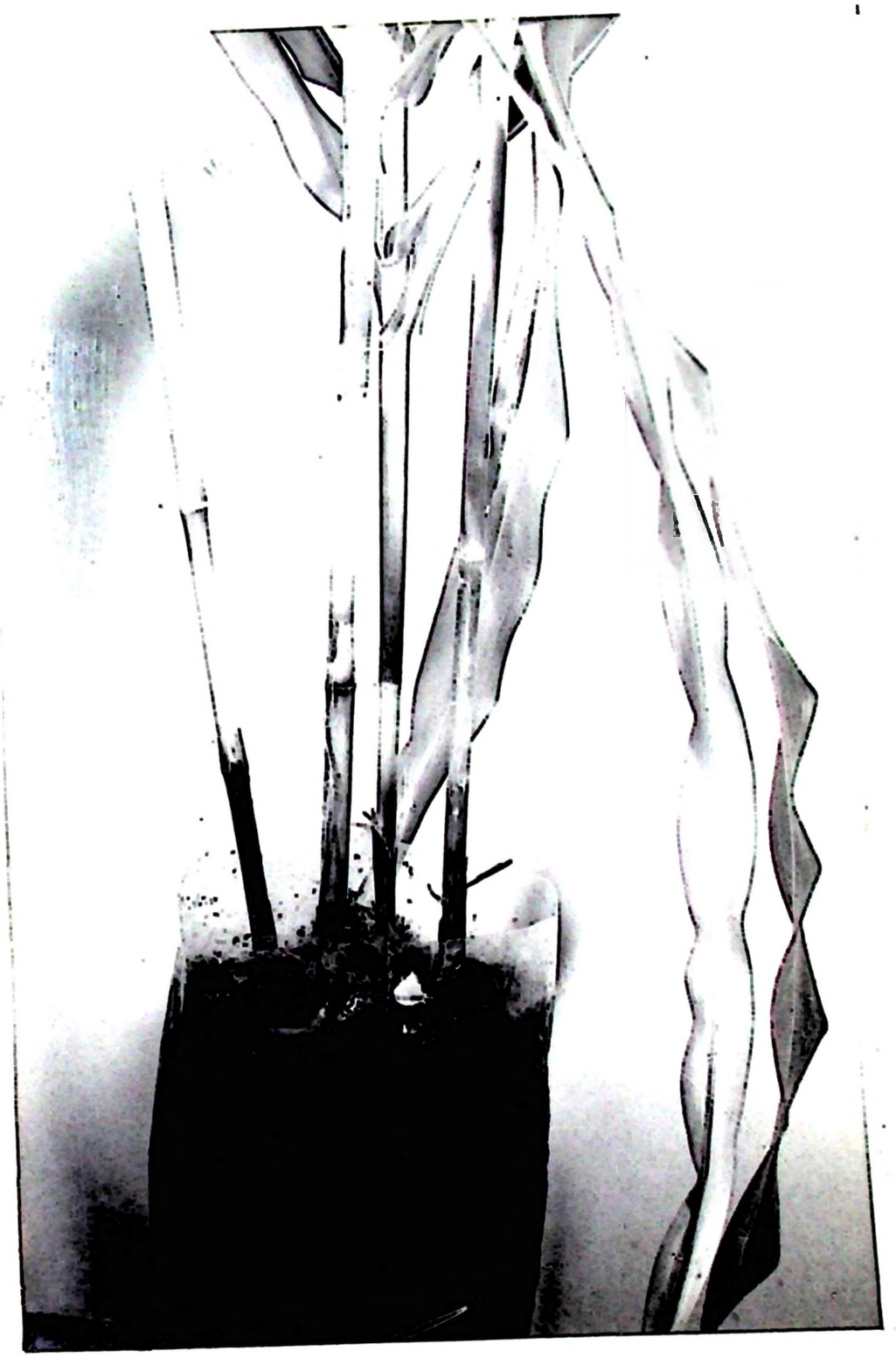


Fig. 17 : Uninoculated (control) plants

Symptoms on sorghum (Sorghum bicolor) were always visible on the seventh day after inoculation. F. moniliforme killed 50% of the plants 14, 20 and 25 days after inoculation and a week later the rest of the plants were all killed. F. graminearum also killed all inoculated plants within three weeks following inoculation. All uninoculated plants remained healthy.

Beans (Phaseolus vulgaris) showed symptoms on the fourth day. Lesions expanded to cause large necrotic areas which were light brown to black in colour. The plants were not killed by both fungus spp. Infection was the same for both F. moniliforme and F. graminearum (Fig. 18a and 18b). Control plants remained healthy. Isolations from infected bean plants produced the original F. moniliforme and F. graminearum.

Fig. 18a : Symptoms produced by F. moniliforme on bean plants

Fig. 18b : Symptoms produced by F. graminearum on bean plants

Only F. graminearum caused superficial lesions to inoculated groundnuts (Arachis hypogaea) three weeks after inoculation. These lesions were very minute, clearly indicating hypersensitivity reaction.

With simsim (Sesamum indicum) only F. graminearum infected and caused minute symptoms. All plants inoculated with F. graminearum showed a hypersensitive reaction while those inoculated with F. moniliforme showed no symptoms and were all healthy.

On cow peas (Vigna unguiculata) only F. graminearum caused symptoms around the eighth day. All plants, however, progressed well till maturity. On one occasion infection took place six days following inoculation but again no plant weakness was detected thereafter (Fig. 19 a). Isolates from both infected simsim and cow peas produced the same fungus, F. graminearum.

Control plants (not inoculated) remained without symptoms (Fig. 19 b).

