

STUDIES ON THE WILT OF EGGPLANT CAUSED BY
Pseudomonas solanacearum E. F. SMITH & ITS CONTROL

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CERTIFICATE

This is to certify that the work recorded in the thesis entitled "Studies on the wilt of eggplant caused by Pseudomonas solanacearum E.F. Smith and its control" submitted by Mr.A.K.M. Anwar Habib for the award of the degree of Doctor of Philosophy in Horticulture of the Bidhan Chandra Krishi viswavidyalaya, is the faithful and bonafide research work carried out under my supervision and guidance. The results of the investigations reported in the thesis have not been submitted for any other Degree or Diploma. The assistance and help received during the course of investigation have been duly acknowledged.


(M. G. Som)

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Aumak
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INTRODUCTION

Vegetables rank next to cereals as source of carbohydrates and for large number of people in developing countries of Africa and Asia, vegetables are supplementary source of nutrients like carbohydrates, proteins, vitamins, minerals and energy (N.R.C., 1978; F.A.O., 1953-1972). In many of the less developed countries of the world diets are chiefly comprised of cereals, pulses and vegetables. Besides nutrition, vegetables are important source of dietary fibre comprised of unassimilable cellulose, hemicellulose, pectin and lignin. These dietary fibres provide the "roughage" and have important effects on gut functioning due to their bulk, their ability to absorb water, and their being substrates for the normal bacterial flora of the gut (Jones, 1978; Burton, 1982).

The word 'comprise' usually not followed by 'of'.

In most developing countries including India and Bangladesh, the population would be double by the end of 20th century and the demand for food in general, and vegetables in particular, would be double or more. To meet this prodigious demand, increase in the production of vegetables capable of growing in all seasons is the goal of agriculture and the research scientists.

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Eggplant (English : Brinjal; Solanum melongena L.) is a solanaceous vegetable which is grown round the year throughout most part of India and Bangladesh (Singh, 1967; Chowdhury, 1980)

and is only second to the potatoes in its popularity (Kochhar, 1981). The unripe fruits of eggplant are consumed as boiled, cooked, fried or roasted vegetable dishes and also as pickles (Herklots, 1972; Kochhar, 1981). The eggplant is of high nutritive value and can well be compared with tomatoes (Chowdhury, 1976). The fruits contain approximately 4 to 6 per cent carbohydrates, 1 per cent protein, 0.3 per cent fat and large amount of minerals including calcium, phosphorus and iron (Chatfield, 1954; Aykroyd, 1963; Chowdhury, 1983; Som and Maity, 1986). The fruits are also quite rich in vitamin A, thiamine, riboflavin, nicotinic acid and vitamin C (Aykroyd, 1963; Chowdhury, 1983).

Many varieties of eggplant are grown in India and Bangladesh round the year (Singh, 1967; Chowdhury, 1980). The yield in India is 25 to 30 metric tonnes per hectare with normal farm management practices (Chowdhury, 1980, 1983). Eggplant is widely cultivated as a vegetable crop in the tropical, subtropical and warm temperate zones of the world. The world production is estimated to be about 4.6 million metric tonnes utilizing 1 million hectares of land (F.A.O., 1981). The leading eggplant producing countries of the world in order of importance are India, China, Japan, Turkey, Italy, Egypt, Iraq, U.S.A., and France (Kochhar, 1981).

Like other food crops, increase in agricultural production of eggplant can be increased by : (i) increase of land area under cultivation, (ii) introducing modern agricultural technologies including high-yielding varieties, adequate fertilizers and

irrigation, (iii) multiple cropping, (iv) controlling the diseases and pests in the field and (v) checking the post-harvest losses.

Of the field diseases of eggplant, bacterial wilt incited by Pseudomonas solanacearum E.F. Smith is by far the most devastating disease causing severe loss of yield and consequent economic losses to farmers wherever the crop is cultivated. In India, yield losses of 30-70% in Kumaon and Nilgiri hills (Sinha, 1985), 15-75% in Karnataka (Rao and Sohi, 1977) and 54-62% in West Bengal (Das and Chattopadhyay, 1955) have been reported. Control of this disease is a necessary pre-requisite for better exploitation of eggplant as a nutritious vegetable for millions and is a challenging problem to agriculture.

Effective control of eggplant wilt is dependent on a comprehensive understanding of the biology of the pathogen and the disease (Buddenhagen and Kelman, 1964; Kelman, 1976; Task Force Committee Report, 1976). The pathogenic variation with the species is enormous and at least three races (1, 2 and 3) have been identified (Buddenhagen et al., 1962; Hayward, 1964). A fourth race with some specificity to ginger has been proposed (Hayward, 1976). Most researchers working on bacterial wilt consider that wilt of eggplant is caused by race 1 of the pathogen (French and Sequeira, 1970; Buddenhagen and Kelman, 1964; Hayward, 1976; Rao, 1976; Addy et al., 1980). The identity of the race of the pathogen causing wilt of eggplant in West Bengal is poorly understood.

Mode of survival of P. solanacearum race 1 in the absence of host plant in nature is quite controversial (Buddenhagen and

Kelman, 1964; Schuster and Coyne, 1974). Several reports indicated persistence of the pathogen on tomato seeds and transmission of the pathogen from seeds to the seedlings (Remadevi and Menon, 1980; Moffett et al., 1981). Information regarding the persistence of the pathogen on eggplant seed and transmission of the disease through seeds of eggplant is insufficient. It is generally believed that race 1 of the pathogen can survive for long time in soil (Kelman, 1953; Buddenhagen, 1965; Buddenhagen and Kelman, 1964; Schuster and Coyne, 1974). Most workers, however, used indirect method of determining disease incidence in fields by sowing susceptible plants or counting population density by using selective medium (Buddenhagen, 1965; Nesmith and Jenkins, 1976). Except one report (Moffett et al., 1983) no attempt has been made so far to exploit direct method of using marker strain of the pathogen. Use of antibiotic resistant marker strain (Obaton, 1971) can provide definitive information with respect to growth and population dynamics of the pathogen in soil. Little is known of the ecological factors that affect the bacterium's longevity in soil or its disease producing potential in different soils (Task Force Committee Report, 1976).

In the past, many attempts have been made to control the wilt of solanaceous plants including eggplant caused by race 1 of the pathogen. Several workers have recommended crop rotation for reducing wilt in solanaceous crops (Foucart, 1959; Johnston, 1964; Bolsewinkel, 1976; Kincaid, 1960; Sohi et al., 1981). Because of the ability of the pathogen to persist in the soil, crop rotation practices have been found to be of limited success

in controlling the disease (Thurston, 1976). Different cultural practices such as manipulation of quantity and form of nitrogen fertilizer, and varying frequency of irrigation attempted by several early workers for potato or tomato wilts were only partially successful or prohibitively expensive (Kelman, 1953; Sequeira and Kelman, 1976; Thurston, 1976). Moreover, fertilizer and irrigation schedules effective for controlling of bacterial wilt of one crop under a particular agroclimatic condition may not be effective for other solanaceous crops. Little is known regarding the efficacy of varying fertilizer and irrigation schedules for controlling the wilt of eggplant caused by race 1 of the pathogen under the agroclimatic conditions of West Bengal.

A large number of chemicals and antibiotics are known to inhibit the pathogen in vitro (Winstead and Kelman, 1960; El-Goorani et al., 1978). Several attempts have been made to control the bacterial wilt by application of chemicals and antibiotics to soil (Sequeira, 1958; Sequeira and Kelman, 1976; Rao, 1976; Enfinger et al., 1979), but most chemicals were either phytotoxic or cost-intensive. The eggplant isolate of the pathogen (race 1) has been found to be sensitive to streptomycin and seedling treatment before planting significantly reduced the wilt incidence (Dutta and Verma, 1969). Little information is available regarding the effects of seed treatment on the incidence of wilt of eggplant.

It is generally believed that the use of disease resistant varieties offers the best control for wilt of solanaceous plants incited by P. solanacearum (Sequeira and Kelman, 1976; Thurston,

1976). Resistant varieties of tobacco and peanut have long been identified and used. Resistance to bacterial wilt of eggplant has been found and bred into commercially acceptable varieties in Puerto Rico (Roque, 1941), the Philippines (Empig et al., 1962), Ceylon (Park and Fernando, 1940) and U.S.A. (Winstead and Kelman, 1960). In India several eggplant cultivars found to be resistant to wilt under Bangalore conditions (Rao et al., 1976) showed susceptibility when tested in Kerala (Sitaramaiah et al., 1981) and Orissa (Das and Panda, 1985). Apparently screening for resistance of a large number of eggplant cultivars in diverse agro-climatic conditions should be conducted before definitive conclusion could be made.

The objectives of the present investigation were : (i) to study the source of primary infection and survival of a marker strain of the eggplant pathogen under the agroclimatic conditions of West Bengal, and (ii) to examine the efficacy of cultural, chemical and resistant cultivars in controlling the disease.

The Section I of the thesis is concerned with the identification and characterization of the race causing wilt of eggplant in West Bengal. Attempts were made to study the survival of the pathogen on seed and in soil by using a double marker strain resistant to 2 mg/ml streptomycin and 1 mg/ml erythromycin. Certain environmental factors affecting the survival of the organism in soil were also included.

The Section II of the thesis is concerned with control of the disease under field condition. A large number of germplasms

collected from India and Bangladesh were screened to identify the resistance source. Efforts were made to control the disease by varying doses of N, P and K, different oilcakes and manipulating the frequency of irrigation. Finally, an attempt was made to determine the efficacy of seed treatment in controlling the disease under natural infection conditions of the field.

In this thesis, attempt has been made only on a few aspects of diverse mode of survival of the pathogen in nature. Emphasis was laid on survival in seed and soil. Survival and transmission of the pathogen by insects, and in crop debris are not included in this study. For control by seed treatment, pre-sowing exposure of the seeds to different chemicals were tested. Treatment of seedlings before transplantation or application of the chemicals to root zones by soil drenching method were excluded.

The background information for the thesis has been drawn freely from previous works done in several laboratories. The excellent syntheses of the available data on bacterial wilt by Kelman (1953), Buddenhagen and Kelman (1964), Sequeira and Kelman (1976) and Alexander (1978, 1981) provided conceptual framework for the present investigation. Information concerned with the survival of the pathogen in nature in the absence of host has been drawn from the excellent reviews by Buddenhagen (1965), Leben (1974, 1981) and Schuster and Coyne (1974).

LITERATURE REVIEW

Bacterial wilt of solanaceous plants was first reported by Burril (1890) and the pathogen Pseudomonas solanacearum E.F. Smith was first described by Smith (1896). It is now well established that the bacterial wilt caused by P. solanacearum can affect more than 200 species of angiosperms and is a serious disease of a number of economically important crop plants such as potato, tomato, banana, eggplant, tobacco and chilli (Kelman, 1953; Buddenhagen and Kelman, 1964; Sequeira and Kelman, 1976). The disease is confined mainly to the tropics, subtropics and warm-temperate regions of the world, although a number of epidemic outbreaks have been reported in cool-temperate areas (Thurston, 1963; Lloyd, 1975; Harris, 1976). The earlier literature on the pathogen and the disease has been comprehensively reviewed (Kelman, 1953). Biological and physiological aspects of the disease and the pathogen including the races has also been reviewed (Buddenhagen and Kelman, 1964; Hayward, 1964). The present review will restrict to the literature relevant to race concept, occurrence, survival in between the planting seasons and control of the pathogen with special reference to eggplant isolate of the pathogen.

Race Concept. — There are distinct differences in the cultural and physiological properties among isolates of P. solanacearum from different hosts which various authors have

considered sufficient to establish sub-specific categories like races, pathovars or biotypes (Okabe, 1954; Okabe and Goto, 1952, 1961; Buddenhagen and Kelman, 1964; Hayward, 1964). Buddenhagen et al. (1962) reported isolates into 3 races : Race 1, affecting solanaceous hosts such as tobacco, tomato, eggplant and chilli and other plants; Race 2, pathogenic on triploid bananas and Heliconias; and Race 3, affecting potato and weakly pathogenic on other solanaceous crops. Hayward (1964) established four biotypes : Biotype I, II and III equivalent to Race 2, Race 3 and Race 1 of Buddenhagen et al. (1962) and Biotype IV affecting ginger mostly. Apparently, the eggplant isolate capable of infecting tomato and tobacco is considered as Race 1 in most of the literature on the pathogen (Sequeira and Kelman, 1976).

Occurrence of eggplant isolate. — Smith (1896) first reported the pathogen from eggplant, tomato and potato in South Carolina. Rolfs (1898) isolated the pathogen from eggplant in Florida. Kelman (1953) in his monograph on the pathogen has summarised the information indicating the existence of the eggplant isolate from different parts of North, Central and South America, Italy, Africa, Ceylon, China and Taiwan.

In India, the earliest record of the disease was on potato in Pune (Butler, 1903). Occurrence of the disease on potatoes was later confirmed from Mysore (Coleman, 1909), Bombay (Butler, 1918), Bengal, (Butler, 1918) Madras (Rajan, 1924) and Assam (Sen, 1930). In undivided Bengal, the disease on tobacco and tomato was reported from Rangpur (Hutchinson, 1913; Butler, 1918), and Dhaka (Hedayetulla and Saha, 1941), respectively. In West

Bengal, bacterial wilt of eggplant was recorded first by Das and Chattopadhyay (1955). Yield losses upto 62.5% due to the disease have been reported (Das and Chattopadhyay, 1955; Paily, 1964). Das and Chattopadhyay (1955), however, identified the pathogen as P. solanacearum var. asiaticum. Since then the bacterial wilt of eggplant, tomato, potato and chilli has been reported as a serious problem in Bihar, Orissa, Karnataka, Madhya Pradesh, Maharashtra and West Bengal (Hingorani et al., 1956; Rao, 1972; Sohi et al., 1981).

It is now well established that in India the disease causes serious losses in eggplant and tomato (Nayar and Mathew, 1982; Rao et al., 1975). In the vast plains of northern India, in the south plateau and in West Bengal, Race 1 of the pathogen has been found to be predominant and race 3 has been recorded only in the hills. The race 2 has not been reported so far from India (Shekhawat, 1976). Most literature on the occurrence of races of the pathogen considered that P. solanacearum var. asiaticum reported from India (Das and Chattopadhyay, 1955; Hingorani et al., 1956) and Australia (Harrison, 1961) is actually race 1 and the information has been summarized (Buddenhagen and Kelman, 1964). Addy et al. (1980) in a study of 30 tomato isolates of P. solanacearum from Assam and Orissa concluded that all isolates belonged to race 1 of Buddenhagen et al. (1962).

Survival and Primary infection. — Bacterial phytopathogens including P. solanacearum do not form resting spores or structures comparable to fungi and nematodes, yet they can survive from season to season (Buddenhagen, 1965; Leben, 1981).

The mechanism of survival of phytopathogenic bacteria in general and P. solanacearum in particular has been extensively studied and the literature has been reviewed (Buddenhagen, 1965; Leben, 1974; Schuster and Coyne, 1974). It is now generally accepted that the phytopathogenic bacteria survive in the absence of hosts in association with following animate or inanimate agencies : (a) Seeds, (b) Weed hosts, (c) Insects, (d) Crop debris and (3) Soil (Schuster and Coyne, 1974; Leben, 1981). The literature with respect to survival and primary inoculum of P. solanacearum is conflicting and has been summarised in this review.

Insect transmission . — Smith (1896) reported that insects like potato beetles (Lepidotarsa decemlineata Say) were the major means of transmission of bacterial wilt of potato caused by P. solanacearum. The insect transmission of potato isolate (Race 3) was refuted later (Nolla, 1931; Kelman, 1953). Bacterial wilt of Moko bananas in Trinidad (Rorer, 1911) and Bluggoe bananas in Honduras (Buddenhagen and Elsasser, 1962), however, has been found to be transmitted by insects. Das and Chattopadhyay (1955) reported that in West Bengal, wilt of eggplant occurred when the plants came to flower and began to fruits and not on young plants. The results suggest the possibility of insect transmission of the pathogen (Buddenhagen and Kelman, 1964). The potential of insect spread of the pathogen on solanaceous hosts including the weed-hosts warrants reinvestigation.