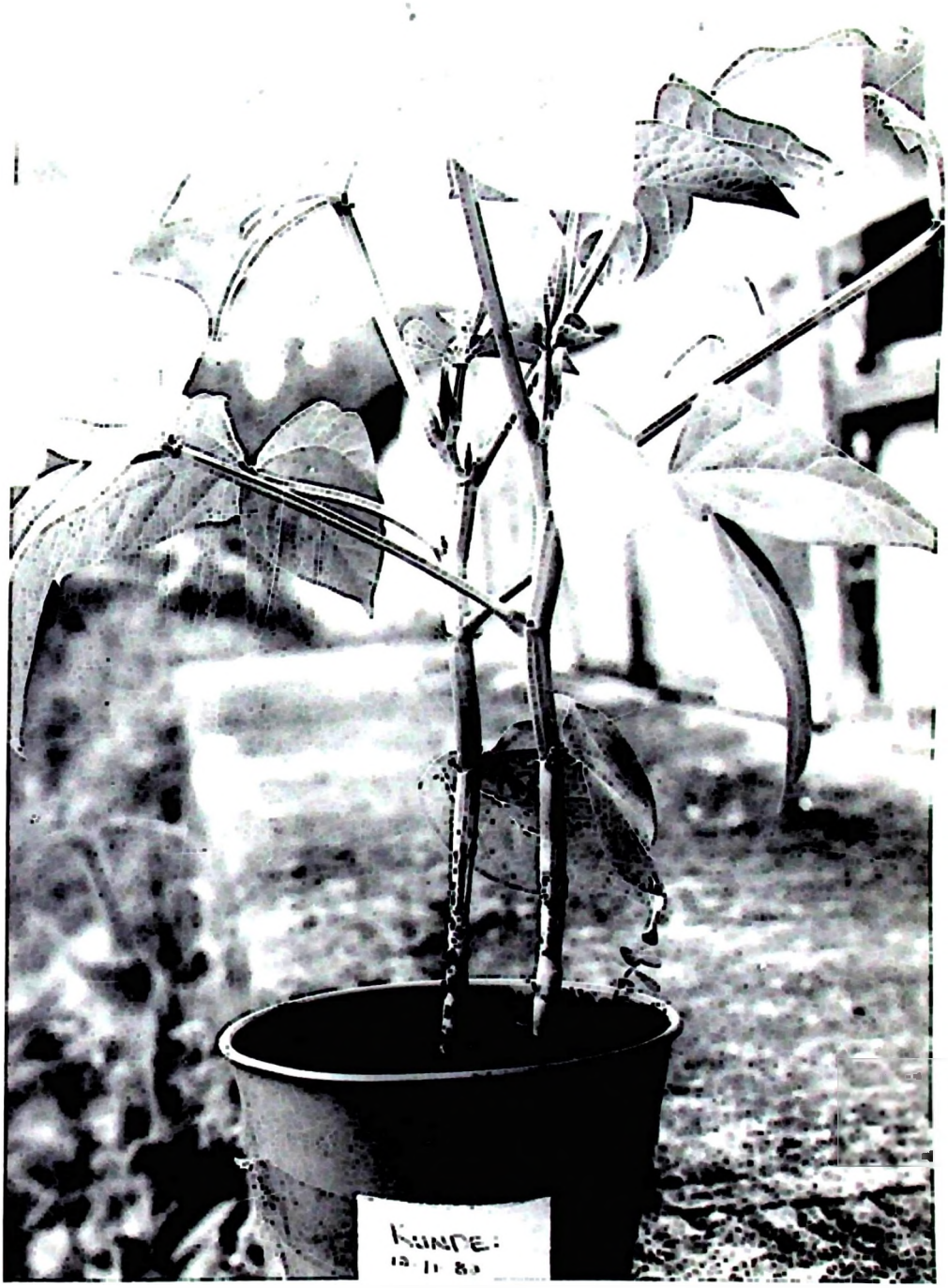


Fig. 19a : Minute specks caused by F. graminearum on cow pea plants

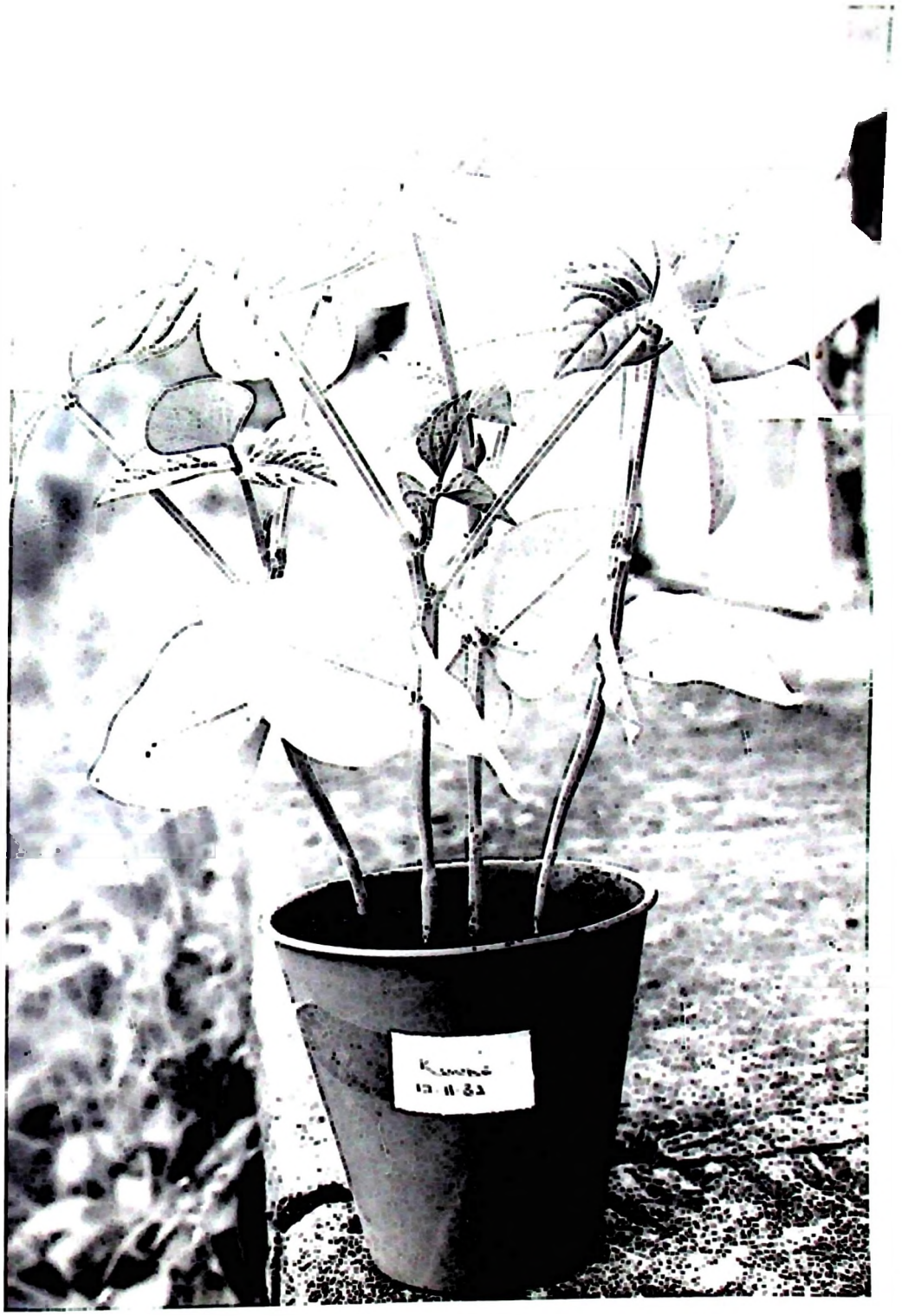


Fig. 19b : Uninoculated (control) cow pea plants

Infection with both F. graminearum and F. moniliforme to green-gram (Phaseolus aureua) normally took place a week after inoculation but symptoms were so minute that they were not visible on photographs (B/W). There was no general plant weakness noticed. All plants continued to grow well till harvest. When the fungus was isolated from the minute specks cultures produced typical F. moniliforme and F. graminearum.

Symptoms on garden peas (Pisum sativum) first appeared between 5 and 8 days with F. graminearum (Fig. 20). F. moniliforme failed to cause symptoms. Infected plants were weakened and on one occasion a plant was killed. The rest of the plants survived but their pods either did not fill or contained shrivelled poor seeds. Reisolation only produced F. graminearum.

Fig. 20 : Symptoms caused by F. graminearum on garden pea plants

4.8 Chemical control

4.8.1 Laboratory fungitoxicity tests against *F. graminearum*

At 0.1% concentration Benlate inhibited mycelial growth throughout all the testing period of 30 days. The initial growth of about 0.5 cm never increased in all petri dishes. Growth on Sisthane was slower attaining a radius of 6 cm at the end of the trial period. Ridomil had very little effect on mycelial growth. All petri dishes containing Ridomil were covered with mycelial growth seventeen days after inoculation (d.a.i.) while the control petri dishes were covered by mycelium within nine days.

At 1% concentration mycelial growth in plates with Benlate, Ridomil and the controls behaved as at the 0.1% concentration. Sisthane had an improved inhibition, delaying growth for seven days. The slow growth which started a week after inoculation continued till the end of the testing period when the mean radius remained 4 cm.

At 1% concentration Benlate and Sisthane equally inhibited mycelial growth inhibition. Rate of growth in the plates with Ridomil and the controls remained the same as in the previously described treatments.

The same results were recorded at 1.5% concentration which was not significantly different from the 1% rate (Fig. 21).