Survival on Seed. — The possibility of survival of P. solanacearum in seeds and transmission of the pathogen through

seeds has been claimed by many and refuted by others. Sherbakoff (1917), Weber and Ramsey (1926), and Van Haltern (1935) claimed that the pathogen may be introduced in a field by sowing infected tomato seeds. Similar claims were made by other workers (Aberdeen, 1945, 1949). Palm (1922) provided evidence that the pathogen was transmitted in peanut seeds. The bacterium could be isolated from the funiculus and the integuments, but not from the embryo. Nakata (1927), however, reported rapid loss of viability of the bacterium on dry tobacco seeds. Moreover, Labrousse (1932) detected no evidence of infection in 500 tomato seedlings planted with seeds obtained from fruits of diseased plants. Similar results have been reported by Shekhawat et al. (1979). These authors found that seeds obtained from infected tomatoes and Chilli plants produced neither diseased plants nor symptomless carriers.

It is now well established that several bacterial phytopathogens are transmitted by seeds (Carter, 1962; Baker and Smith, 1966; Baker, 1972; Schuster and Coyne, 1974). It is also known that a few pathogens such as Xanthomonas campestris and X. incanae infected the tomato seeds through the vascular elements (Schuster and Coyne, 1974). The tomato-canker pathogen Corynebacterium michiganense invades the tomato fruit via the vascular elements, but seed contamination results during extraction of seeds from the fruit (Schuster and Coyne, 1974). Apparently C. michiganense is externally seed borne on tomato.

Recently Ramadevi and Menon (1980) reported that P. solanacearum was carried externally on tomato seeds. The incidence of

the pathogen on freshly extracted seeds and pulp of fruits obtained from infected plants was high. Drying or surface sterilization of the seeds reduced the incidence of the pathogen significantly. These authors could not detect the pathogen in embryo of the seeds. Seed transmission of the pathogen was also recorded by Moffett et al. (1981). These authors demonstrated transmission of P. solanacearum from contaminated seeds to the cotyledons of capsicum and tomato at high (60-92%) relative humidity of the ambient atmosphere. The possibility of seed infection by P. solanacearum and dispersal of the pathogen through seeds of other solanaceous hosts including eggplant needs thorough investigation.

Survival in seed tubers. — Smith (1896) recognized that P. solanacearum could spread from season to season through infected potato tubers. This was confirmed by several authors later (Orton, 1913; Meier and Link, 1923; LeClerg, 1942). In India, the possibility of spreading the pathogen by infected seed tuber potatoes has been pointed out by several early workers (Coleman, 1909; Mann and Nagpurkar, 1919, 1921). Certified disease free seed tubers has minimised the occurrence of the disease in most advanced countries but similar facilities are not available in most developing countries. Dispersal of the pathogen in potato tubers and rhizomes of bananas and ginger to areas nearby or to distant countries has occurred (Anonymous, 1960; Buddenhagen, 1961). Spread of the potato strain (Race 3) of the pathogen through infected seed tubers has been found to be quite important in Africa, Australia and South America (Thurston, 1963; Harrison, 1961).

Long term survival of race 3 in latently infected self sown tubers has been reported in cool temperate climate of Northern part of New South Wales (Graham et al., 1979). Shakhawat et al. (1979) reported that in India apparently healthy potato tubers from summer as well as autumn crops in the hills carry the pathogen and develop the disease when planted next year.

Survival in Soil. — There is much evidence to show that many phytopathogenic bacteria survive from season to season in soil (Buddenhagen, 1965; Schuster and Coyne, 1974; Leben, 1981). Phytopathogenic bacteria in relation to their soil phase have been categorised into 3 groups : transient visitors, resident visitors, and residents (Buddenhagen, 1965). In the first group the soil phase of the organism declines rapidly and not contributing to the perpetuation of the pathogens. Most phytopathogenic bacteria fall in this category (Schuster and Coyne, 1974). The second group is characterized by organisms whose numbers gradually decline in soil. Their long-term occurrence in soil is host-dependent, and their populations increase or gradually decrease according to cropping practices. Pathogens with an extended soil phase include Agrobacterium tumefaciens and P. solanacearum. The third group is characterized by bacteria that reproduce in soil or on rhizoplane and whose relation to plant disease is erratic and transitory. These include the true soil saprophytes and green fluorescent pseudomonas spp.

It is now well established that many isolates of P. solanacearum can survive in soil for a long time (Buddenhagen, 1965; Schuster and Coyne, 1974; Kelman, 1976). Smith (1944) showed that race 1 of the pathogen survived in bare-fallowed field

plots for at least 4 years. Roque (1933) found that the pathogen survived upto 10 years in soils cropped with non-susceptible plants. The disease caused by race 1 has been reported to occur in the 1st planting of a susceptible crop on virgin land (Schwarz, 1926) and this has been attributed to the presence of weed hosts carrying the pathogen on the roots (Sequeira, 1962). Apparently, different strains of the wilt bacterium vary in their soil survival capacity. The race 1, however, can be considered a true soil pathogen and is one of the most successful pathogens in this regard (Schuster and Coyne, 1974).

Despite the above evidence, poor survival ability of race 1 in soil has been reported by many authors (Dukes et al., 1965; Tanaka and Noda, 1973). Several workers have reported erratic distribution and unpredictable persistence of race 1 in field soils under natural conditions (Kelman, 1953; McCarter et al., 1969). Kelman (1953), noted that race 1 can survive in some soils for long periods in the absence of susceptible crops, whereas in other soils it declined rapidly even with the cultivation of susceptible hosts (Buddenhagen and Kelman, 1964; Dukes et al., 1965). Furthermore, strains of a particular race may vary in their soil survival capacity (Sequeira, 1962).

In a study of the survival of race 3 (Potato strain) of the pathogen, Graham et al. (1979) found that the pathogen could be detected in soil and debris collected from a heavily infested potato field 33 weeks after the field area was abandoned. Infested debris that was mixed into non-infested soil

remained infested for 26 to 32 weeks. The extent of survival of race 3 in soil free of host debris, however, was not investigated. Moffett et al. (1983) reported prolonged survival of biovars 2 (race 3) and 3 (race 1) in clay and sandy soils in the absence of hosts. Apparently, both races 1 and 3 are capable of surviving in soil as resident visitors (Buddenhagen, 1965).

Rangaswami and Thirunavukarasu (1964) reported that the eggplant isolate can survive in sterile and non-sterile soils for more than 250 days (less than one year) when introduced artificially as a water suspension. The population in non-sterile soils, however, declined more rapidly than in sterile soils. In contrast, Rao et al. (1959) were of the opinion that the wilt pathogen of the solanaceous vegetables can not survive in soil. Positive survival of the potato isolate of P. solanacearum in the central plateau of Madhya Pradesh, Karnataka and eastern plains has been reported (Keshwal, 1976; Shekhawat et al., 1978). Shekawat et al. (1978) showed that the pathogen can survive for at least 2 years in north-western hills, Deccan plateau and central and eastern plains of India in a variety of soil types having strongly acidic to alkaline reactions. In the survival study of eggplant isolate in artificially inoculated soils using brinjal seedlings as indicators, it was observed that the pathogen can not survive for more than 16 weeks (Nayer and Mathew, 1982). Apparently, information with respect to the survival of the eggplant isolate in soil is conflicting and warrants further investigation.

Various soil factors such as soil types, soil texture,

temperature, moisture, pH, fertilizer amendments are known to affect survival of race 1 in soil (Buddenhagen, 1965; Cook and Papendick, 1974; Keshwal, 1976). Furthermore, Rangaswami and Thirunavukarasu (1964) have shown that the survival of the pathogen in soil is dependent on initial inoculum concentration added to the soil.

Conflicting evidence has been presented regarding the effect of soil moisture on the survival of the pathogen. Survival is known to be affected by soil desiccation and flooding (Buddenhagen and Kelman, 1964; Govindarajan and Gnanamanickam, 1980). Buddenhagen and Kelman (1964) reported that dry soil conditions reduced survival and sporadic drying resulted in a decrease in viability. Ramos (1968) investigating the survival of P. solanacearum in sterile soil at different moisture levels found that the populations of the pathogen declined at moisture levels below field wilt point. In a water saturated soil column he found that populations of races 1 and 3 of P. solanacearum initially increased, but later race 3 declined at a greater rate than race 1 (Ramos, 1976). Field experiments indicated that survival was enhanced under wet conditions. Tanaka and Noda (1973) found that the growth rate of P. solanacearum in sterile soil was higher at high (80-100% water content) soil moisture than in soil with 40% water holding capacity. However, such increases did not occur in non-sterilized soil regardless of water content. Contrasting results were obtained by Okabe (1971), who found that the organisms grew more actively in soil with 15-20% water content than in moist soil with 40-50% water content. In a

comparative study of the survival of races 1 and 3 in soils moistened at a constant pressure potential, Moffett et al. (1983) observed that the population of both the races declined with increasing pressure potentials of the soil and the population of race 3 declined more rapidly than race 1. Moreover, there was also a tendency of better survival of both the races in the drier soil treatments. Furthermore, a definite effect of the soil type has been recorded by these authors. The rate of population decline of both the races was greater in clay loam than in sandy loam soils. Nesmith and Jenkins (1983) reported that in certain soils of North Carolina the pathogen survived well, whereas in other soils were suppressive. Soil moisture levels and antagonism by soil microorganisms were found to be major reasons for poor survival of race 1 in the suppressive soils (Nesmith and Jenkins, 1985). These authors provided evidence that soil moisture expressed as matric potential of the soil significantly affected survival of race 1 in unsterile soils, regardless of soil types. The most favourable soil moisture for survival of race 1 was -0.5 to -1 bars. Flooded or dried (-5 to -15 bars) soils were not conducive for survival.

Excess soil moisture has an adverse effect on survival of P. solanacearum. Although in various parts of the world it has been a generally accepted fact that high soil moisture levels due to heavy rainfall usually favour development of disease (Kelman, 1953). Van Schreven (1948) provided definitive evidence that flooding the soil could effectively eliminate the tobacco isolate in Java.

There are many reports of increased bacterial wilt incidence during periods of hot weather and the earlier literature has been reviewed (Kelman, 1953). Several controlled experiments have shown that disease development following inoculation is more rapid with increasing soil temperature from 26 to 37.8 C (Vaughan, 1944; Gallegly and Walker, 1949). The optimum soil temperature for the pathogen and the disease was in the range of 28 to 32 C (Gallegly and Walker, 1949). Tanaka (1976) reported that the tobacco isolate of the pathogen survived for less than 5 days at 30 C when air-dried on glass slide. In water saturated and heavily fertilized volcanic ash soil (pH 6.1) kept at different temperature conditions, prolonged survival occurred at temperature 20 C or more, but from 30 C or more the survival was significantly less (Tanaka, 1976). In non-sterilized soil, the organisms survived poorly, under identical condition of soil manuring and temperature regimes.

It is apparent from the above discussion that results concerned with the survival ecology of the pathogen in soil and especially the effects of soil temperatures, soil moistures and soil treatments on the persistence and epiphytotics caused by the pathogen are highly variable. A survey of literature indicates that most of the studies on survival of the pathogen in soils have been indirect, based on disease incidence of susceptible plants sown or population count using selective media (Buddenhagen and Kelman, 1964; Buddenhagen, 1965; Karganille and Buddenhagen, 1972). The disease incidence data is difficult to interpret in terms of effects upon the pathogen itself or the host. Although several selective media were developed, none was

thought to be satisfactory by Nesmith and Jenkins (1976). Selective media so far developed for P. solanacearum can not eliminate all soil bacteria and the different races of the pathogen show differential tolerance or sensitivity to them with consequent erroneous quantification of the population in soil. In recent years, use of antibiotic resistant marker strains for studying the population dynamics of introduced bacteria in soil have received increased attention (Obaton, 1971; Schwinghamer and Dudman, 1973; Alexander, 1978, 1981). The method appears to be easier and quicker than other methods (Alexander, 1981). The effects of various soil factors on the actual growth and survival of the pathogen in soil environment can be tested by this method. Recently Moffett et al. (1983) has used mutant strains of P. solanacearum biovar 2 (race 3) and 3 (race 1) resistant to 2 mg streptomycin sulphate ml⁻¹ for determining the effects of soil moisture and soil types on their survival. Information with respect to growth and survival of marker strains of different races of P. solanacearum as affected by diverse soil factors is insufficient.

Varietal resistance. — It is generally accepted that the use of resistant varieties is the cheapest, safest and the most effective method of controlling the wilt of eggplant. Screening of cultivars for resistance to diseases is a pre-requisite for development of resistant cultivars. Screening of eggplant varieties for resistance to wilt has been made in several countries and a few resistant germplasms have been identified in Puerto Rico (Nolla, 1931; Roque, 1941), the Philippines

(Anonymous, 1962; Empig et al., 1962), Ceylon (Park and Fernando, 1940), South Africa (Wager, 1944, 1946). Japan (Kuneida, 1953; Suzuki et al., 1964), Martinique (Daly, 1972), USA (Winstead and Kelman, 1960; Henderson and Jenkins, 1972), India (Rao and Tikeo, 1975; Rao et al., 1976; Sitaramaiah et al., 1981, 1984; Sheela et al., 1984), and Malayasia (Mak and Vijiarungam, 1980).

In the United States of America, the cultivars Matale and Kopek were found to be highly resistant, but Florida Market, Florida High Bush and Fort Myers Market although commercially superior and resistant to Phomopsis blight were found to be intermediate in resistance to wilt (Winstead and Kelman, 1960). In the Philippines, the cultivars Aroman and Sinampiro (Empig et al., 1962) and in Malaysia (Mak and Vijiarungam, 1980) 9 local varieties have been found to be resistant to the disease. Rao et al. (1976) tested three Philippino (Dingras Multiple Purple, Dumagnete Long Purple and Sinampiro), one Javanese (Kopek), one Ceylonese (Matale) and 14 Indian varieties for wilt resistance under greenhouse and field conditions of Bangalore, India. The varieties Dingras Multiple Purple and Sinampiro from Philippines and Pusa Purple Cluster from India were found resistant; Kopek and Dumagnete Long Purple were moderately resistant; and all other exotic and Indian cultivars were rated as highly susceptible. Mak and Vijiarungam (1980) confirmed the resistance of Pusa Purple Cluster but not of Pusa Kranti in a trial in Malaysia. In contrast in a sick soil trial of 12 Indian commercial varieties [Pusa Purple Long, Gulla, SM 2, Purple Round, Black Beauty, Black Long, Black Beauty - 2, Improved Muktakeshi, Round Black, Round

White Chinese, Pusa Purple Cluster and the wild species S. torvum]

Siddarama Gowda et al. (1974) reported resistance in the local variety Gulla and the wild plant S. torvum. Earlier Srinivasan et al. (1969) have shown that only the wild brinjal S. melongena var. insanum was resistant to wilt; the local popular commercial varieties Purple Long Datta was highly susceptible under Kerala condition.

Conflicting results were obtained by Khan (1974) in a separate experiment using wilt sick plots in Bangalore. Several eggplant varieties such as Long Purple, Udipi, Improved Muktakeshi, Purple Long, Pusa Purple Cluster and wild species S. torvum and S. Xanthocarpum were found to be resistant to wilt pathogen. Many of the varieties claimed to be resistant by Khan (1974) were reported as susceptible by earlier works. Apparently, the inoculum potential of the wilt sick plots used by different authors was variable with consequent variation in disease incidence and wilt rating. Gopimony and George (1979) compared the wilt tolerance of Pusa Purple Cluster with local varieties of eggplant and two wild eggplant species. The variety Pusa Purple Cluster which was reported to be resistant earlier (Khan, 1974; Rao et al., 1976; Mak and Vijiarungam, 1980) was found to be susceptible under Kerala conditions. Cultivars Velleyani and Annamalai and both the wild species showed resistance, but all these produce small fruits and the yield of the local varieties were quite low. Sitaramaiah et al. (1981) reported that most of the lines reported resistant to wilt in India viz., Dingras Multiple Purple, Sinampiro, Pusa Purple Cluster, Pusa Purple Round, Vijay Hybrid

and Banaras Giant Green wilted under the warm humid climate of Kerala. In contrast, in a field trial at Pantnagar of Northern India, Pusa Purple Round, T-3, Vijay Hybrid, Pusa Purple Cluster, Banaras Giant Green, S-3 and six other breeding lines were found to be resistant to race 3 of the pathogen (Sitaramaiah et al., 1984).

Sheela et al. (1984) evaluated the resistance of thirty-four breeding lines of eggplant including SM 6, a resistant line earlier selected by Kerala Agricultural University (1980) and 4 other species of *Solanum* viz., *S. indicum*, *S. macrocarpon*, *S. integrifolium* and *S. sisymbirifolium*. This work confirmed the presence of resistance in SM 6 and Annamalai along with other resistant breeding lines such as SM 48, SM 56, SM 71, SM 72 and SM 74. Moreover, a prickly line SM 6-1 with long purple fruits, was found immune to wilt. Transfer of bacterial wilt resistance from the wild eggplant species *S. insanum* to cultivated variety has been demonstrated by Gopimony and Srinivasan (1970) and confirmed by Swaminathan and Srinivasan (1971).

Recently Das and Panda (1985) investigated the field reaction of 60 brinjal germplasms to bacterial wilt in Orissa in wilt sick plots. Out of sixty varieties, Berhampuri Pendi (5.8%), Bhaujanagar Local (8.8%), Purusothampur Local (9.3%) and Cuttack Pendi Round (6.2%) were found to be resistant, Malipada Local was moderately resistant and the rest 55 varieties including Pusa Purple Cluster were highly susceptible. These authors, however, have not mentioned about the race of pathogen involved in their study. In West Bengal Pusa Purple Long has been found to be highly susceptible (72%) to the local isolate of *P. solanacearum*, whereas Pusa Purple Cluster (12%), Improved Muktakeshi (28%), and Pusa

Kranti (36%) were found to be moderately resistant (Mukherjee and Mukhopadhyay, 1982). Apparently, the eggplant varieties exhibit differential response to wilt pathogen under different agroclimatic conditions of the experimental site. The differential response of a particular variety in two locations might be due to difference in the virulence of the race of isolate of the pathogen. The wilt response of large number of eggplant germ-plasms against race 1 and race 3 of Pseudomonas solanacearum under diverse agroclimatic condition of India warrants investigation.

Influence of fertilizers on wilt incidence. — The relationship of host nutrition especially nitrogen nutrition and disease severity has been studied in many host-parasite interacting system and the results have been reviewed (McNew, 1953; Barbar, 1968; Huber and Watson, 1972; 1974). There are several reports of disease-increase due to nitrogen fertilization of the field. The form of nitrogen also influences the severity of many diseases (Huber and Watson, 1974). Inorganic mineral nutrients appear to influence disease potential and the source of nitrogen appears more important than the rate (Baker and Martinson, 1970). Organic amendments may decrease disease even though the population of the pathogen is increased (Nash and Snyder, 1962). Information with respect to host nutrition and incidence of wilt of solanaceous crops caused by race 1 are insufficient.

Application of high rates of nitrogen, potassium and phosphorus, caused significant reduction of wilt incidence in White

Stem tobacco variety susceptible to P. solanacearum. Similar doses of N, P or K did not influence the reaction of the resistant variety 79-A (N.C.Agr. Expt. Sta., 1941). In greenhouse experiments, Gallegly and Walker (1949) found that wilt severity of Bonny Best tomato plants was inversely related to nutrient concentrations supplied as balanced Hoagland solution. These authors further observed that in summer months disease indices were usually decreased in the high-N series and increased in the low-K series. The reverse was found in winter months. Different levels of phosphorus did not have any effect on the disease severity irrespective of summer or winter months.

Both the form of nitrogen and the concentrations of nitrogen were found to affect wilt development in tobacco and tomato plants (Kelman, 1950, 1953; Huber and Watson, 1974). It has been reported that high concentration of nitrate-nitrogen (NO_3) caused increase (Gallegly and Walker, 1949) and ammonia-nitrogen (NH_4) caused decrease (Smith, 1944) in wilt incidence of tobacco and tomato. In contrast, Kelman (1950) observed that wilt development of tomato plants was most rapid in plants supplied with nitrogen in the ratio of ammonium-N to nitrate-N of 3:1 and least rapid in plants supplied with nitrate-N alone.

Amendment of soil with high rates of urea has been found to cause significant reduction of bacterial wilt of tobacco (Smith, 1944). Urea applied at the rate of 100 lbs per acre reduced 54% wilt in tobacco (Smith, 1944; N.C.Agr. Expt. Sta., 1941). Little or no wilt appeared in tomato, potato, eggplant,

chilli, castor bean, petunia and black night shade plants treated with urea at the rate of 1000 lbs per acre 10 weeks prior to planting (Smith and Clayton, 1943; N.C.Agr.Expt.Sta., 1943 a, b). Application of 1,000 to 2,000 lbs of urea per acre caused economic control of wilt of tobacco in Florida (Kincaid, 1950). Treatment of soil with urea in North Carolina, however, was not as effective in reducing disease on potatoes as on tobacco (Nielson and Todd, 1945). Control of bacterial wilt of eggplant by application of urea as nitrogen fertilization has been reported in recent years (Clairon, 1984). Application of urea at 700 units/ha be applied before planting, the remaining dose (400-500 units/ha) should be applied as split doses every 3-4 weeks until the end of the crop cycle.

In an evaluation of different sources of phosphate on incidence of tomato wilt, it was reported that low grade superphosphate reduced infection; high grade superphosphate and basic slag were less effective; di-calcium phosphate and ground rock-phosphate produced no appreciable reduction in disease severity (Van der Poel, 1937, 1938).

Several authors have investigated the agronomic response of eggplant to different levels of nitrogen, phosphorus and potassium. In general, it was found that all growth characters and fruit yield showed increasing effect with increasing doses of nitrogen. Increased levels of phosphorus and potassium had no positive correlation with yield (Patnaik and Farooqui, 1964; Martia and Chauhan, 1970; Gnanakumari and Satyanarayana, 1971; Usmani and Khot, 1973; Rathore

and Dayal, 1974; Verma et al., 1974).

Influence of organic manures on wilt incidence. — There are several reports of influence of organic manures, such as, oilcakes, seedmeal, composts and crop residues on the incidence of diseases caused by soil-borne phytopathogens (Patrick and Toussoun, 1965; Hoitink and Fahy, 1986). Application of high doses of organic materials to soils is known to cause decrease in wilt, caused by P. solanacearum (Van der Poel, 1938; Celino and Gottlieb, 1952; Tanaka, 1976). Van der Poel (1938) first reported that peanut meal was very effective and reduced wilt incidence of tomato by 50%. With tobacco as test plant, wilt incidence was decreased by application of urea and peanut meal in separate plots but the decrease was the greatest in plots treated with peanut meal. In contrast, no significant reduction in brown rot of potato was observed due to application of peanut or, copra meal to soil (Van Eek and Thung, 1950).

Jayaprakash and Rajan (1975) reported that non-edible seed-meals such as rubber seedmeal and neemcake in combination with inorganic fertilizer and pesticide caused significant reduction in the percentage of wilted plants of tomato and population of P. solanacearum in soil, but remarkable increase in soil saprophytes. Similar results were obtained both in pot and field experiments. There are several reports that amendment of soil with neemcake suppressed the population of nematodes, some fungi and many insect pests of eggplant (Dryer, 1987; Singh et al., 1987). Bhattacharya and Goswami (1987) found that application of

powdered neemcake to soil at a concentration of 4% caused significant reduction of nematode population which indirectly adversely affected P. solanacearum in soil with consequent decrease in wilt incidence.

Mitra and Mukherjee (1988) studied the effects of peanut meal, mustardcake, neemcake and mahucake on the profiferation of bacteria, fungi, actinomycetes and ammonifying bacteria in soil. These authors reported that peanut meal and mustardcake supported more number of fungi, bacteria and actinomycetes than neem or mahua cakes. The stimulation of bacteria and fungi was found to be the least with neemcake, and mahuacake caused least increase of fungi and ammonifying bacteria. Furthermore, neemcake is now widely used in many parts of India to promote the growth of tomatoes, cabbages, cauliflowers and potatoes. In the experimental plots of International Rice Research Institute (IRRI) at the Philippines, neemcake caused a boost to crop yield by upto 1.3 tons/ha (Singh et al., 1987). Neemcake treated with urea was 20% more effective than untreated urea for rice production (Reddy and Prasad, 1975). There is no work on the efficacy of edible and non-edible oilcakes in controlling the bacterial wilt of eggplant caused by race 1 of Pseudomonas solaracearum. The agronomic response of eggplant to soil amendment by various oilcakes also needs reinvestigation.

Disease control by irrigation. — The favourable and unfavourable effects of irrigation and soil moisture on disease development have long been known and the subject has been reviewed (Chupp, 1946; Cook and Papendick, 1971, 1972; Cook et al., 1972;

Griffin, 1963, 1969; Rotem and Palti, 1969). It is now well established that most root diseases including wilt caused by P. solanacearum are favoured by wet soils (Kelman, 1953; Cook and Papendick, 1972). The development of the pathogen under different irrigation management, and its effect on disease, has been studied by several authors and the information has been summarised (Kelman, 1953). Several early workers suggested that reduction of soil moisture to low levels could effectively control the wilt of tobacco in Sumatra (Palm, 1926) and Japan (Nakata, 1927). Drying out of soils in seedbeds following treatment with soil chemicals gave good control of wilt in Java (Thung, 1939). Hayward (1964) in a study of the characteristics of different isolates of P. solanacearum observed that wilt incidence increased with the water holding capacity of the soil. There are several reports that incidence of disease was the highest during the dry spells followed by heavy rainfall (Nolla, 1931; Seneviratne 1969; Nayar and Mathew, 1982). Stolp and Starr (1963), however, reported that the pathogen survived poorly in very moist soils, i.e. soils saturated with moisture and having poor drainage facilities. Poor drainage of soil has been long known to be conducive for the disease (Earle, 1900; Smith, 1914; Thomas, 1939). Similar conclusions were made by several Indian workers (Rao and Sohi, 1977; Nayar and Mathew, 1982).

Following the early report by Van Schreven (1954) that flooding of soil could effectively control P. solanacearum in Java, several authors have reported similar results. Tanaka and Noda (1973) observed rapid decline of the wilt bacterium

in flooded soils in Japan. Govindarajan and Gnanamanickam (1980) reported that moko wilt pathogen failed to survive in flooded soils or soils held at -15 bars. Apparently, both extreme drought and wetness, were unfavourable for the disease. Control of tobacco wilt by flooding of soil has been confirmed in recent years (Abedin et al., 1983; Iqbal and Kumar, 1985). The incidence of wilt in artificially inoculated tobacco plants was brought down from 44.7% to practically zero by flooding the soils for six months before planting.

There are several reports that high soil moisture conditions are agronomically suitable for good growth of eggplant and fruit yield (Balasa et al., 1970; Abdel-Fattah and Abdel-Salam, 1972; Hafeez and Cornillon, 1976; Cornillon and Dauple, 1981; Subramanyam and Gupta, 1982). An increase in frequency of irrigation is known to cause better plant growth, i.e. plant height, fruit weight, early flowering and total yield (Hafeez and Cornillon, 1976). It is now well established that moisture levels equivalent to 80% field capacity are favourable for high yield of fruit by eggplant (Balasa et al., 1970; Cornillon and Dauple, 1981).

Chemical Control. — Chemical control of bacterial wilt by disinfecting soil with toxic chemicals have been attempted by several authors and the earlier literature has been reviewed (Kelman, 1953). Chemicals such as chloropicrin (Smith, 1944), and ammonium thiocyanate (Nielson and Todd, 1945) caused effective control of the disease, but were highly expensive. Several fungicides such as copper carbonate, Bordeaux mixture, potassium sulfide, corrosive sublimate, copper acetate, copper

sulfate, and formalin gave poor control of the disease and most caused severe injury to the tobacco crop (Stevens, 1906; Kelman, 1953). The soil fumigant D-D (dichloropropane-dichloropropene mixture) caused significant control of Granville wilt of tobacco, but the plants showed high degree of chlorine injury (Smith, 1947).

Winstead and Kelman (1960) evaluated various antibiotics like aureomycin, streptomycin and achromycin, and chemicals like vapam, formalin, and bleaching powder for the control of bacterial wilt of potato. The final disease incidence ratings were 83% in control, 70% in streptomycin, 66% in vapam, 62% in bleaching powder and 12% in formalin treated plots. Perez (1961) reported that the tomato isolate of P. solanacearum could be controlled by two additions of 4 μ g/ml streptomycin sulfate to the nutrient solution at an interval of 7 days. It was further reported that slightly wilted plants recovered after the application of streptomycin and no further infection took place.

El-Goorani et al. (1978) conducted in vitro experiments on the efficacy of various fungicides, insecticides and antibiotics against race 3 isolates of P. solanacearum collected from various parts of Egypt. Growth of the bacterium was found to be inhibited by ampicillin, kanamycin, oxytetracycline, tetracycline, streptomycin, nabam (Dithane A-40), maneb (Dithane M-22), Zn-ion, maneb complex (Dithane M-45), and the insecticide chlorothion. Systemic fungicides like carbendazim and benomyl proved ineffective in inhibiting the growth in vitro. Drenching the soil with Dithane M-22 (0.25%; wt/vol.), Dithane M-45 (0.25%; wt./vol.) or chlorothion (50 μ g/ml, active ingredient) decreased the

incidence of potato wilt induced by artificial infestation of the field with the pathogen. Mondal and Mukherjee (1978) were able to protect tomato plants against the pathogen by using streptomycin sulfate at the rate of 500 ppm. Seedling treatment of eggplant with streptomycin (1 g/40 l) for 30 minutes before planting reduced the disease incidence (Dutta and Verma, 1969). Titatarn (1985) further reported that the ginger isolate (biotype IV) which is distinctly different from other isolates was also sensitive to streptomycin.

Newer antibiotics like myomycin C was more effective than streptomycin in inhibiting growth of the pathogen in vitro. Its protective value in field was higher than that of streptomycin (Katasuyoshi et al., 1978).

In laboratory tests, captan, maneb, mancozeb and thiram at the rate of 1000 and 10,000 ppm showed significant inhibition of P. solanacearum (Leandro and Fucikovsky, 1983). In greenhouse tests maneb (1 and 10 g., a.i.), captan (10 g., a.i.) and mancozeb (5 and 10 g., a.i.) protected potato plants from infection, but the high rates of application (5 and 10 g., a.i.) per plant caused reduction of plant height and tuber production (Leandro and Fucikovsky, 1983). Under field conditions, maneb and mancozeb at the rate of 5 g., a.i. per plant caused significant reduction of disease severity index, and percentage of wilt incidence, but both the fungicides were phytotoxic and reduced the number of tubers per plant.

Re-examination of the earlier report of effective control of the disease by chloropicrin was conducted (Enfinger et al.,

1979; Balasubramanya and Patil, 1975). Chloropicrin was the only chemical of the 8 evaluated that provided full season control of the pathogen causing wilt of tomato (Enfinger et al., 1979). Similar results were found in eggplant (Balasubramanya and Patil, 1975). The phytotoxic effects of chloropicrin, however, were mentioned by these authors. Rao (1976) confirmed the efficacy of formalin in reducing the bacterial wilt (Stevens, 1906) of tomato.

Since the bacterial wilt of tomato and eggplant is more severe in fields infested with nematode, application of soil fumigants which control nematodes could significantly reduce the wilt incidence also (Bhattacharya and Goswami, 1987). In recent years, attempts have been made for integrated control of bacterial wilt by soil fumigation with Di-Trapex, soil mixing with Saroton-G and application of S 6035 (morpholinium phenylphosphinate). The combined treatments have been found to be most effective and the yield was 4 times more than that of untreated control plots (Murokoshi and Takahashi, 1984).