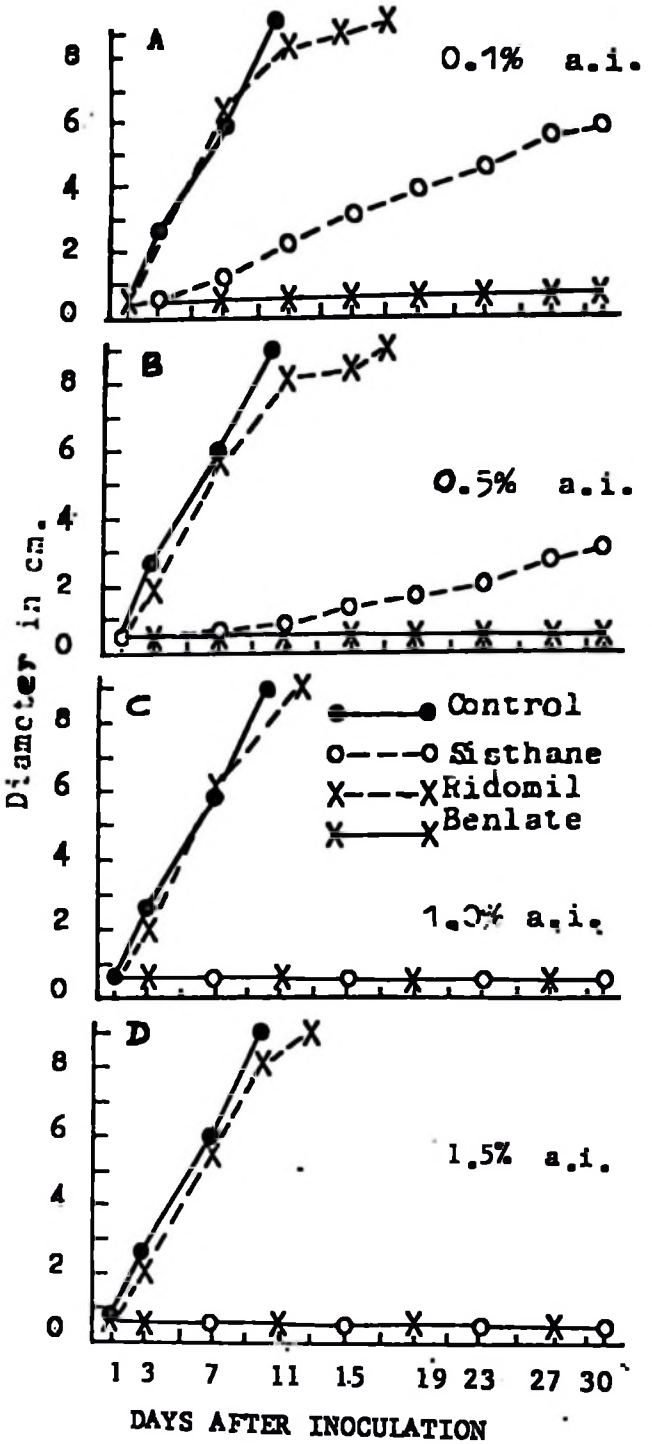


Fig. 11: Reaction of F. graminearum to Sisthane Ridomil and Benlate when tested in the laboratory using a fungitoxicity method.

4.8.2 Laboratory fungitoxicity tests against *F. moniliforme*

At 0.1% concentration, Benlate inhibited mycelial growth throughout the entire experimental period of 30 days. Sisthane at 1.0 and 1.5% concentration also caused some inhibition to mycelial growth. Plates with Sisthane, mycelium grew to no more than 6 cm 30 days after inoculation while all the Ridomil - treated plates were fully covered by mycelium 21 days after inoculation.

At the 0.5% concentration Benlate and Ridomil behaved as at 0.1% but Sisthane delayed growth for 11 days. 30 d.a.i. mycelium in the Sisthane plates had grown only up to a radius of 3 cm.

At 1 and 1.5% concentrations Benlate and Sisthane inhibited mycelial growth equally while there was no change in reaction to Ridomil.

Mycelial growth in all uninoculated control plates was rapid and covered the plates after 9 days (Fig. 22).

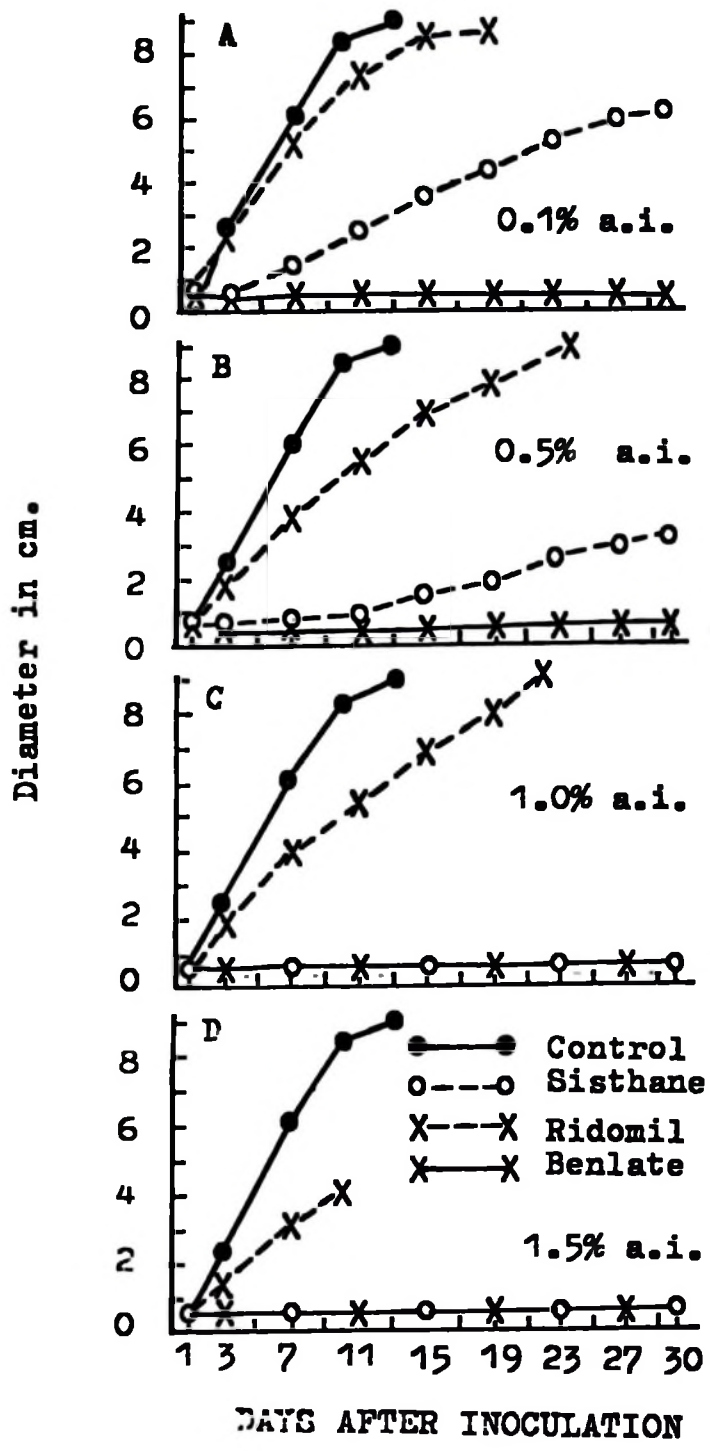


Fig. 22 : Reaction of F. moniliforme to Sisthane, Ridomil and Benlate when tested in the laboratory using a fungitoxicity method.

4.8.3 Laboratory difusibility tests against F. moniliforme and F. graminearum

At 0.1% concentration there was almost no inhibition to mycelial growth by all three fungicides. At 0.5% and 1% Benlate and Sisthane showed some limited inhibition, Benlate giving better results to F. moniliforme. At 1.5% Benlate still gave better results than the other two fungicides (Fig. 23 and 24).

Mycelium overgrew filter papers dipped in Sisthane and Ridomil solutions but was inhibited by those dipped in Benlate at 1% (Fig. 25 and 26 lower plates). Benlate had a toxic effect on F. moniliforme at the 1% concentration but at 0.1% however, filter papers were also overgrown (Fig. 26). Filter papers dipped in 0.1% Benlate solution were completely overgrown by F. graminearum but those dipped in 1% Benlate solution were only partially covered (Fig. 23 and 24).