Since the pathogen can persist on the seeds of certain solanaceous vegetables, the efficacy of seed treatment in reducing the incidence of the disease warrants re-examination. Information with respect to control of eggplant wilt by treatment of seeds with chemicals is insufficient.

MATERIALS AND METHODS

Plant Materials

Seeds of sixtythree germplasms of eggplant were obtained from the Department of Horticulture, Bidhan Chandra Krishi Viswavidyalaya (BCKV), Mohanpur, Nadia, West Bengal and eighteen germplasms were collected by courtesy of the Director General, Bangladesh Agricultural Research Institute (BARI), Dhaka. The code number, source and fruit characteristics of the germplasms are listed in the Tables 1 and 2 respectively. Besides these, bulk seeds of commercial eggplant variety, Improved Muktakeshi and tomato varieties, namely, Marglobe Supreme and Best of All required for pathogenicity tests were obtained from Sutton Company, Calcutta.

Eggplant seedlings were raised in seedbeds (3m x 1m) which were prepared in the University experimental fields. Seeds were sown and the beds were watered at 2 days interval or adjusted according to soil moisture condition. Seedlings usually emerged after 7 days of sowing and were transplanted to the experimental plots of 3.6m x 3.0 m size with a spacing of row to row 90 cm and plant to plant 60 cm. There were a total of 20 plants per plot with 5 plants per row.

Tomato seedlings were grown under greenhouse conditions for pathogenicity tests. Tomato seeds were surface sterilized

with 0.1% HgCl_2 followed by rinsing with sterile distilled water and sown in earthenware pots containing garden soil:sand (1:1). For pathogenicity test, 7-day-old seedlings were uprooted, inoculated with the pathogen (described later) and transplanted into pots containing garden soil and sand (3:1) and incubated on greenhouse benches till symptoms developed.

Table 1. List of Indian germplasms of eggplants used

Sl. No.	Name of the germplasm	Source	Fruit characteristics: Shape, colour, surface and fruit arrangement
(1)	(2)	(3)	(4)
IG-1	Pusa Purple Long	IARI, New Delhi	Long with blunt apex; uniformly dull/reddish purple; smooth; fruits singly borne
IG-2	Banaras Giant Green	IARI, New Delhi	Big, round with blunt apex; uniformly light green; wrinkle; fruits singly borne
IG-3	Pusa Kranti	IARI, New Delhi	Long with blunt apex; deep purple, non-uniform; smooth; fruits singly borne
IG-4	Pusa Purple Cluster	IARI, New Delhi	Small with blunt apex; uniformly dark purple; wrinkle; fruits in cluster
IG-5	Improved Muktakeshi	IARI, New Delhi	Big, oblong with blunt apex; uniformly deep purple; wrinkle; fruits singly borne
IG-6	Nurki	Habra, Nadia	Long-oval with blunt apex; uniformly purple; wrinkle; fruits in cluster
IG-7	K-812	Katrain, Himachal Pradesh	Round-oval with blunt apex; uniformly purple; smooth; fruits in cluster
IG-8	Kanta Brinjal	Habra, Nadia	Oblong with tapering apex; purple, not uniform; wrinkle; fruits singly borne

Table 1. List of Indian germplasms used

(1)	(2)	(3)	(4)
IG-9	SM 65(1)	IARI, New Delhi	Long - oval with tapering apex; uniformly purple; smooth; fruits in cluster
IG-10	SM 65(2)	IARI, New Delhi	Long with tapering apex; light purple and uniform; smooth; fruits in cluster
IG-11	T-3	Kalyanpur, U.P.	Round with blunt apex; pink, not uniform; wrinkle; fruits in cluster
IG-12	KG Local	Krishnagar, Nadia	Round with blunt apex; uniformly purple; wrinkle; fruits singly borne
IG-13	SM-50(A)	IARI, New Delhi	Long with blunt apex; purple, non-uniform; wrinkle; fruits singly borne
IG-14	Long - 1	Kalpi, Nadia	Long, oval with tapering apex; purple, not uniform; wrinkle; fruits singly borne
IG-15	K-8(1)	Katraia, Himachal Pradesh	Oval with tapering apex; purple, uniform; smooth; fruits in cluster
IG-16	Pant Rithuraj	Pantnagar, U.P.	Round with blunt apex; light purple, not uniform; wrinkle; fruits singly borne
IG-17	S-62(1)	Kalyani, Nadia	Long with cone-apex; uniformly white; wrinkle; fruits singly borne
IG-18	Karimpur	Karimpur, Nadia	Medium-oval with blunt apex; greenish-white, not uniform; smooth; fruits in cluster
IG-19	Bhangar-1	Habra, 24-Pgs.	Long with tapering apex; greenish white, non-uniform; smooth; fruits singly borne
IG-20	H-4(1)	Hissar, Haryana	Oval with blunt apex; purple (White stripes at apex), non- uniform; smooth; fruits singly borne

Table 1. List of Indian germplasms of eggplant used

(1)	(2)	(3)	(4)
IG-21	SM-227	IARI, New Delhi	Oblong with blunt apex; light purple, not uniform; smooth; fruits singly borne
IG-22	SM-48	IARI, New Delhi	Oblong with tapering apex; green white stripes, not uniform; smooth; fruits singly borne
IG-23	Salimakra	Habra, 24-Parganas	Long with tapering apex; uniformly purplish green; smooth; fruits singly borne
IG-24	Kalo Chaila	Awalsidhi, 24-Parganas	Oblong with blunt apex; uniformly dull purple; smooth; fruits singly borne
IG-25	SM-71	IARI, New Delhi	Big, round with blunt apex; white, non-uniform; smooth; fruits singly borne
IG-26	Muktakeshi -I	Gadamara, 24-Parganas	Round with blunt apex; uniformly purple; wrinkle; fruits singly borne
IG-27	Garia	Haringhata, 24-Parganas	Oval with blunt apex; uniformly purple; smooth; fruits in cluster
IG-28	Type 3(3) Long	Krishnagar, Nadia	Long with blunt apex; uniformly green; wrinkle; fruits singly borne
IG-29	Gourigajar	Gadamara, 24-Parganas	Oval with blunt apex; light green with white spots, not uniform; smooth; fruits in cluster
IG-30	Pusakesar	IARI, New Delhi	Oblong with blunt apex; reddish purple, not uniform; wrinkle; fruits singly borne
IG-31	Type 3(2) Long	Krishnagar, Nadia	Long-oval with tapering apex; light green fruits, not uniform; smooth; fruits singly borne
IG-32	Green Long	Sevakgram, Burdhaman	Long with tapering apex; uniformly light green; wrinkle; fruits singly borne

Table 1. List of Indian germplasms of eggplant used

(1)	(2)	(3)	(4)
IG-21	SM-227	IARI, New Delhi	Oblong with blunt apex; light purple, not uniform; smooth; fruits singly borne
IG-22	SM-48	IARI, New Delhi	Oblong with tapering apex; green white stripes, not uniform; smooth; fruits singly borne
IG-23	Salimakra	Habra, 24-Parganas	Long with tapering apex; uniformly purplish green; smooth; fruits singly borne
IG-24	Kalo Chaila	Awalsidhi, 24-Parganas	Oblong with blunt apex; uniformly dull purple; smooth; fruits singly borne
IG-25	SM-71	IARI, New Delhi	Big, round with blunt apex; white, non-uniform; smooth; fruits singly borne
IG-26	Muktakeshi -I	Gadamara, 24-Parganas	Round with blunt apex; uniformly purple; wrinkle; fruits singly borne
IG-27	Garia	Haringhata, 24-Parganas	Oval with blunt apex; uniformly purple; smooth; fruits in cluster
IG-28	Type 3(3) Long	Krishnagar, Nadia	Long with blunt apex; uniformly green; wrinkle; fruits singly borne
IG-29	Gourigajar	Gadamara, 24-Parganas	Oval with blunt apex; light green with white spots, not uniform; smooth; fruits in cluster
IG-30	Pusakesar	IARI, New Delhi	Oblong with blunt apex; reddish purple, not uniform; wrinkle; fruits singly borne
IG-31	Type 3(2) Long	Krishnagar, Nadia	Long-oval with tapering apex; light green fruits, not uniform; smooth; fruits singly borne
IG-32	Green Long	Sevakgram, Burdhaman	Long with tapering apex; uniformly light green; wrinkle; fruits singly borne

(1)	(2)	(3)	(4)
IG-33	Muktakeshi -II	Mondouri, Nadia	Oblong with blunt apex; green- ish purple, not uniform; smooth; fruits singly borne
IG-34	Black Round	Krishnagar, Nadia	Long, oblong with blunt apex; purple with green stripes, not uniform; wrinkle; fruits singly borne
IG-35	Katamakra	Habra, 24-Parganas	Round-oval with blunt apex; uniformly greenish purple; wrinkle; fruits singly borne
IG-36	Silimum	Kalyani, Nadia	Oblong with blunt apex; light green with green stripes, not uniform; wrinkle; fruits singly borne
IG-37	PPR-7	Pantnagar, U.P.	Long with tapering apex; light purple, not uniform; smooth; fruits singly borne
IG-38	B-5	Bankura, West Bengal	Oblong with blunt apex; light purple, not uniform; wrinkle; fruits singly borne
IG-39	Guzrat	Guzrat	Oval with blunt apex; purple, not uniform; wrinkle; fruits singly borne
IG-40	Long White	Nadia, West Bengal	Long, thick with tapering apex; uniformly light green; smooth; fruits singly borne
IG-41	Mysore	Mysore, Karnataka	Long with tapering apex; uniform- ly purple; wrinkle; fruits in cluster.
IG-42	Siliguri	Jalpaiguri, West Bengal	Long with tapering apex; uni- formly deep purple; wrinkle; fruits singly borne
IG-43	K-3	Katrai, Himachal Pradesh	Round oval with blunt apex; uniformly green; smooth; fruits singly borne
IG-44	Azad Kranti	Kalyanpur, U.P.	Long-oblong with blunt apex; uniformly purple; wrinkle; fruits singly borne

(1)	(2)	(3)	(4)
IG-45	Dharampur- Local	Dharampur, 24-Parganas	Oblong with blunt apex; green with purple stripes, non-uniform; smooth; fruits singly borne
IG-46	K-2	Katrain, H.P.	Oblong with blunt apex; uniformly light green; smooth; fruits in cluster
IG-47	SM-31	IARI, New Delhi	Big, oblong with blunt apex; uniformly white; wrinkle; fruits singly borne
IG-48	SM-273	IARI, New Delhi	Oval with blunt apex; uniformly whitish green; smooth; fruits singly borne
IG-49	Bhangar-II	Awaisidhi, 24-Parganas	Oblong with blunt apex; light green with white stripes, not uniform; smooth; fruits in cluster
IG-50	White Brinjal	Burdhaman, W.B.	Long, slender with tapering apex; uniformly white; smooth; fruits singly borne
IG-51	Paichabani	Gadamara, 24-Parganas	Small, oval with blunt apex; light green, not uniform; smooth; fruits in cluster
IG-52	KNG-1	Krishnagar, Nadia	Small; round with blunt apex; uniformly purplish green; smooth; fruits in cluster
IG-53	Pant Samrat	Pantnagar, U.P.	Oval with cone apex; uniformly purple; smooth; fruits singly borne
IG-54	H-8	Hissar, Haryana	Small, round with blunt apex; greenish white, not uniform; smooth; fruits in cluster
IG-55	Makra	Habra, 24-Parganas	Big, round with blunt apex; uniformly green; smooth; fruits singly borne

(1)	(2)	(3)	(4)
IG-56	Manjorigota	Rahuri, Maharashtra	Small, round with blunt apex; white with pink stripes, non-uniform; smooth; fruits in cluster
IG-57	Purulia Purple	Purulia, W.B.	Oblong with cone apex; uniformly purple; smooth; fruits singly borne
IG-58	PBR-91-2	Pantnagar, U.P.	Small, round with blunt apex; purple, not uniform; smooth; fruits in cluster
IG-59	Muktakeshi- III	Chandemari, Nadia	Big size, oblong with blunt apex; uniformly deep purple; wrinkle, fruits in cluster
IG-60	KNG-II	Krishnagar, Nadia	Small, round with blunt apex; uniformly greenish purple; smooth; fruits singly borne
IG-61	KT-4	Katraia, H.P.	Long - oval with cone apex; uniformly light purple; smooth; fruits singly borne
IG-62	S-54	Kalyani, Nadia	Round-oval, blunt apex, light purple with white stripes, not uniform; smooth; fruits in cluster
IG-63	Patakanta	Habra, 24-Parganas	Round with blunt apex; uniformly whitish purple; smooth; fruits singly borne.

Table 2. List of Bangladeshi germplasms of egg plant used

Sl.No.	Name of the germplasm	Fruit characteristics: shape, colour surface and fruit arrangement
(1)	(2)	(3)
BG-1	D.R. Chowdhury	Oblong with cone-apex; purple, non-uniform; wrinkle; fruits singly borne
BG-2	Black Long	Big, round with blunt apex; uniformly purple; wrinkle; fruits singly borne
BG-3	Rangpur No.1	Medium, Oval with blunt apex; purple, non-uniform; smooth; fruits in cluster
BG-4	Laljhuri	Oblong with blunt apex; reddish-purple, non-uniform; wrinkle; fruits in cluster
BG-5	Bhangar	Medium, round with blunt apex; purplish green, non-uniform; smooth; fruits singly borne
BG-6	Black ACC	Big, round with blunt apex; uniformly purple; wrinkle; fruits singly borne
BG-7	Rajsahi No.6	Medium, oval with blunt apex; uniformly white ; smooth; fruits singly borne
BG-8	Dohazari	Oblong with blunt apex; light-green, non-uniform; smooth; fruits singly borne
BG-9	Saramasi	Medium, round with blunt apex; greenish, non-uniform; wrinkle; fruits in cluster
BG-10	Rajsahi No. 9	Medium, long with tapering apex; light purple, non-uniform; smooth; fruits singly borne
BG-11	Uttara	Medium, oval with blunt apex; uniformly purple; smooth; fruits singly borne

Table 2. List of Bangladeshi germplasms of eggplant used

(1)	(2)	(3)
BG-12	Muktakeshi Round	Round with blunt apex; uniformly purple; wrinkle; fruits singly borne
BG-13	Khatkhatia Long	Long, slender with blunt apex; dull purple, non-uniform; smooth; fruits singly borne
BG-14	Longla	Long with tapering apex; uniformly bright purple; smooth; fruits singly borne
BG-15	Islampuri	Big, round with blunt apex; light purple, non-uniform; smooth; fruits singly borne
BG-16	Singhnath	Long with blunt apex; uniformly blackish purple; smooth; fruits singly borne
BG-17	Local Chittagong	Small, round with blunt apex; whitish green, non-uniform; smooth; fruits singly borne
BG-18	Sada Jhumki	Small, round with blunt apex; uniformly white; smooth; fruits in cluster.

The pathogen. — A virulent strain of Pseudomonas solanacearum was used. The organism was locally isolated from a wilted eggplant. Wilted eggplants from farmers' fields were collected and brought to the laboratory. Root portions were surface sterilized by treating with 0.1% HgCl_2 for 2 minutes followed by washing with several changes of sterilized distilled water. Surface-sterilized root portions were then placed on Potato-Dextrose Agar (PDA: peeled potato, 250 g; dextrose, 20 g; agar, 20 g; distilled water, 1ℓ; pH 7.0) in petridishes. Single well separated colonies growing near the periphery of the roots^{portions} were transferred to PDA slopes and incubated at 28 C with weekly subculturing. The identity of the organism was confirmed by determining the biochemical and pathogenicity properties.

Bulk inoculum suspension of the pathogen for field or pot inoculation experiments were prepared by growing the organism on PD broth. Erlenmeyer flasks (1ℓ) containing 400 ml of PD broth each were inoculated with the organism and incubated at 28 C without shaking. After 5 days of incubation a thick suspension of bacteria was formed and used as inoculum. The cell density per ml was determined by dilution plating technique using PDA. Unless otherwise stated the cell density of the above preparation was 10^7 - 10^8 cells/ml of suspension.