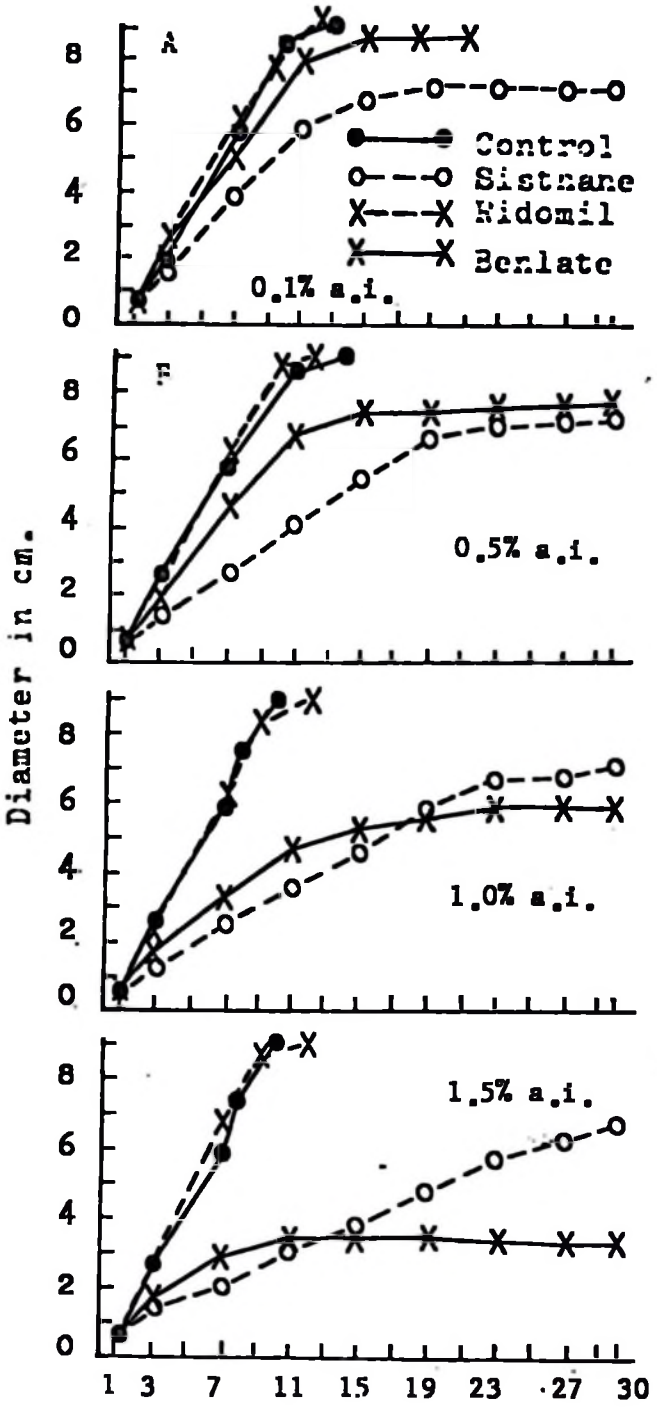


Fig. 23 : Reaction of F. graminearum to Sisthane, and Benlate when tested in the laboratory using a diffusibility method.

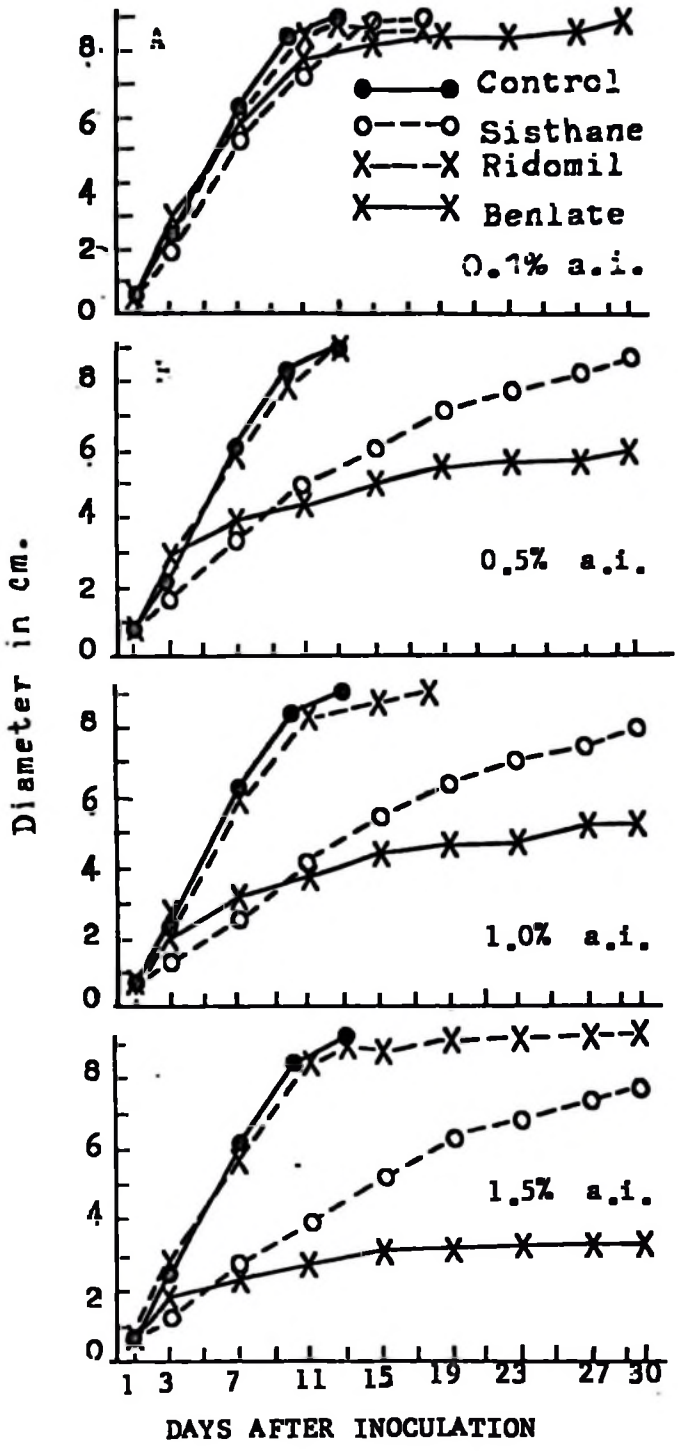


Fig. 24 : Reaction of F. moniliforme to Sisthane, Ridomil, and Benlate when tested in the laboratory using a diffusibility.

4.8.4 Effect of Benlate to sunflower plants inoculated with F. moniliforme and F. graminearum

Symptoms caused by both F. moniliforme and F. graminearum to plants sprayed 24 hours before inoculation were the same as those inoculated 24 hours after they were sprayed. Three days after inoculation all plants showed symptoms starting with small specks which later enlarged to cover big portions of the stems. Severely infected plants started dying on the fourth day and by the end of the eighth week many of them had already been killed, (Tables 7 and 8).

Although spraying 24 hours after inoculation resulted in more deaths than when it was done 24 hours before, F. graminearum killed more plants than F. moniliforme. Unsprayed plants which were inoculated with F. moniliforme survived better than sprayed indicating that Benlate had some phytotoxicity to young sunflower plants.

In the field, Benlate had very little effect in preventing symptom development on inoculated plants. Four days after inoculation all sprayed and unsprayed plants developed symptoms similar to those formed in the glass house.

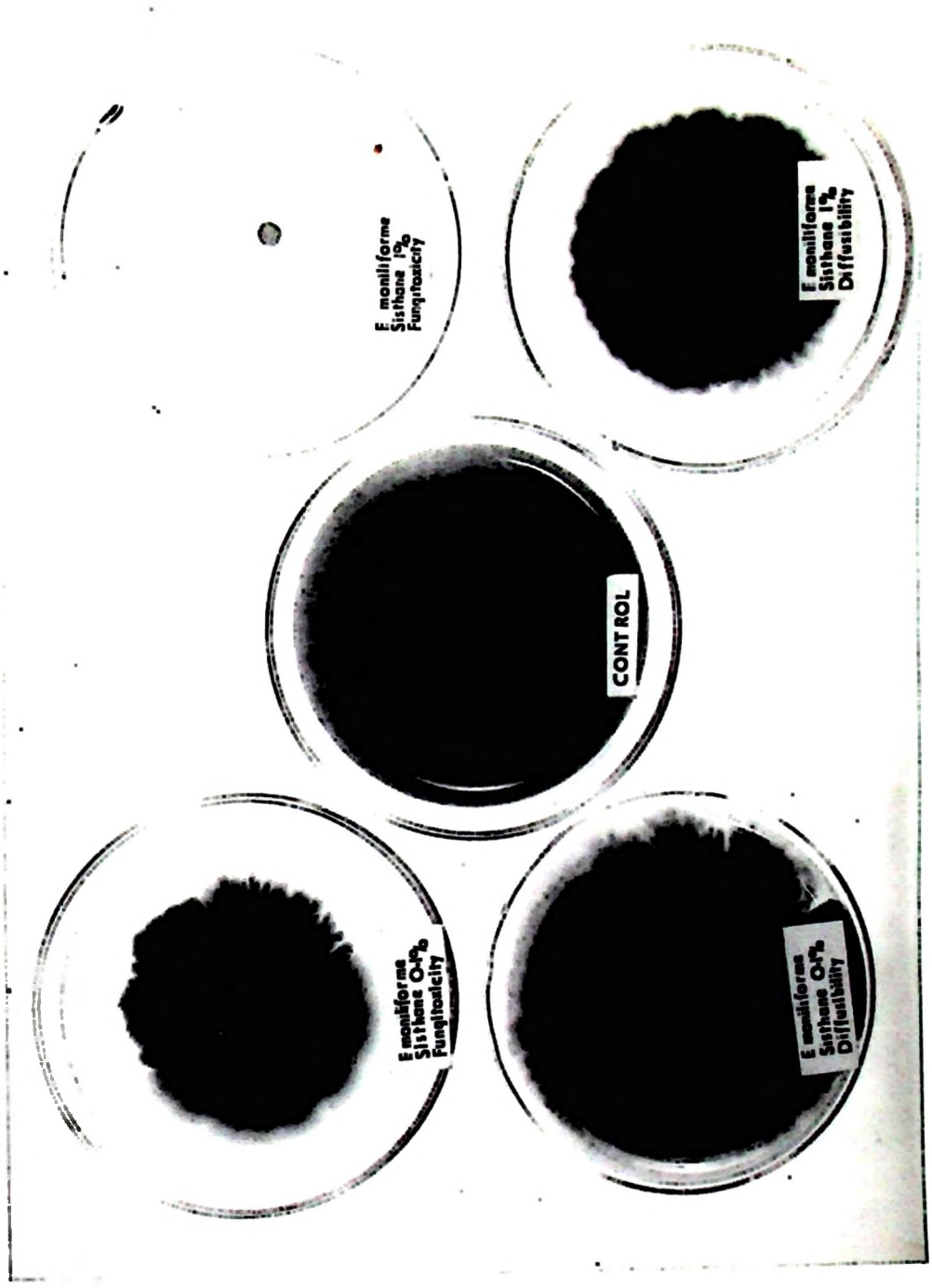


Fig. 25 : Fungitoxicity (left) and Diffusibility (right) testing of Sisthane against F. moniliforme with a control (untreated) plate in the centre

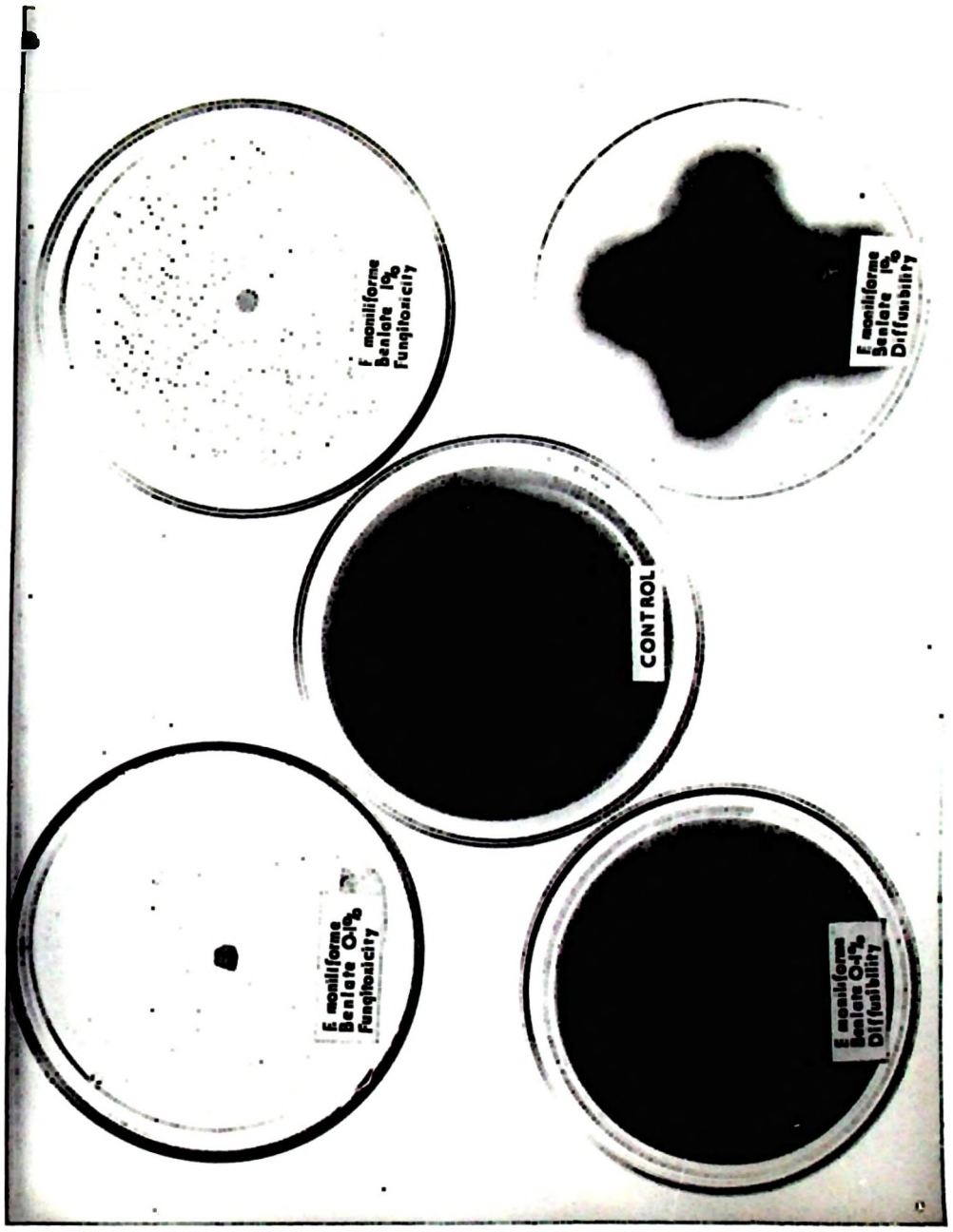


Fig. 26 : Fungitoxicity (left) and Diffusibility (right) testing of Benlate against F. moniliforme with the control plate in the centre

Table 7 : Number of plants killed by F. moniliforme (Fm) and F. graminearum (Fg) when benomyl 50% WP was sprayed 24 hours after inoculation in a glasshouse

% a.i.	Species	WK.1	WK.2	WK.3	WK.4	WK.5	WK.6	WK.7	WK.8	Total % killed out of 25
0.5	Fm	0	0	0	0	0	0	2	10	48
	Fg	5	0	0	0	0	0	0	11	44
1	Fm	0	0	0	0	0	0	0	0	0
	Fg	5	0	4	0	0	6	0	5	80
1.5	Fm	0	0	0	0	0	0	6	4	40
	Fg	0	9	0	0	0	6	0	6	84
0	Fm	0	0	0	0	1	1	2	0	16
0	Fg	6	1	0	0	0	0	0	0	28
0	0	0	0	2	2	0	0	0	1	12

Table 8 : Number of plants killed by F. moniliforme (Fm) and F. graminearum (Fg) when benomyl 50% WP was sprayed 24 hours before inoculation in a glasshouse

% a.i.	Species	WK.1	WK.2	WK.3	WK.4	WK.5	WK.6	WK.7	WK.8	Total % killed out of 25
0.5	Fm	0	0	0	0	0	0	0	0	0
	Fg	0	0	10	5	0	0	0	0	60
1	Fm	0	0	0	0	0	0	0	5	20
	Fg	0	0	0	0	0	0	4	10	56
1.5	Fm	0	0	0	0	0	0	0	6	24
	Fg	0	4	0	0	0	0	0	12	64
0	Fm	0	0	0	0	0	0	0	6	24
	Fg	25	0	0	0	0	0	0	0	100
0	0	0	0	0	0	0	0	0	0	0

5. DISCUSSION

During a routine plant disease survey in the University farm near Morogoro town, Professor C.L. Keswani came across an unusual diseases attacking sunflower plants. Within a few weeks the same disease was reported to be severe in a large farm at Dakawa some 50 kn north of Morogoro town.

A quick survey made in other fields revealed that the then unknown disease had already spread in many neighbouring villages within Morogoro rural and urban districts. The manner in which the stem rot appeared to be spreading and the apparent threat which faced the sunflower industry triggered off interest in carrying out more detailed studies on this disease which appeared potentially threatening.