Pathogenicity test. — Infecting ability of the wild type and mutant marker isolates (described later) were determined by using tomato seedlings. Aseptically grown 15-day-old seedlings of tomato were placed in petridishes separately (5 seedlings/dish). After cutting the root-tips, the seedlings were flooded

with 10 ml cell suspension (10^8 cells/ml) of the pathogen. After 2 hours (of host-pathogen interactions), ^{data} the seedlings were sown in pots containing garden soil and sand (3:1). The control seedlings were similarly treated with sterilized distilled water. The percentages of wilt of the inoculated seedlings were determined and compared with that of the control seedlings.

Seed pathological studies. — The ^{seed} pathology studies were made following the agar plate method of Neergaard (1977). Seed samples obtained from different sources were plated on PDA (10 seeds per plate). For testing the biocidal effects of different compounds, seeds were pre-treated with the compounds and then plated on PDA. The microorganisms isolated from seed samples were maintained on PDA slopes at 28 C with monthly subculturing.

Preparation of experimental plots. — The site selected had a previous history of eggplant cultivation for 3 consecutive years and with report of fair amount of wilt incidence in each year. The experimental plots were prepared by cross ploughing and subsequent laddering. Stubbles, weeds, pieces of bricks and stones etc. were removed.

Debris of previous years' eggplant crop were not removed since these were sources of inoculum. For increasing the inoculum potentials of the field, diseased eggplants showing typical bacterial wilt symptoms were collected, chopped into pieces and added to the soil. Unless otherwise stated all the experimental fields were fertilized with N:P:K (90:60:60 kg/ha) in the form of urea, single superphosphate and muriate of potash. Full

doses of phosphorus and potassium but only half dose of nitrogen (45 kg/ha) were applied during the preparation of the experimental plots. The rest of the nitrogen, dose was applied 45 days after transplantation of the seedlings to the field.

Transplantation procedure. — Five-week old seedling of cultivar Improved Muktakeshi from seed beds were transplanted to experimental plots with different treatment schedules.

Screening for varietal resistance. — For screening the resistance of Indian and Bangladeshi germplasms, the natural disease incidence under field conditions was evaluated in the first year. The germplasms which showed resistance under natural field conditions were subjected to further tests in the following seasons by creating epiphytotic conditions by inoculation. Unless otherwise stated, the seedlings were carefully uprooted, washed thoroughly with distilled water and inoculated with distilled water and inoculated with a thick suspension of bacterial cells (10^8 cells/ml) by root clipping method. After 2 hours of interaction in the laboratory of the roots of the seedlings with the pathogen, seedlings were transferred to the respective field plots. The inoculated seedlings were reinoculated by pouring 5 ml of bacterial suspension (10^8 cells/ml) per plant employing root drenching method after 30 days of transplanting.

Amendment of field soils. — The experimental plots were amended with different doses of N:P:K or oilcakes to find out their efficacy on the incidence of bacterial wilt. Three different doses of each N, P and K in different combinations

were used. The doses were 60, 90 or 120 kg/ha for N and 40, 60 or 80 kg/ha for each of P and K. A total of 27 treatment combinations as shown in Table 3 were used.

For oilcake experiments, the plots were prepared as described before and received the basal dose of N:P:K (90:60:60 kg/ha). The oilcakes used were Karanja, Mahua, Mustard and Neem. Oilcakes purchased from local markets were dried, powdered and sieved through a 2 mm mesh sieve. The powdered cakes were mixed with the soil and moistened by irrigation. Three doses of oilcakes used were : 10, 20 or 40 quintal/ha. Control plots received the basal dose of N:P:K only.

Irrigation schedule. — The standard irrigation schedule used was watering of the plots at 10 days interval unless otherwise stated. During rainy season irrigation was given as and when necessary depending on the moisture level of the soil. For determining the effects of frequency of irrigation on the wilt incidence, the experiment was conducted during rabi (winter) season, when the moisture level of the soil in West Bengal is naturally low. Plots were prepared and fertilized with basal dose of N:P:K as described before and were irrigated every 5, 10, 15 or 20 days interval following transplantation of the seedlings.

Seed treatment. — For seed treatments, chemicals used were captan (N-trichloromethylthio-4-cyclohexene-1,2-dicarboximide), vitavax (2,3-dihydro-6-methyl-5-phenylcarbamoyl-1,4-oxathia) and streptomycin sulphate. For in vitro experiments, 5 g seeds of Improved Muktakeshi were treated separately with

Table 3. Different combinations of N:P:K used for fertilizer experiments.

$N_{60}P_{40}K_{40}$	$N_{90}P_{40}K_{40}$	$N_{120}P_{40}K_{40}$
$N_{60}P_{40}K_{60}$	$N_{90}P_{40}K_{60}$	$N_{120}P_{40}K_{60}$
$N_{60}P_{40}K_{80}$	$N_{90}P_{40}K_{80}$	$N_{120}P_{40}K_{80}$
$N_{60}P_{60}K_{40}$	$N_{90}P_{60}K_{40}$	$N_{120}P_{60}K_{40}$
$N_{60}P_{60}K_{60}$	$N_{90}P_{60}K_{60}$	$N_{120}P_{60}K_{60}$
$N_{60}P_{60}K_{80}$	$N_{90}P_{60}K_{80}$	$N_{120}P_{60}K_{80}$
$N_{60}P_{80}K_{40}$	$N_{90}P_{80}K_{40}$	$N_{120}P_{80}K_{40}$
$N_{60}P_{80}K_{60}$	$N_{90}P_{80}K_{60}$	$N_{120}P_{80}K_{60}$
$N_{60}P_{80}K_{80}$	$N_{90}P_{80}K_{80}$	$N_{120}P_{80}K_{80}$

10 ml of water or different concentrations of treatment chemicals. Seeds were treated for 15 or 30 minutes by shaking on a rotary shaker. After the treatment time, the seeds were removed from the treatment solution, plated on PDA and incubated at 28 C in the dark. The percentage of treated seeds yielding microorganisms was compared with that of control untreated seeds. For field experiments captan and vitavax were used as dry seed treatment at the rate of 0.25% and 0.40% respectively. For streptomycin application, seeds were treated with 250 ppm aqueous solution of streptomycin sulphate for 6 hours. Seeds after treatment with chemicals were sown in separate seed beds and 5-week-old seedlings were transplanted to experimental plots for natural incidence of disease. Seedlings raised from non-treated seeds served as controls.

Isolation of microorganisms from soil. — Soil microorganisms were isolated by dilution plate technique following the method of Pramer and Schmidt (1967) using rose bengal streptomycin agar medium (glucose, 10g; peptone, 5 g; K_2HPO_4 , 1 g; $MgSO_4 \cdot 7H_2O$, 0.5 g; agar, 15 g; distilled water, 1 ℓ; rose bengal, 35 mg; and streptomycin, 35 mg, (streptomycin was filter sterilized and added just before pouring the plates). Soil dilution plates were prepared and incubated at 28 C. Fungal species differing in colony morphology were transferred to PDA and maintained by incubating at 28 C with monthly subculturing.

Soil bacteria and actinomycetes were isolated using nutrient agar (NA ; beef extract, 8 g; peptone, 5 g; agar 15 g; distilled water, 1 ℓ; pH was adjusted to 6.8). Soil dilution plates were

prepared and incubated at 37 C. Colonies differing in morphology were maintained at 20 C with fortnightly transfer to fresh medium.

Isolation of Streptomycin and Erythromycin resistant strains of *Pseudomonas solanacearum*.— Mutant strains resistant to streptomycin sulphate and erythromycin were selected and isolated following the method of Obaton (1971). The step-wise development of the double marker strain is stated in Fig. 1. Thick suspensions of cells (10^8 cells/ml) of 4-day-old culture of *P. solanacearum* were prepared and streaked on PDA plates containing 600 μ g/ml of streptomycin sulphate. The survivors after multiplying on PDA slopes were streaked again on PDA plates containing 1 mg/ml of streptomycin sulphate. The surviving colonies from the latter were isolated, multiplied on PDA slopes and exposed to PDA containing 2 mg/ml streptomycin sulphate. The colonies capable of growing on the latter medium were considered as mutants resistant to 2 mg/ml of streptomycin.

For development of double marker strain of the pathogen, the streptomycin resistant (2 mg/ml) isolate was grown on PDA containing increasing concentrations of erythromycin in three steps. Erythromycin concentrations used were 200 μ g/ml, 600 μ g/ml and 1 mg/ml respectively. The colonies surviving on PDA containing 200 μ g/ml of erythromycin were successively transferred to media containing 600 μ g/ml and 1 mg/ml erythromycin. The colonies surviving on 1 mg/ml of erythromycin were considered as resistant to erythromycin.

Characterization of *Pseudomonas* isolates. — The wild type

and mutant strains of Pseudomonas were characterized by morphological, biochemical and pathogenecity properties of the isolates.

For determining morphological characters, presence or absence of flagella, capsule and spore were studied. Catalase activity, hydrolysis of starch and liquification of gelatin were determined following the method of Smith (1946). Acid and gas production by the utilization of various carbon sources were determined following the method of Graham and Parkar (1964), and Hayward (1976).

Assessment of disease incidence. — Disease incidences following different treatment schedules were ascertained at intervals by determining the following parameters : (i) Percentage of wilted plants/plot, (ii) Average height of surviving plants, (iii) Number of primary branches/surviving plant, (iv) Average length (cm) and diameter (cm) of fruit, (v) Average fruit weight (g), (vi) Yield perplant (kg), (vii) Total yield/plot (kg), and (viii) Projected yield in quintal/hectare.

Plants showing wilt or incipient wilt symptoms were subjected to "ooze test". Root segments were removed, washed thoroughly, split longitudinally and placed in a beaker containing distilled water. Within a few minutes milky-white "ooze" of bacterial mass came out of the infected parts only. Plants showing positive "ooze" test were considered as wilted plants. The percentage wilt data were transformed into angular values, following the formula $Q = \sin^{-1} \sqrt{P/100}$, where P = percentage of wilted plants (Little and Hills, 1975) and presented in parentheses. For parameter (ii) and (iii) five surviving plants were randomly selected and the number of primary

branches and the heights of the plants were recorded.

For yield data per plant (vi), all mature fruits from the plants at the time of fifth harvest were weighed and average yield in kg/plant was determined. For parameter (iv) 10 fruits were randomly selected and the average length and diameter of those fruits were determined. Average fruit weight (g) was determined by dividing the total weight of fruits by total number of fruits per plot at the time of fifth harvest. For parameter (vii) cumulative weight of the fruits harvested per plot at different times was taken. Projected yield was calculated by using the formula :

$$\text{Quintal/ha} = x - \frac{x(15)}{100} \quad \text{where } x = \frac{\text{Yield/plot} \times \text{area (ha)}}{\text{Plot area} \times 100}$$

Statistical methods. — For N:P:K trial, Randomised Block Design with 3^3 factorial concept was followed. The rest of the experiments were also done with Randomised Block Design. All the experiments were replicated 2-6 times and repeated in two successive years. The data obtained were analysed by the analysis of variance method as described by Panse and Sukhatme (1978). Two years data were averaged together and their mean values were analysed statistically. The level of significance used was $P=0.05$.

Collection and storage of soil samples.—Soil samples(Loam,pH 6.8) from a fallow land of the Kalyani University campus were collected. For collecting samples, upper 5 cm layer of soil containing the roots of grass and other herbs were discarded. Five centimeter layer of soil beneath that depth was shovelled out, kept in closed plastic bags and brought to the laboratory as quickly as possible.

Samples were air dried and sieved through a 2 mm mesh sieve. Moisture content and water holding capacity (W.H.C.) of the samples were determined following the method of Pramer and Schmidt (1967). Samples were then adjusted to 50% WHC and stored in closed plastic containers under laboratory conditions. Soils were sterilized by autoclaving at 15 p.s.i. for 1 hour on three consecutive days. Absence of growth of microorganisms in soil dilutions plated on nutrient agar ensured sterility of the soils. Non-sterilized soil was considered as natural soil. All soil samples were used within two months of collection.

Determination of moisture content and water holding capacity of soil. — Moisture content and WHC of soil samples were determined by the method of Pramer and Schmidt (1967). Approximately, 10 g of soil was taken in a weighing bottle and weighed. The bottle with the soil sample was dried at 110 C for 24 hours, cooled in a desiccator for 12 hours and weighed again. Moisture content was calculated as per cent of oven dry weight of soil.

For determination of WHC of soils, a circle of moist filter paper was placed on the brass screen of a Hilgard cup and the complete unit was weighed. The cup was filled with a test soil sample previously dried in an oven at 110 C for 24 hours. The soil was compacted by dropping the cup ten times from a height of approximately 3 cm. After levelling the top with a spatula, the unit was reweighed. The cup was then immersed overnight in a tray containing water deep enough to wet the bottom of the soil column. After the soil was saturated with water, the cup was

placed in a moist chamber and the excess water was allowed to drain out. The unit was weighed again. The amount of moisture retained per unit weight of dry soil sample was calculated and expressed as the water holding capacity of the soil (W.H.C.).

Determination of pH of soils. — The pH values of soil samples were determined following the method of Schofield and Taylor (1955). Aqueous 0.01 M CaCl_2 solution (100 ml) was added to 50 g of air dried soil, shaken thoroughly and allowed to stand for overnight. The pH of the supernatant fluid was measured without disturbing the sediments using a pH meter with glass electrode (Philips, India). Results are average of three readings for each soil sample.

Survival studies. — For survival studies, 30 g portions of air dried soils were taken in 250 ml. glass bottles. The soil samples were inoculated with required volumes of cell suspension of the mutant strain of Pseudomonas solanacearum. The inoculum was prepared by growing the mutant strain in potato dextrose broth without shaking. Cells after 24 hours of incubation were harvested, centrifuged and washed twice with sterilized aqueous salt solutions (0.02% KH_2PO_4 and 0.08% K_2HPO_4). Cells after washing were resuspended in the same salt solution and were used for inoculation. The inoculum was mixed thoroughly with the soil to get a population density of 10^8 cells per g of soil and the moisture of the soil was adjusted to 60% W.H.C. Bottles containing inoculated soils were kept in vertical position at 30 C in the dark. Soil samples from the bottles were taken out at intervals and the number of surviving

Pseudomonas cells per g of soil was determined. Soil samples were serially diluted and plated on PDA (selective medium) supplemented with streptomycin (2 mg/ml) and erythromycin (1 mg/ml) following the standard soil dilution plate technique. The plates were incubated at 28 C in the dark. After 4 or 5 days colonies growing on the selective medium were counted to obtain the number of cells surviving in unit weight of soil. Data presented in this report are based on triplicate plates for each serial dilution of soils. Replicate soil samples were taken for each treatment type.

For recovery study, different doses of cell suspensions with known number of cells of the test Pseudomonas strain were mixed thoroughly with the soil and the percentage of cells recovered at zero time was determined by soil dilution plating method.

Effect of temperature. — Temperature effects on survival of Pseudomonas cells in soil conditions were determined. Inoculated soil samples were incubated in different temperature controlled incubators. The population of surviving cells at intervals was determined.

Effect of drying. — For determining the effect of drying, soil samples were inoculated with mutant cells and allowed to air dry in petridishes kept at 30 C in the incubator in closed and open-lid conditions. The population of surviving cells was determined at indicated intervals.

Effect of soil moisture. — For determining the effect of soil moisture on survival, sterilized and non-sterilized soil samples were adjusted to different water holding capacity by adding required

volumes of sterilized distilled water. Soil samples were then mixed with cell suspensions to obtain 10^8 cells/g of soil as described previously and incubated at 30 C in the dark. The population of surviving cells was determined at indicated intervals.

For flooding experiments, soil samples were inoculated with Pseudomonas cells (10^8 cells/g) and then flooded with sterilized distilled water. The water levels above the submerged soil surface were adjusted to various depths of standing water. Flooded soils were incubated at 30 C in the dark and the number of surviving cells/g of soil were determined at indicated intervals.