To identify the causal organism(s) of the new disease, tow fungi ware always isolated from all diseased samples collected from fields in Morogoro Region. These were identified as Fusarium moniliforme and F. graminearum. They were readily grown on potato dextrose agar, malt extract agar and Czapek Dox agar. When young sunflower plants were inoculated with the above cultures they became diseased showing symptoms exactly the same as those observed in the field. Isolates made from such laboratory inoculated plants yielded F. moniliforme and F. graminearum, thus positively completing Koch's postulate. This was the first record not only in Tanzania but also elsewhere in the world. However, all isolates always produced both species in the same culture and in order to separate them, it was always necessary to subculture them into diffe-

rent plates according to the colour, shape and growth characteristics. This was a clear indication that the sunflower stem rot was being caused by both F. moniliforme and F. graminearum in combination.

Pathogenicity tests proved that F. graminearum alone was more pathogenic to sunflower plants than F. moniliforme since it always killed all young test plants. When a mixture of both species was inoculated to plants, symptoms developed more slowly than those of F. graminearum alone but quicker than those caused by F. moniliforme. The mixture took a longer time to kill incoulated plants and in many cases not all plants were killed as it was the case with F. graminearum alone. Hence a synergistic interaction of F. graminearum to F. moniliforme and an anergistic interaction of F. moniliforme to F. graminerarum.

In the field not all diseased plants died some remained weak produced small heads but survived. The most susceptible plants were found to be those which were weak and short (Fig. 2c). Most such plants always lodged and later died. The stronger and more robust plants either were not infected or if they were, the necrotic areas remained dry and all such plants survived with normal heads . Under field conditions, though, due to differences in time of germination, competition for light induced by close spacing and the traditional broadcasting planting "method" the number of short and weak plants in any sunflower field at or a few weeks before flowering was considerably high. In places where infection

was serious, almost all short and weak plants lodged and later died resulting in heavy plant losses.

Fortunately, the disease has only been confirmed in Morogoro Region. The survey carried out in all major sunflower growing regions (Fig. 5) revealed that only plants in Morogoro Region were infected. Besides the presence of inoculum in the Morogoro environment and the fact that Morogoro is on a much lower altitude no other reasons could be advanced as why the disease was confined to Morogoro alone.

Climatic conditions, time of planting and soil type were all investigated. The sunflower growing areas in Arusha, Kilimanjaro, Iringa and Mbeya were in many aspects similar to those of Morogoro Region. Artificial inoculations carried out in Arusha (at TPRI) both in the laboratory and in the field produced typical symptoms. This gives an indication that climatically, conditions in Arusha were not unsuitable for the occurrence of stem rot on sunflower.

Temperatures ranging from 18 to 30°C were found suitable for the general growth of both F. moniliforme and F. graminearum. Culture of F. moniliforme grown in incubators set at different temperatures grew fastest at between 20 and 30°C (Fig. 14). Those of F. graminearum grew fastest at between 20 and 25°C (Fig. 14). Booth (1971) states that optimum growth of F. moniliforme is attained at temperatures ranging from 25 - 35°C and of F. graminearum at 24 - 26°C. Both temperature ranges are in agreement with those found suitable in the TPRI laboratory.

During the months of May and June when sunflower was found susceptible to the stem rot in Morogoro, day time air temperature was found to lie between 22 and 24°C (Appendix 11) a relatively conduceve climate for both F. moniliforme and F. graminearum. Although the disease was not found outside Morogoro Region, temperature in Iringa, Kilimanjaro and Arusha ranged between 17 and 23°C (Appendix 10) a climate also suitable to disease development.

Data on relative humidity taken at 0900 and 1500 hours in major sunflower growing districts were also not found significantly differing from those taken in Morogoro, (Appendices 10 and 11). Chances of disease spread to such regions, therefore, remain a likely possibility.

Trials conducted to identify the most suitable medium for optimum growth of the pathogens revealed that all tested media were suitable for normal growth of both species (Fig. 15). This finding agreed with Booth's statement (1971) that "Fusaria will grow on most standard mycological media.

Traditionally, sunflowers grown in Tanzania have for a long time been referred to as "mixed", the composition of which has been cultivars distinguishable by their colours, including white, black or striped. This fact was confirmed during the survey when it was found that most small scale farmers particularly in Iringa and Mbeya Regions were still growing a complete mixture of different coloured seeds. Large scale farmers who have mushroomed up in recent years have adopted the use of known pure varieties in many cases the

black and striped. In this colour grouping there happened to be the most widely grown varieties, Record and Jupiter which also fetched premium prices on local official markets (Appendix 4).

More recently, however, many more varieties have been introduced into this country in order to select the more adaptable cultivars.

When F. moniliforme and F. graminearum were mechanically inoculated to the most promising and the already widely cultivated varieties in the laboratory all of them got infected. While F. graminearum killed all test plants, none of the plants within the batches inoculated with F. moniliforme got killed.

When all these varieties were planted in a TPRI field for natural infection (1979 and 1980), all of them remained healthy. This was another confirmation to the earlier findings, during the survey of Arusha and Kilimanjaro regions that both regions were still free from the sunflower stem rot.

All tested varieties got naturally infected when they were planted at a farm in Morogoro (Table 5). The degree of infection differred among varieties and when results obtained in two seasons (1980 and 1981) were computed, it was concluded that all tested varieities were susceptible. However, since varieties like Record, Jupiter, Gor 104 and Dwarf comet had comparatively less overall mean number of diseased plants; and the same varieties are still commecially preferred, in places where disease pressure is low such

varieties should be recommended to growers till better lines are found.

When most crops grown in association with sunflower were mechanically inoculated with F. moniliforme and F. graminearum in the laboratory, maize, sorghum garden peas and beans were susceptible to the disease. Groundnuts, simsim cowpeas and greengram, though some minute lesions were recorded, both fungi had no effect on their general growth.

Field observations made in all infested areas during this study failed to locate any infection among the intercropped plants under natural conditions.

The control of stem rot in sunflower fields by using fungicides was found ineffective. Ridomil and Sisthane failed to inhibit fungal growth in the laboratory and both were therefore discontinued from subsequent field trials. Benlate which was found effective in the laboratory tests was later proved ineffective in the field. Further, it was found to induce more deaths to sunflower seedlings than unsprayed controls (Table 7) indicating that, Benlate sprayed at the concentration found effective in the laboratory was phytotoxic to young sunflower.

6. CONCLUSIONS

These studies have established the status of sunflower stem rot in Tanzania. It was confirmed that this disease is confined to Morogoro Region alone although other regions like Arusha, Kilimanjaro, Iringa and Mbeya were found to have similar conducive climatic conditions.

Among the many intercropped plants maize, sorghum, beans and garden peas were found sufficiently susceptible to both F. moniliforme and F. graminearum when mechanically incoulated. However, under natural conditions, even in heavily diseased fields, all crops grown in association with diseased sunflowers remained disease-free.

Since intercroppings is already a popular practice all over the country villages where stem rot is endemic, farmers should be encouraged to intercrop with resistant crops like groundnuts, cowpeas, greengram and simsim. In places where farmers must intercrop with susceptible crops like maize, sorghum and beans, spacing must avoid overcrouding and sunflower varieties, which were found less infected by shem rot in the field like: Record, Jupiter, Dwarf comet and Gor 104 should be made available to farmers. Also, during weeding and thinning the susceptible weak plants must be removed to avoid too humid conditions and to give room to robust plants which were found less affected by the stem rot.

Stem rot control by using fungicides was not found feasible. All tested fungicides were not effective to artificially inoculated plants and therefore could not be considered for any further field trials.

The only logical method which can reduce losses likely to be caused by the sunflower stem rot is the use of resistant varieties. Of all tested introductions Record, Jupiter Gor 104 and Dwarf comet were less diseased under field conditions when disease pressure was low. These could still be grown by farmers in affected zones; but efforts should be directed in screening more varieties so that better lines can be obtained for future breeding work.