Soil amendments. — Soil samples (Loam, pH 6.8) from Kalyani were amended with different supplements in separate experiments.

Fertilizer and manures used were : ammonium sulphate, potassium nitrate, urea and cowdung. Oilcakes used were linseed cake, mahua cake, neemcake and mustardcake. Organic supplements in the form of leaf residues of eggplant leaf (Solanum melongena L.) and cauliflower leaf (Brassica oleracea var. botrytis L.) were also used. Fertilizers, manures and oilcakes were purchased from local market. Leaves of eggplant and cauliflower were collected, sundried for 7 days, pulverized, sieved through 2 mm sieve and stored until used.

Required amount for obtaining a particular concentration of the individual supplement was weighed and mixed thoroughly with the soil. Soil samples thus amended with different concentrations of individual supplements were inoculated with the test marker

strain of Pseudomonas. Growth and survival of the strain in these amended soils were determined.

RESULTS

SECTION I : PRIMARY SOURCE OF INFECTION AND SURVIVAL OF THE PATHOGEN

Isolation and characterization of the pathogen. — The pathogen was isolated from the roots of wilted eggplants. Plants showing external wilt symptoms were tested for production of "ooze" in the field. Root pieces were brought to the laboratory and the pathogen was isolated following surface sterilization of the samples and plating on PDA. The pathogen was also isolated from the seed samples of eggplant obtained from the local market. All isolates were identified by comparing biochemical and (pathogene)^{pathogen} city properties. Biochemical and pathogenecity properties of a typical isolate of P. solanacearum are shown in Table 4.

It is evident from the results that the isolates showed typical morphological and biochemical characteristics of P. solanacearum. It is also apparent from biochemical characters that the organism could utilize hexose alcohols mannitol and sorbitol and other characters similar to race 1. Isolates which caused rapid wilting of susceptible Marglobe Supreme variety of tomato were selected for further studies. Of these the wild type isolate KUPL were highly virulent and used in most of the experiments.

Development of marker strain of the pathogen. — In preliminary experiments the sensitivity of P. solanacearum isolate KUPL

Table 4. Typical morphological, biochemical and pathological properties of Pseudomonas solanacearum

Test characters	Wild type isolates
Gram staining	- ^a
Spore staining	-
Presence of flagella	+ ^b
Presence of capsule	-
Hydrolysis of starch	+
Lignification of gelatin	-
Carbon sources for growth	
Glucose	+
Ribose	+
Lactose	+
Maltose	+
Fructose	+
L-Arabinose	-
Sucrose	+
Cellulose	+
Starch	+
Mannitol	+
Sorbitol	+
Malonate	-
Ethanol	-
Pathogenecity to tomato ^c seedlings	+

^a(-) indicates the absence of the characters or negative reaction to the test

^b(+) indicates the presence of the characters or positive reaction to the test

^c Seedlings of tomato variety Marglobe Supreme were used.

to streptomycin and erythromycin was compared with that of rhizosphere soil microflora of eggplant. The test organisms were plated on PDA supplemented with different doses of streptomycin sulphate or erythromycin.

It is apparent from the results that the wild type isolates KUPL of P. solanacearum and the rhizosphere soil microorganisms were highly sensitive to streptomycin and erythromycin (Table 5).

The double mutant isolate KUPL-16 of the pathogen showing resistance to 2 mg/ml of streptomycin and 1 mg/ml of erythromycin was developed following the method of Obaton (1971). The flow-sheet of the procedure for the development of the double marker strain is shown in Fig. 1.

It may be noted (Fig. 1) that initially KUPL-9 isolate was selected having the ability of tolerating 2 mg/ml of streptomycin in the medium. This single marker strain was then further used for the development of erythromycin resistance in a stepwise manner. Thus the isolate KUPL-10 showed resistance to streptomycin (2 mg/ml) and erythromycin (200 μ g/ml). The tolerance of KUPL-12 to erythromycin increased to the level of 600 μ g/ml and finally KUPL-16 was selected with the desired double marker resistance (2 mg/ml streptomycin and 1 mg/ml erythromycin).

In preliminary experiments the double marker mutant isolate KUPL-16 was mixed with soil and recovered at zero time by soil dilution plating technique using PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml). The individual colonies

to streptomycin and erythromycin was compared with that of rhizosphere soil microflora of eggplant. The test organisms were plated on PDA supplemented with different doses of streptomycin sulphate or erythromycin.

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In preliminary experiments the double marker mutant isolate KUPL-16 was mixed with soil and recovered at zero time by soil dilution plating technique using PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml). The individual colonies

Table 5. Comparison of antibiotic sensitivity of different isolates of Pseudomonas solanacearum and other soil microorganisms

Test isolates	No.anti-biotic	Growth on medium containing					
		Streptomycin/ml			Erythromycin/ml		Streptomy-cin (2 mg/ml) and Erythro-mycin(1 mg/ml)
		600/ug	1 mg	2 mg	600 /ug	1 mg	
Soil micro-organisms							
Bacteria	+	-	-	-	-	-	-
Fungi	+	-	-	-	-	-	-
KUPL	+	-	-	-	<u>+</u>	-	-
KUPL-6	+	+	+	-	<u>+</u>	-	-
KUPL-9	+	+	+	+	<u>+</u>	-	-
KUPL-12	+	+	+	+	+	-	-
KUPL-16	+	+	+	+	+	+	+
KUPL-16(R) ^a	+	+	+	+	+	+	+

^a KUPL-16 isolates recovered from soil were designated as KUPL-16 (R).

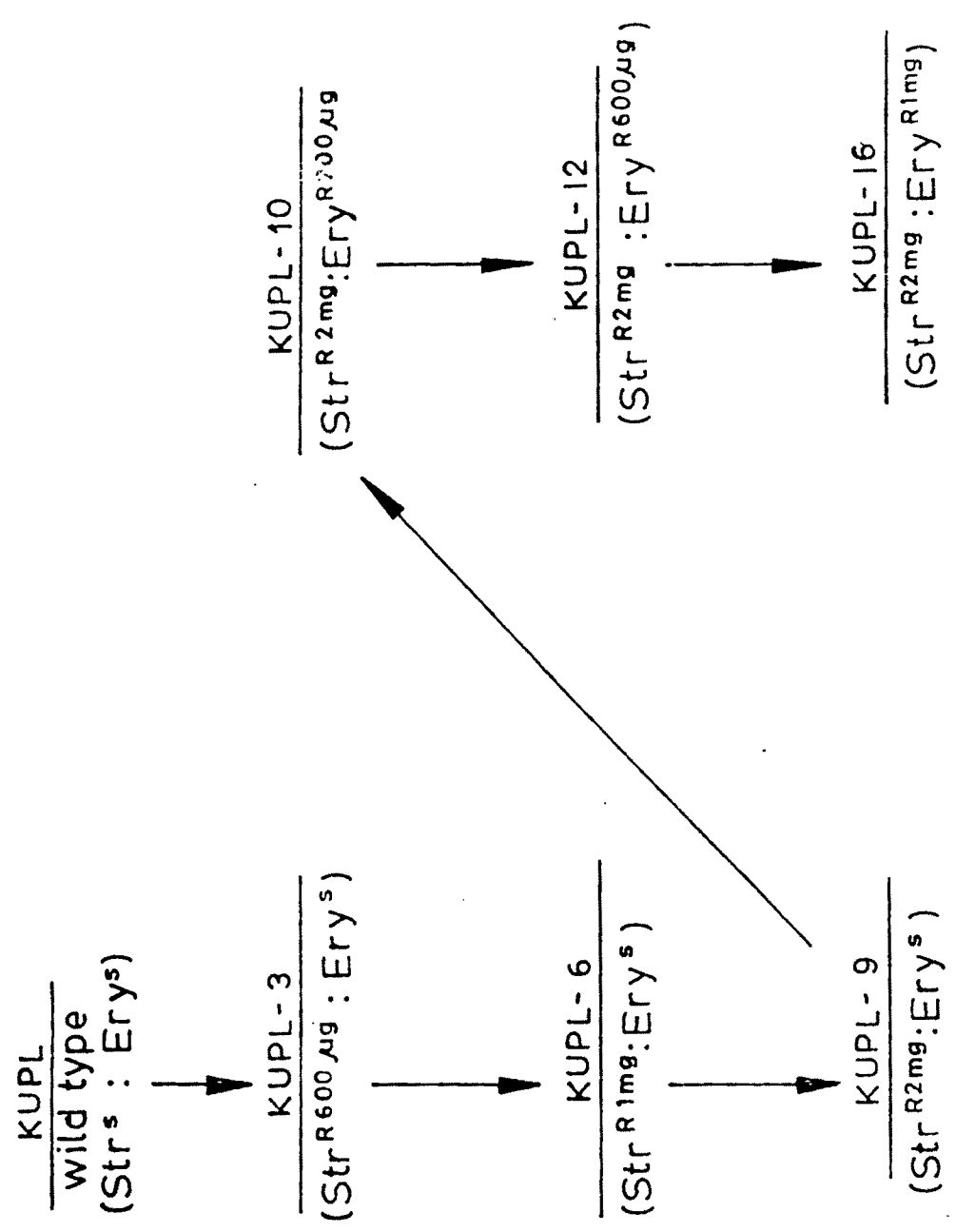


Figure 1. Flow sheet of development of double antibiotic resistant marker strain of *Pseudomonas solanacearum*
 Str = Streptomycin
 S = Sensitive
 Ery = Erythromycin
 R = Resistant

of KUPL-16 recovered from soil were maintained on PDA. The biochemical properties of the wild type isolate KUPL, the double marker strain KUPL-16 and the isolates recovered from the soil (KUPL-16 R) were compared and shown in Table 6. The pathogenecity potentials of KUPL and KUPL-16 were also compared and the results are shown in Table 7.

It is evident from these results that the double marker strain and the isolates recovered from soil showed biochemical characters, carbon source utilization properties and pathogenecity potential similar to that of the wild type isolate. Apparently, the isolate KUPL-16 differed from the wild type isolate by two mutant genes only.

Survival in seed. — In this series of experiments the presence of the eggplant isolate of Pseudomonas solanacearum in seeds of different germplasms of eggplant was examined. Seeds of germplasm collection of BCKV and the cultivars obtained from local markets were plated on PDA and the percentages of total number of infected seeds, and those infected by bacteria, fungi or actinomycetes were determined. The results are presented in Table 8. A summary of the findings is presented in Table 9.

It may be noted from the data that the degree of infection of seeds varied from 36 to 100% depending on the cultivars tested. It was of interest that approximately 30% of the seed samples showed 100% of the seeds with infection. It was also evident that bacterial infection was dominant and 38-100% of the infected seeds yielded bacterial colony including P. solanacearum. Percentages

Table 6. Comparison of the morphological and biochemical characters of different isolates of Pseudomonas solanacearum

Characters compared	Wild type	KUPL-16	KUPL-16(R) ^a
Gram staining	-	-	-
Spore staining	-	-	-
Presence of flagella	+ ^c	+	+
Presence of Capsule	-	-	-
Hydrolysis of Starch	+	+	+
Liquification of gelatin	-	-	-
Carbon sources for growth			
Ribose	+	+	+
Glucose	+	+	+
Maltose	+	+	+
Fructose	+	+	+
L-Arabinose	-	-	-
Sucrose	+	+	+
Cellulose	+	+	+
Starch	+	+	+
Mannitol	+	+	+
Sorbitol	+	+	+
Malonate	-	-	-
Ethanol	-	-	-

^a KUPL-16 isolates recovered from soil were designated as KUPL-16(R)

^b (-) indicates the absence of the characters or negative reaction to the test

^c (+) indicates the presence of the characters or positive reaction to the test.

Table 7. Comparison of pathogenicity of different isolates of P. solanacearum using tomato seedlings susceptible to the pathogen

Observed after days	Percentage of wilted plants of tomato varieties after days		
	Inoculated with	Marglobe Supreme	Best of All
1	Control ^a	0	0
	Wild type isolate ^b (KUPL)	38	17
	KUPL-16 ^c (Double marker)	35	18
2	Control	0	0
	Wild type (KUPL)	69	36
	KUPL-16	71	34
3	Control	0	0
	Wild type (KUPL)	100	67
	KUPL-16	100	65
4	Control	0	0
	Wild type (KUPL)	100	67
	KUPL-16	100	65

^a Control plants were treated with sterilised distilled water

^b Plants were inoculated with suspension of the wild type isolate KUPL

^c Plants were inoculated with suspension of the marker strain KUPL-16.

Table 8. Seed pathological study of Indian germplasms of eggplant.

Code number	Name of the Germplasms	Percentages of total number of seeds infected by				
		Infected	Bacteria	Fungi	Actino-mycetis	Mixed
(1)	(2)	(3)	(4)	(5)	(6)	(7)
IG-1	Pusa Purple Long	100	54	-	-	46
IG-2	Banaras Giant Green	94	84	8	-	2
IG-3	Pusa Kranti	100	82	-	-	18
IG-4	Pusa Purple Cluster	86	72	2	-	12
IG-5	Improved Mukta-keshi	80	74	2	-	4
IG-6	Nurki	98	66	2	-	30
IG-7	K.812	96	58	10	-	28
IG-8	Kanta Brinjal	100	96	-	-	4
IG-9	SM 65(1)	90	76	2	-	12
IG-10	SM 65(2)	98	96	-	-	2
IG-11	T-3	100	92	-	-	8
IG-12	KG Local	100	84	2	-	14
IG-13	SM 50 (A)	100	38	12	-	50
IG-14	Long (1)	66	62	2	-	2
IG-15	K-8(1)	100	70	4	-	26
IG-16	Pant Rithuraj	100	92	2	-	6
IG-17	S-62 (1)	100	80	4	-	16
IG-18	Karimpur	98	84	2	-	12
IG-19	Bhangar-1	84	82	-	-	2
IG-20	H-4(1)	96	80	4	-	12
IG-21	SM-227	96	92	-	-	4
IG-22	SM-48	80	58	2	-	20
IG-23	Salimakra	92	72	12	-	8
IG-24	Kalo Chaila	100	56	-	-	44

Table 8. Seed pathological study of Indian germplasm of eggplant

(1)	(2)	(3)	(4)	(5)	(6)	(7)
IG-25	SM-71	90	64	8	-	18
IG-26	Muktakeshi-1	90	52	4	-	34
IG-28	Long Type (3)	70	68	2	-	-
IG-29	Gourigazar	100	94	-	-	6
IG-30	Pusakesar	94	74	4	-	16
IG-33	Muktakeshi-II	96	82	2	-	12
IG-34	Black Round	86	70	8	-	8
IG-35	Katamakra	84	54	22	-	8
IG-36	Silimum	84	70	4	-	2
IG-37	PRR-7	94	92	2	-	-
IG-38	B-5	96	82	2	-	12
IG-39	Guzrat	98	98	-	-	-
IG-40	Long White	90	56	6	-	28
IG-44	Azad Kranti	80	70	4	-	6
IG-45	Dharampur Local	88	58	12	-	18
IG-46	K-2	90	70	8	-	12
IG-47	SM-31	50	42	4	-	4
IG-48	SM-273	78	68	10	-	-
IG-49	Bhangar-II	100	100	-	-	-
IG-50	White Brinjal	36	36	-	-	-
IG-51	Paichabani	52	36	8	-	8
IG-52	KNG-1	100	98	-	-	2
IG-55	Makra	92	74	10	-	8
IG-57	Purulia Purple	100	98	2	-	-

^a Germplasms were obtained from the department of Horticulture, BCKV

^b Seeds when plated on PDA yielded both bacteria and fungi.

Table 9. Summary of seed pathology of Indian germplasma of eggplant

	Percentage of Infection
Degree of infection	36-100
100% of seeds infected	30
Bacterial infection	38-100
Fungal infection	2-34
Actinomycetes infection	0
Mixed infection	2-40

of the seeds infected with fungi ranged from 2 to 34% and seeds with mixed infection ranged from 2 to 40%, whereas no seed samples yielded actinomycetes.