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8. APPENDICES

Appendix 1 : Major sunflower producing countries (1,000 mt)

	1977	1978	1979	1980	1981	1982	1983
Albania	25	26	37	37	38	38	53
Angola	13	10	10	10	10	10	2
Argentina	900	1000	1430	1650	1280	1780	2300
Australia	75	158	186	159	142	139	1
Bulgaria	423	390	426	415	457	507	448
Canada	81	120	218	166	165	94	51
Chile	15	30	33	38	7	5	7
France	92	82	172	273	413	623	837
Hungary	215	225	419	456	627	610	638
Iran	19	20	22	10	15	15	1
Kenya	13	14	15	15	15	16	17
Malawi	2	3	3	3	4	4	4
Mexico	3	7	31	25	9	9	20
Morocco	19	16	40	18	20	21	19
Mozambique	10	11	22	22	20	20	20
Romania	807	816	888	817	810	835	705
South Africa	484	453	320	332	518	260	202
Spain	388	470	504	488	300	656	674
Turkey	455	485	590	750	575	620	725
Tanzania	6	59	31	40	40	41	42
Uruguay	34	72	51	48	41	46	20
U S A	1252	1732	3309	1726	2098	2661	1447
U S S R	5904	5333	5414	4650	4600	5300	5300
Yugoslavia	479	539	525	306	327	227	135

Source : FAO Production Year Book 32 (1978), 130; 34 (1980), 131, 36 (1982) 142 and 37 (1983) 144. Food and Agric. Organisation of the United Nations, Rome.

Appendix 2 : World Regional Sunflower Production
1977 to 1983 (1,000 ha and 1,000 mt)

	1977	1978	1979	1980	1981	1982	1983
World Area	9818	11480	12165	12374	12119	13015	12779
Total Production	11956	13069	15276	13174	14204	16226	15766
Yield kg/ha	1218	1138	1256	1065	1172	1247	1234
Africa - Area	528	643	534	506	557	549	482
Total Production	576	594	478	478	671	414	354
Yield kg/ha	1091	924	896	946	1204	800	733
N & C America - Area	963	1235	2386	1683	1710	2135	1315
Total Production	1337	1859	3558	1917	2272	2764	1518
Yield kg/ha	1387	1505	1490	1129	1329	1294	1155
South America - area	1304	2148	1673	2039	1394	1693	1975
Total Production	950	1705	1521	1756	1369	1881	2349
Yield kg/ha	728	794	909	861	982	1111	1189
Asia - area	537	828	921	1214	1757	1800	1932
Total Production	614	820	1063	1363	2050	1910	2463
Yield kg/ha	1144	989	1154	1122	1167	1102	1275
Europe - area	1777	1847	2055	2128	2267	2448	2601
Total Production	1501	2600	3058	2853	3103	3661	3690
Yield kg/ha	1408	1408	1488	1341	1369	1509	1418
Australia and Oceanic area	270	440	822	450	396	356	338
Total Production	150	316	372	318	278	230	186
Yield kg/ha	1112	1436	1428	1410	1403	1396	1100
U S S R - area	4574	4558	4334	4580	4235	4250	4305
Total Production	5904	5333	5414	4650	4600	5300	5300
Yield kg/ha	1291	1170	1249	1015	1086	1247	1231

SOurce : FAO Production year book 32 (1978) 130; 34 (1980), 131, 36 (1982) 142 and 37 (1983) 144. Food and Agric. Organisation of the United Nations, Rome

Appendix 3 : Regional collections of sunflower seeds by GAPEX in metric tons (1971/72 - 1979/80)

Region	Season	1971/ 72	1972/73	1973/74	1974/75	1975/76	1976/77	1977/78	1978/79	1979/80
Arusha		49	75	47	73	24	20	237	402	322
Dodoma		756	1143	767	721	616	801	1668	3079	3654
Mtwara		57	67	258	27	72	57	10	9	12
Kilimanjaro		62	135	70	199	82	108	177	138	55
Tanga		30	38	10	9	3	6	5	8	5
Mwanza		320	465	343	22	15	180	78	241	500
Morogoro		2059	2532	2638	2612	1789	960	1533	3502	1463
Tabora		415	600	262	519	369	-	789	1081	2103
Shinyanga		1261	1843	1389	1258	1120	1010	888	1816	2242
Mbeya		19	27	22	53	82	17	123	189	302
Iringa		1110	2453	303	1537	1482	1383	1342	2116	2796
Rukwa		-	-	-	-	1	7	2	3	59
Coast		-	-	-	-	-	-	2	-	-
Singida		78	66	129	-	267	74	367	336	1760
Kagera		-	-	-	-	-	-	-	-	-
Kigoma		-	-	4	1	-	-	-	-	-
Lindi		-	-	-	-	-	-	1	4	3
Mara		8	20	10	-	-	-	-	-	48
Ruvuma		-	-	-	-	-	-	106	156	264
Total		6164	9464	6247	7031	5922	4663	7228	12080	15497

Source: Anonymous, 1980, Bulletin of Crop Statistics 1979. Statistics Section, Planning Division Ministry of Agriculture, Dar es Salaam, Tanzania.

* Up to 31-03-1980

Appendix 3 contd.

Region	1980/81	1981/82	1982/83	1983/84	1984/85
Arusha	373	184	73	50	108
Dodoma	2469	1540	686	1120	1589
Mtwara	5	2	-	-	4
Kilimanjaro	69	5	1	-	-
Tanga	25	12	27	7	-
Mwanza	342	389	205	49	109
Morogoro	1357	1191	499	168	287
Tabora	1238	583	523	239	465
Shinyanga	2092	1243	703	503	638
Mbeya	190	35	1	118	7
Iringa	1647	1157	140	150	323
Rukwa	362	44	3	5	8
Coast	-	-	1	-	-
Singida	2507	2628	2579	1877	3429
Kagera	-	12	1	-	-
Kigoma	-	18	17	9	-
Lindi	776	2	1	3	1
Mara	-	13	12	6	66
Ruvuma	46	314	240	233	70
Total	13498	9372	5712	4537	7103

Sources : Anonymous 1985. Price policy recommendations for the 1985, Agricultural price review Vol. 3, Oil seeds(prices for 1986/87 marketing season).

Marketing Development Bureau, Dar es Salaam 1985 R 4/85.

Appendix 4 : Producer prices for sunflower (1972/73 - 1986/87)

Price in T.shs. per metric ton

Year	Black	Jupiter	Others
1972/73	570	570	
1973/74	550	550	550
1974/75	750	750	750
1975/76	1000	1000	750
1976/77	1100	1100	800
1977/78	1500	1500	1250
1978/79	1700	1500	1300
1979/80	1700	1500	1400
1980/81	1800	1600	1500
1981/82	200	1800	1700
1982/83	2900	2600	2500
1983/84	4000	3500	3200
1984/85	6000	5250	4800
1985/86	8400	7400	6700
1986/87	9250	8150	7400

Sources : Bulletin of Crop Statistics, Section Planning Division, Ministry of Agriculture and Livestock Development

and

Price Policy Recommendations for the 1985 Agricultural Price Review Vol. 3 Oilseeds (prices for 1986/87 marketing season)

Marketing Development Bureau Dar es Salaam 1985 R 4/85.

Appendix 5 : Mean daily growth of F. moniliforme at different temperature in cm

Degrees Celsius	Days after inoculation															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	21
8	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.4	0.4	0.4	0.5	0.5	0.5	0.6	0.7	1.3
10	0.0	0.0	0.3	0.4	0.6	1.6	1.0	1.8	1.8	2.1	2.5	2.5	2.8	2.8	3.2	3.06
18	0.1	1.5	2.6	3.4	4.5	6.6	7.4	0.0	8.7	9.0						
20	-	2.2	3.6	4.7	4.9	7.0	8.0	8.6	9.0							
25	0.1	1.5	2.8	3.7	4.5	5.5	6.5	6.9	7.5	8.0	8.5	9				
30	-	1.8	2.5	3.1	4.4	4.6	4.9	5.5	6.1	6.7	7.2	8.2	8.7	9.0		
35	-	-	0.2	0.5	1.1	1.1	1.1	2.1	2.5	2.8	2.8	3.2	3.3	3.4	6.5	
40	0	0	0	0	0	0	0	0	0	0	0	0	0	0		

Appendix 6 : Mean daily growth of F. graminearum at different temperatures in (cm)