Majority of seed samples yielding bacterial colony also yielded P. solanacearum as one of the component of contaminating microflora of the seed surface. The colonies were transferred to tetrazolium chloride medium (Kelman, 1954) to test their ability to grow on the medium and the biochemical properties of P. solanacearum were determined.

Survival of the marker strain on eggplant seeds. — In this series of experiments known number of cells of the marker strain

of eggplant isolate of P. solanacearum was placed on the surface of healthy non-infected eggplant seeds and incubated at different temperature condition. At 7 days intervals, the surviving cells on the surface of the seeds were recovered by plating the seeds - on PDA containing 2 mg of streptomycin and 1 mg erythromycin per ml of medium. The results are presented in Table 10.

It may be noted that during the first 7 days of incubation, the population of the marker strain increased on the surface of seeds irrespective of the temperature conditions (except 40 C) followed by a gradual decline of the population density thereafter. It is also evident from the data that at low temperature (22 C), the population declined less rapidly, whereas at high temperature (40 C), population declined rapidly and became zero after 28 days of incubation. At 30 C, after 28 days of incubation, approximately 300 marker cells per seed could be recovered.

In a separate series of experiments the effects of temperature and relative humidity of the ambient atmosphere on the survival of the marker strain in eggplant seeds were determined. Seeds were inoculated with a known number of bacterial cells and then incubated at constant temperature of 30 C with variable relative humidity (%) of the ambient atmosphere in humidity chambers. At 7 day intervals, the cells surviving in seeds were determined by plating the seeds on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml). The results are presented in Table 11.

It may be noted that the organism survived well on seeds when

Table 10. Survival of the marker strain of Pseudomonas solanacearum on eggplant seeds kept under constant relative humidity and different ambient temperature conditions

Incubation temperature (C)	Number of CFU ^a recovered ($\times 10^5$)/100 seeds after days of incubation following inoculation ^b				
	0 ^c	7	14	21	28
22	300 (7.48) ^d	1600 (8.20)	200 (7.30)	40 (6.60)	9 (5.95)
30	300 (7.48)	1000 (8.0)	0.5 (4.70)	1 (5.00)	0.3 (4.48)
35	300 (7.48)	800 (7.90)	3.5 (5.54)	3 (5.48)	0.9 (4.95)
40	300 (7.48)	0.5 (4.70)	0.01 (3.00)	0.0001 (1.00)	0

^aCFU = Colony Forming Units of the marker strain on PDA

^bHealthy non-infected seeds were inoculated by mixing with thick suspension of marker cells of the pathogen

^cNumber of marker cells on seed surface was determined at '0' time and at indicated intervals

^dFigures in the parentheses indicate log transformation values.

Table 11. Survival of the marker strain of Pseudomonas solanacearum on eggplant seeds kept at 30 C and different relative humidity conditions

RH (%)	Number of CFU ^a recovered ($\times 10^5$)/100 seeds after days of incubation ^b				
	0 ^c	7	14	21	28
1.7	200 (7.30) ^d	1000 (8.00)	38 (6.58)	36 (6.56)	29 (6.46)
16.9	200 (7.30)	1600 (8.20)	200 (7.30)	40 (6.60)	9 (5.95)
33.8	200 (7.30)	2000 (8.30)	270 (7.43)	45 (6.65)	40 (6.60)
50	200 (7.30)	100 (7.00)	10 (6.00)	0.8 (4.90)	0.1 (4.00)

^aCFU = Colony Forming Units of the marker strain on PDA

^b Healthy non-infected seeds were inoculated by mixing with thick suspension of marker cells of the pathogen

^c Number of marker cells on seed surface was determined at '0' time and at indicated intervals

^d Figures in the parentheses indicate log transformation values.

the RH values ranged from 1.7 to 33.8%. In all these moisture conditions, the population increased initially and then declined, but a significant number of colony forming units of the marker strain could be isolated from the seeds after 28 days of incubation. At RH 50%, however, the organisms declined even after 7 days of incubation and became approximately 100 colony forming units per seed. Apparently low temperature, and dry condition favoured persistence of the marker strain on the carphosphere of the plant for a long time.

Survival in soil. — There are many reports in the literature that Pseudomonas solanacearum is soil-borne and can survive for long term in the soil (Buddenhagen and Kelman, 1964; Buddenhagen, 1965; Leben, 1981; Dukes et al., 1965). All the studies on the survival of the pathogen in soil, however, were of indirect nature, based on disease incidence of the susceptible plants sown in the soil. The soil survival ability of the double marker strain was examined in this study. Known number of cells of the marker strain were mixed with natural or sterilized soil of Kalyani (Loam, pH 6.8) and incubated at 30 C. Soil samples were collected at zero time or at indicated intervals and plated on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml). The number of surviving cells which formed discrete colonies on the selective medium were counted.

In preliminary experiments the effect of initial population density on the recovery percentage of the marker strain from the soil was examined. Natural and sterilized soil samples were inoculated with different cell densities of the marker strain

from the soil was examined. Natural and sterilized soil samples were inoculated with different cell densities of the marker strain separately and cells were then recovered at zero time by plating the soil dilutions on the selective medium containing streptomycin and erythromycin. The number of cells recovered per gram of soil was determined and expressed as percentage recovery of the initial cell density added to unit weight of soil. The results are shown in Table 12.

It may be noted that initial cell density had influence on the percentage recovery of cells from natural or sterilized soils. Maximum number of cells could be recovered from natural soil when 8×10^7 cells/gram were used as initial cell density. Cell densities higher or lower than this yielded less percentage recovery of cells from natural soil. The recovery pattern from sterilized soil also indicated that with decreasing cell density less percentage of cells were recovered. Based on the recovery pattern from natural or sterilized soil it appeared that 8×10^7 cells in general, gave good percentage of recovery (75% and 50% in natural and sterilized soils respectively). In most of the subsequent experiments therefore, the initial cell density used was 10^8 cells per gram of soil.

Factors affecting survival in soil. — In this series of experiments environmental and nutritional factors affecting the survival of the double marker strain in soil were investigated. In preliminary experiments the effect of slow drying of soil on the survival ability of the marker strain was tested. The marker strain was mixed with natural and sterile soil and

Table 12. Effect of initial cell density on the recovery of marker strain of Pseudomonas solanacearum cells from soil

Initial ^b cell density ($\times 10^7$)/g of soil	Number ^a ($\times 10^7$) and percentages of cells recovered from soil type			
	Natural ^c		Sterilized ^d	
	Number	Percentage	Number	Percentage
5000	3000	60.0	3000	60.0
800	500	62.5	500	62.5
30	10	33.33	11	36.7
8	6	75.0	4	50.0
0.8	0.4	50.0	0.3	37.5
0.06	0.02	33.3	0.02	33.3
0.007	0.004	57.1	0.004	57.1

^a Number of cells recovered per gram of soil at zero time after mixing were determined by plating on PDA containing streptomycin and erythromycin

^b A thick suspension of 48 hour old cells were prepared and the cell density was determined by a Haemocytometer. Portion of suspension was added to known weight of soils to get the required cell density

^c Unsterilized soil was considered as natural soil

^d Natural soil was sterilized at 15 p.s.i. for 15 minutes on 3 consecutive days.

incubated for different days. In one set, the inoculated soils were kept in closed petridishes and the other set was incubated with the lids of the petridishes open. Soil samples were collected at intervals and the percentage of surviving cells was determined. The results are shown in Table 13. The log numbers of surviving cells are presented in Figure 2.

It is evident from the data that the double marker strain could survive well in sterilized soil kept in a closed vessel. The population increased during the first two weeks of incubation but decreased to some extent during the next two weeks. The population after four weeks was approximately ten times more than the initial number added to the soil. In contrast, in natural soil the population increased during the first seven days but then declined rapidly and after four weeks it became somewhat less than the initial number of cells added to the soil.

When the inoculated soils were allowed to dry slowly by keeping the lid of the petridishes open, the population in natural soil declined much more rapidly and became approximately hundred times less than the original number of cells. Slow drying affected the survival in the sterilized soil also. The population growth was less as compared to the closed vessels and the final population density was also significantly less than that of the closed vessels.

Effect of temperature on survival. — To test the effect of ambient temperature on the survival ability of the marker strain, soil samples were inoculated and adjusted to 30% or 60% W.H.C. The bottles were then incubated at different temperature

Table 13. Survival of cells of double marker strain of Pseudomonas solanacearum in soil

Incubation days	Number of cells ^a /gm of soil ($\times 10^7$) surviving in soils ^b kept in vessels with			
	Lid closed ^c		Lid open ^d	
	Natural	Sterilized	Natural	Sterilized
0	5 (7.70) ^g	7 (7.85)	4 (7.60)	7 (7.85)
7	349 (9.54)	851 (9.93)	180 (9.26)	753 (9.88)
14	48 (8.68)	863 (9.94)	29 (8.46)	651 (9.81)
21	31 (8.49)	401 (9.60)	0.3 (6.48)	99 (9.00)
28	9 (7.95)	99 (9.00)	0.09 (5.95)	4 (7.60)

^a Initial cell density was 1×10^8 cells/g of soil

^b Soils were adjusted to 60% W.H.C. and incubated at 30 C

^c Soils were kept in 20 cm petridishes with lids closed

^d Soils were kept in 20 cm petridishes with lids open

^e Soil from a fallow land of the campus was used and considered

^f Soil samples were sterilized in an autoclave at 15 p.s.i. for 15 minutes on 3 consecutive days to get sterilized soils

^g Figures in parentheses indicate log transformation values.

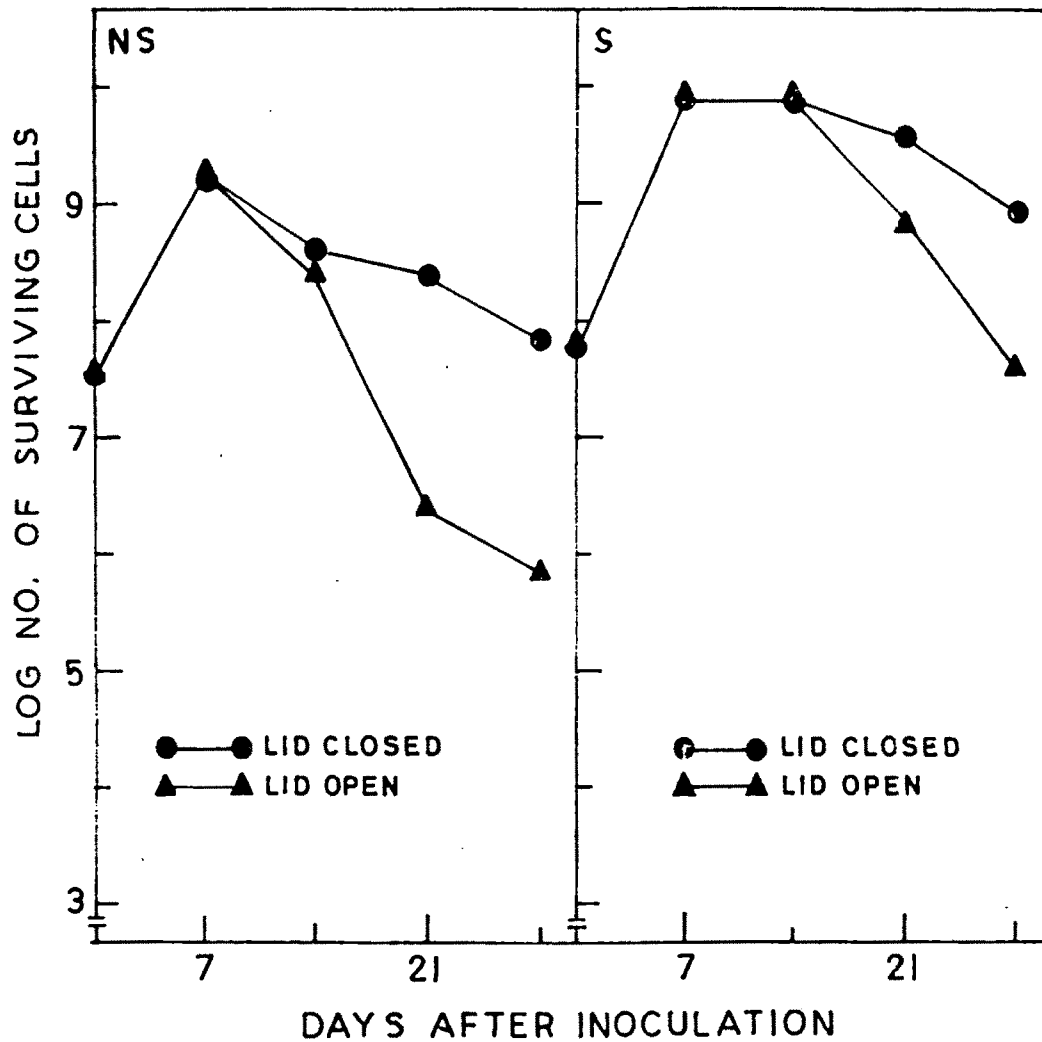


Figure 2. Survival of double marker strain of *P. solanacearum* in natural (NS) and sterilized (S) soils as a function of days of incubation.

conditions. The surviving cells were counted at 7 day intervals by soil dilution plate technique. The results are presented in Tables 14 and 15. The log number of surviving cells are presented in Figures 3 and 4.

It is evident from the data that at 30% W.H.C. and at ambient temperature of 40 C the organism failed to survive in both natural and sterilized soils. In all lower temperature regimes ranging from 15 to 30 C the survival of marker strain was not affected in sterilized soil. The population of the marker strain in inoculated sterilized soil increased during the first 7 days irrespective of the temperature conditions. Thereafter, the population density remained almost constant during the tenure of the experiment. At 40 C, however, the population declined rapidly after 7 days of incubation and became zero after 28 days. The survival pattern in the natural soil was similar to that in sterilized soil, except that at lower temperatures (15 to 30 C) the population slowly declined and after 28 days of incubation became approximately one log unit less than that of the initial cell density added to the soil (Table 14 and Figure 3).

The survival of the marker strain in soil adjusted to 60% W.H.C. and incubated at different temperatures (Table 15 and Figure 4) exhibited the similar pattern observed in soils adjusted to 30% W.H.C. It is apparent from the results that incubation of soils at 40 C was significantly inhibitory to the survival of the marker strain irrespective of the two moisture levels tested in this study. The population of the marker strain declined rapidly at 40 C and became zero after 28 days of incubation.

Table 14. Effect of ambient temperatures on the survival of cells of double marker strain of Pseudomonas solanacearum in soil adjusted to 30% W.H.C.

Soil type	Ambient ^b Temperature (C)	Number of cells recovered ^a /g. of soil ($\times 10^7$) after days of incubation				
		0	7	14	21	28
Natural ^c	15	7 (7.85) ^e	260 (9.41)	55 (8.74)	15 (8.18)	1 (7.00)
Sterilized ^d	15	7 (7.85)	390 (9.59)	810 (9.91)	1592 (10.20)	980 (9.99)
Natural	25	7 (7.85)	1000 (10.00)	323 (9.51)	22 (8.34)	1 (7.00)
Sterilized	25	9 (7.95)	1121 (10.05)	1592 (10.20)	1920 (10.28)	1120 (10.05)
Natural	30	6 (7.78)	310 (9.49)	25 (8.40)	19 (8.28)	0.9 (6.95)
Sterilized	30	8 (7.90)	779 (9.89)	782 (9.89)	372 (9.57)	65 (8.81)
Natural	40	5 (7.70)	251 (9.40)	0.009 (4.95)	0.00001 (2.00)	-
Sterilized	40	8 (7.90)	310 (9.49)	0.01 (5.00)	0.00002 (2.30)	-

^a Streptomycin (2 mg/ml) and erythromycin (1 mg/ml) resistant cells were introduced to soil at an initial cell density of 1×10^8 . At intervals number of surviving cells was determined by plating on PDA containing 2 mg/ml streptomycin and 1 mg/ml erythromycin

^b Soil samples in glass bottles containing the marker strain were incubated at different temperature controlled cabinets

^c Soil from a fallow land of the campus was used and considered as natural soil

^d Soil samples were sterilized for 3 consecutive days in an autoclave at 15 p.s.i. for 15 minutes to get sterilized soils

^e Figures in parentheses indicate log transformation values.