Degrees Celsius	Days after inoculation														
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	21
8	0.0	0.0	0.0	0.0	0.15	0.25	0.6	0.78	1.83	1.05	1.23	1.28	1.55	1.63	2.2
10	0.0	0.0	0.0	0.0	0.7	0.98	0.95	2.13	2.3	2.6	2.8	2.9	3.4	3.5	5.0
18	0.0	1.2	2.5	3.2	4.2	5.2	6.3	8.8	7.8	8.6	9.0				
20	0.0	1.5	2.7	3.9	5.1	6.3	7.2	7.8	8.5	9.0					
25	0.0	1.0	1.8	3.0	4.5	6.0	7.0	8.0	8.5	9.0					
30	0	0	0.0	0.1	0.3	0.4	0.5	0.6	1.0	1.1	1.18	2.0	2.2	2.3	2.4
35	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
40	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Appendix 7 : Mean daily growth of F. moniliforme on different media at between 20-25°C

(diameter in cm)

Media/ days	1	2	3	4	5	6	7	8	9	10	11	12	13	14	21
MEA	0.9	1.8	2.7	3.5	4.6	6.2	7.2	7.8	8.4	8.9	9.0				
DOX	0.7	1.6	2.7	4.0	4.4	7.5	8.0	8.5	9.0						
MEA - 3	0.5	1.2	2.9	5.1	6.1	7.5	8.5	9.0							
PDA	0.1	1.5	2.8	3.7	4.5	5.5	6.5	6.9	7.5	8.0	8.5	9.0			

Appendix 8 : Mean daily growth of F. graminearum on different media at 20-25°C

(diameter in cm)

Media/ Days	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
MEA	0.4	0.7	1.2	1.7	2.2	2.9	2.2	3.8	4.5	5.0	5.4	5.9	6.5	7.0	8.5	9.0
DOX	0.8	1.8	3.1	4.5	5.9	7.5	8.5	9.0								
MEA-3	0.2	1.2	2.2	3.5	4.3	5.3	6.6	7.5	8.5	9.0						
PDA	0.1	1.5	2.8	3.7	4.5	5.5	6.5	6.9	7.5	8.0	8.5	9.0				

Appendix 9 : Elevation, sowing/harvesting months and types of soils for some of the main sunflower growing areas

Area	Metres a.s.l.	Sowing	Harvest	Soils
Iramba	1524	January	May/June	Sandy loam
Singida	1524	January	May/June	Snady loam
Mpwapwa	987.6	January	May/June	Redish sandy light loam
Kongwa	1005.8	January	May/June	Redish sandy light loam
Kahama	1219.2	November	April	Mbuga sandy loam
Morogoro	531.8	March/April	July/August	Light sandy loam
Iringa	1371.6	March/April	July/August	Redish sandy loam
Mbeya	1706.9	Feb/March	June/July	Clay loam
Arusha	1432.6	April	August	Volcanic sandy loam
Kilimanjaro(Moshi)	835.4	March/April	July/August	Volcanic red loam

Source : Operational Navigation Chart Map. Stock No. ONCXXM05 and Correspondence in File TP3/4/12 from Regional Agricultural Development Officers.

a.s.l.= above sea level.

Appendix 10 : Long term mean temperature (^{o}C) and relative humidity (Rh) in % at 1500 hours for some sunflower growing areas

Station	January		Feburary		March		April		May		June		July		August	
	^{o}C	Rh%	^{o}C	Rh%	^{o}C	Rh%	^{o}C	Rh%	^{o}C	Rh%	^{o}C	Rh%	^{o}C	Rh%	^{o}C	Rh%
Mpwapwa	24	55	23	60	*23	53	23	60	21	51	20	44	19	43	20	39
Tabora	23	55	23	56	23	57	23	56	22	46	21	47	21	34	22	33
Arusha	21	53	21	52	21	60	20	68	19	69	17	66	17	63	18	57
Kilimanjaro (Moshi)	25	41	23	41	25	46	24	58	23	64	21	58	21	54	21	50
Morogoro	26	55	26	67	26	60	25	60	24	68	22	59	21	54	22	49
Iringa	21	61	21	64	21	65	20	61	20	51	18	44	18	44	17	-
Mbeya	18	69	18	69	18	71	18	70	15	61	14	52	13	47	15	44

* Underlined months - Expected susceptibility of sunflower to stem rot

Source : Director General,
Monthly Farming Weather,
Directorate of Meteorology,
P.O.Box 3056,
DAR ES SALAAM

Appendix 11 : Mean relative humidity. Average for several years upto 1984 in Percentages

	0900 hours						
	Jan.	Feb.	M	A	M	Jn	Jl
Dar es Salaam	79	79	83	87	86	84	85
Arusha	79	80	84	91	89	87	86
Moshi	72	72	76	85	86	82	82
Tabora	84	84	83	81	73	65	61
Dodoma	80	82	81	81	76	72	73
Mbeya	81	81	81	79	75	75	72
Mtwara	84	85	86	86	79	79	80
Morogoro	88	81	90	80	79	83	81
Songea	86	87	88	87	83	78	76

Source : Anonymous (1984) Statistical Abstracts. Bureau of Statistics Ministry of Finance Planning and Economic Affairs, Dar es Salaam.

Appendix 12 : Means in percentages of infected sunflower at Morogoro field in 1980

Varieties/Reps	I	II	III	IV	Total	$\bar{X}$
Record	0.0	10.5	25.0	5.0	40.5	10.1
Jupiter	6.3	0.0	9.0	0.0	15.3	3.8
Gor 104	18.8	31.3	0.0	6.7	56.8	14.2
Dwarf comt	12.5	7.7	13.3	29.4	62.9	15.7
Giant 549	62.5	12.5	22.2	23.5	120.7	30.2
1S 8944	35.3	38.1	52.3	33.5	159.0	39.8
1S 893	23.1	33.3	26.7	26.3	109.4	27.4
1S 844	46.7	20.0	23.8	50.0	140.5	35.1
56 F5	7.7	33.3	27.8	38.9	107.7	26.9
6 F5	83.3	71.4	66.7	75.0	296.4	74.1
	296.2	258.1	266.8	288.1	1009.2	
X^2	15414.4	10472.8	10612.1	12925.3	49424.59	

C.F. $1109.2^2 \div 40 = 30758.1$

Total SS = 49424.6 - CF = 18666.5

Reps SS = 308533.9 $\div$ 10 - CF = 95.29

Treats SS = 180067.34 $\div$ 4 - CF = 14258.74

Error SS = 18666.5 - (95.29 + 14258.74)

Reps	DF	MS	F	0.05
	3	95.29		
Treats	9	14258.74	1584.3	9.92^{**}
Error	27	4312.47	159.7	

S.E $\frac{2 \times 159.7}{4} = 8.94$

LSD (P = 0.05) = 8.94 x 2.052 (t) = 18.3

(P = 0.01) = 8.94 x 2.771 (t) = 24.8

Appendix 13 : Means in percentages of infected sunflower at Morogoro field 1981

Varieties/Reps	I	II	III	IV	Total	Means
Gor 704	24.07	10.34	7.14	17.34	58.89	14.72
Giant 549	15.00	23.72	28.81	13.79	81.32	20.33
56/F5	21.05	18.75	21.17	8.62	69.79	17.40
IS 8944	8.47	16.67	14.28	17.85	57.27	14.32
IS 849	18.51	20.68	13.46	14.54	67.19	16.80
IS 893	13.33	13.35	15.51	23.03	65.22	16.31
Dwarf comet	16.32	10.90	14.28	10.71	52.21	13.15
Record	19.15	7.27	10.00	18.64	55.06	13.77
Jupiter	5.35	27.45	15.00	20.37	68.17	17.04
6/F5	23.63	23.65	14.00	5.00	66.15	16.54
	164.88	172.65	153.65	149.89	641.03	
x^2	3059.58	3383.25	2679.73	2527.65	11650.21	

CF = $641.07^2 \div 40 = 10274.27$

Total SS = 11650.21 - CF = 1375.94

Reps SS = 103068.77 ÷ 10 - CF = 32.61

Treats SS = 41752.2 ÷ 4 - CF = 163.78

Errors SS = 1375.94 - (32.61 + 163.78) = 1179.56

	DF	MS	F	0.05	
Reps	3	32.61	10.87		
Treats	9	163.78	18.2	0.43	NS
Error	27	1179.56	43.69		

S.E. = $\frac{43.69}{4} \times 2 = \underline{4.67}$