Table 15. Effect of ambient temperatures on the survival of double marker strain of Pseudomonas solanacearum in soil adjusted to 60% W.H.C.

Soil type	Ambient temperature (C) ^b	Number of cells recovered ^a /gm of soil ($\times 10^7$) days after inoculation				
		0	7	14	21	28
Natural ^c	15	7 ³ (7.85)	561 (9.75)	80 (8.90)	22 (8.34)	12 (8.08)
Sterilized ^d	15	8 (7.90)	600 (9.78)	990 (10.00)	1801 (10.26)	1300 (10.11)
Natural	25	9 (7.95)	1191 (10.08)	401 (9.60)	99 (9.00)	3 (7.48)
Sterilized	25	8 (7.90)	1260 (10.10)	1800 (10.26)	2200 (10.34)	1190 (10.08)
Natural	30	7 (7.85)	347 (9.54)	49 (8.69)	33 (8.52)	7 (7.85)
Sterilized	30	7 (7.85)	860 (9.93)	867 (9.94)	407 (9.61)	96 (8.98)
Natural	40	5 (7.70)	290 (9.46)	0.01 (5.00)	0.00001 (2.00)	-
Sterilized	40	8 (7.90)	380 (9.58)	0.04 (5.60)	0.00007 (2.85)	-

^a Streptomycin (2 mg/ml) and erythromycin (1 mg/ml) resistant cells were introduced to soil at an initial cell density of 1×10^8 . At intervals number of surviving cells were determined by plating on PDA containing 2 mg/ml of streptomycin and 1 mg/ml of erythromycin.

^b Soil samples in glass bottles containing the marker strain were incubated at different temperature controlled cabinets

^c Soil from a fallow land of the campus was used and considered as natural soil.

^d Soil samples were sterilized for 3 consecutive days in an autoclave at 15 p.s.i. for 15 minutes to get sterilized soils.

^e Figures in parentheses indicate log transformation values.

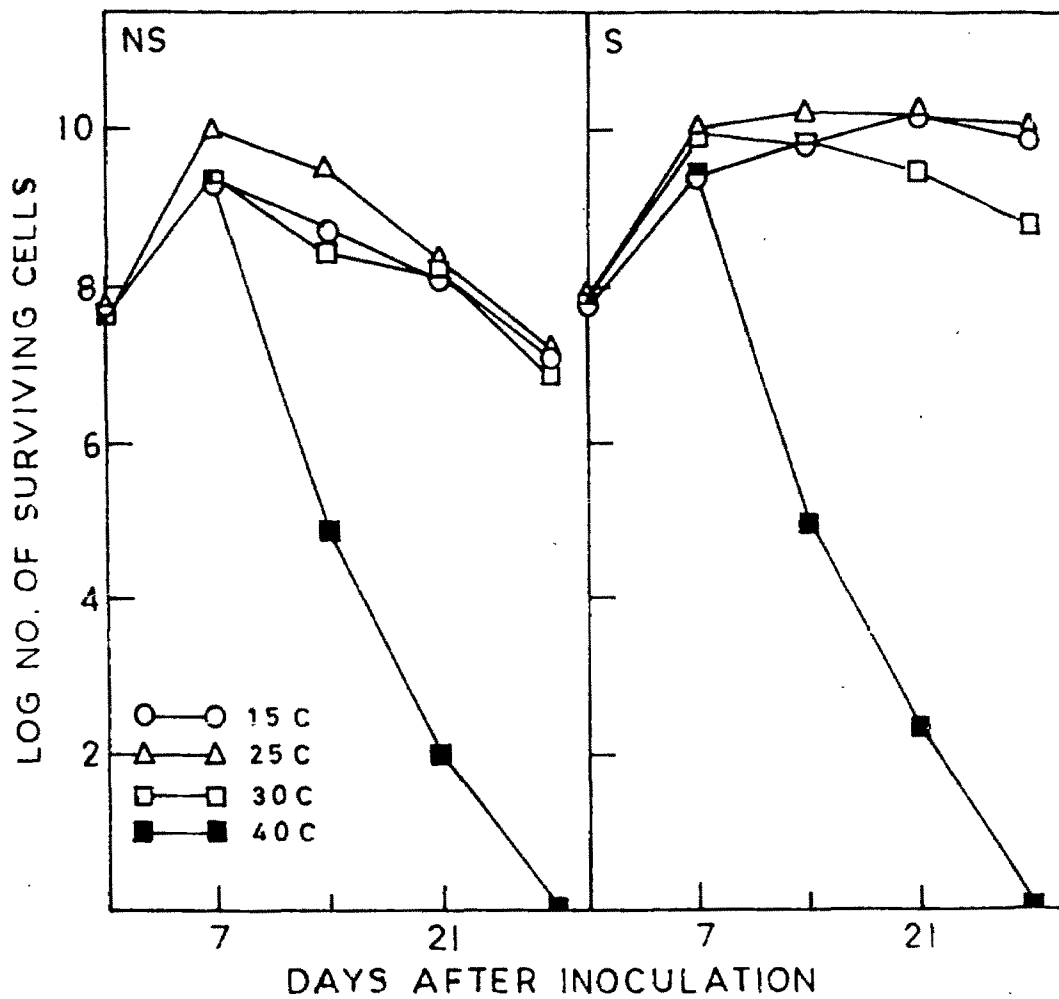


Figure 3. Survival of the double marker strain of *P. solanacearum* in natural (NS) and sterilized (S) soils adjusted to 30% W.H.C. and kept at different temperature conditions as a function of days of incubations.

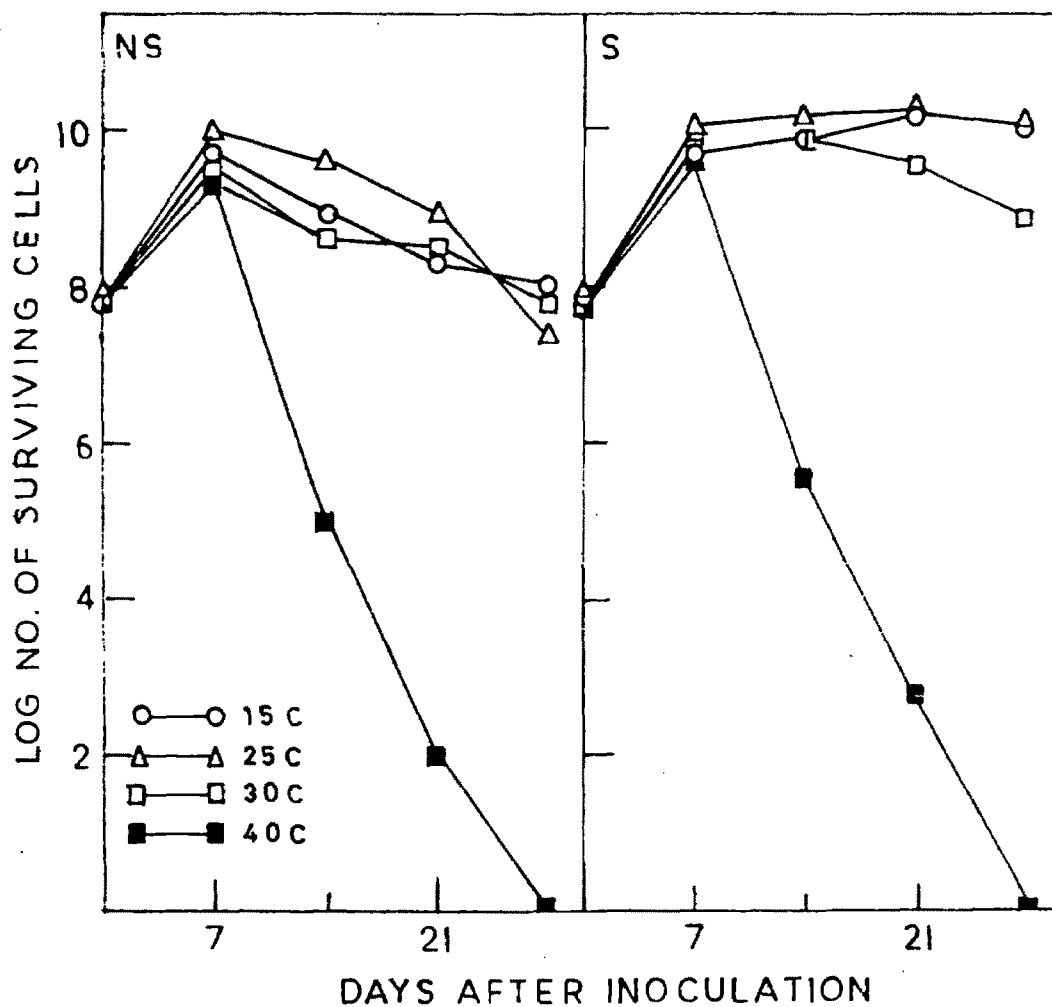


Figure 4. Survival of the double marker strain of *P. solanacearum* in natural (NS) and sterilized (S) soils adjusted to 60% W.H.C. and kept at different temperature conditions as a function of days of incubation.

Effect of moisture on survival. — Moisture is an important parameter for survival of phytopathogenic bacteria in soil. In the previous experiment two moisture levels tested such as 30% and 60% WHC had no apparent effect on the survival of the test marker strain. This experiment, therefore, was designed to test the survival ability of the pathogen incubated at variable moisture content of the soil. The natural and sterilized soil samples were inoculated, adjusted to different moisture levels and incubated at 28 ± 2 C under laboratory conditions. The moisture level of the soil was expressed as % WHC and % water content of the soil. The surviving cells were determined at 7 days intervals. The results are presented in Table 16 and Figure 5.

It is apparent from the results that soil moisture levels had little or no effect on the survival of the test marker strain in sterilized soil. In natural soil, however, moisture levels (10% and 14%) were to some extent inhibitory and the population density declined about two log units after 28 days of incubation. At higher moisture levels the decline in population was less and more slow as compared to the lower moisture levels.

Effect of flooding and water-logging of soil on survival. — In this experiment the effects of flooding and consequent water-logging of the soil on the survival of the marker strain were examined. Natural and sterilized soil samples were inoculated with known number of the marker strain, flooded under different depth of water levels and the water-logged soils were incubated

Table 16. Effect of moisture content of soil on the survival of the marker strain of Pseudomonas solanacearum race 1

Soil type	Moisture content		Number of cells recovered/g of soil ($\times 10^7$) ^a				
	of soil ^b		after days of incubation				
	WHC(%)	Water(%)	0	7	14	21	28
Natural ^e	24	10	7 (7.85)	222 (9.35)	71 (7.00)	0.9 (6.95)	0.07 (5.85)
Sterilized ^f	24	10	6 (7.78)	680 (9.83)	682 (9.83)	290 (9.46)	15 (8.18)
Natural	32	14	9 (7.95)	250 (9.40)	5 (7.70)	1 (7.00)	0.2 (6.30)
Sterilized	32	14	5 (7.70)	700 (9.85)	705 (9.85)	305 (9.48)	25 (8.40)
Natural	40	17	6 (7.78)	310 (9.49)	25 (8.40)	19 (8.28)	0.9 (6.95)
Sterilized	40	17	8 (7.90)	779 (9.89)	782 (9.89)	370 (9.57)	65 (8.81)
Natural	45	20	9 (7.95)	327 (9.51)	30 (8.48)	25 (8.40)	1 (7.00)
Sterilized	45	20	7 (7.85)	793 (9.90)	799 (9.90)	379 (9.58)	77 (8.89)
Natural	53	23	6 (7.78)	331 (9.52)	38 (8.58)	29 (8.46)	2 (7.30)
Sterilized	53	23	5 (7.70)	801 (9.90)	809 (9.91)	383 (9.58)	85 (8.93)
Natural	59	25	5 (7.70)	347 (9.54)	49 (8.69)	33 (8.52)	7 (7.85)
Sterilized	59	25	7 (7.85)	860 (9.93)	867 (8.94)	407 (9.61)	96 (8.98)

^a Initial number of cells added to soil was 1×10^8 cells/g of soil

^b All soils were incubated at 30 C

^c Per cent water holding capacity of the soil

^d Per cent water content (wt/wt) of soil

^e Soils from a fallow land of the campus was used and considered as natural soil

^f Soil samples were sterilized at 15 p.s.i. for 15 minutes on 3 consecutive days

^g Figures in parentheses indicate log transformation values.

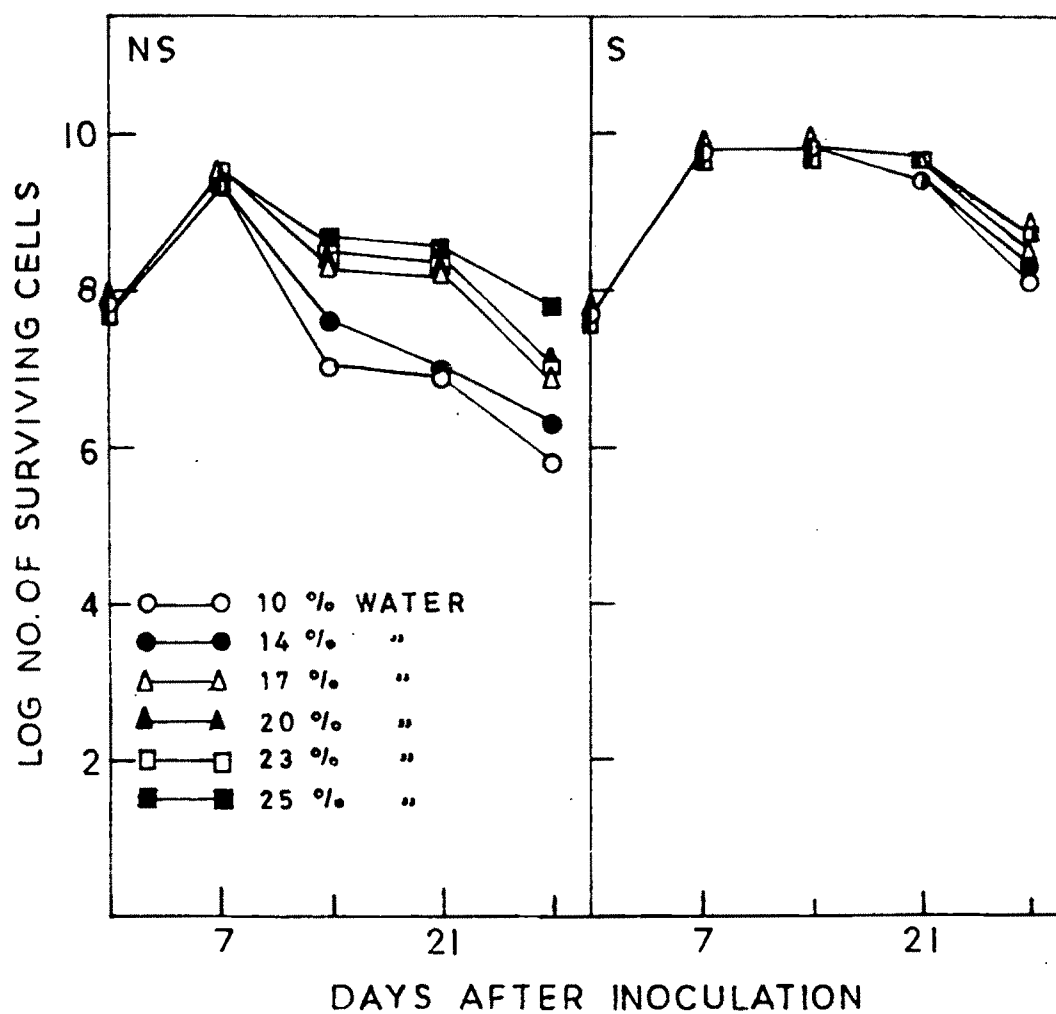


Figure 5. Survival of the double marker strain of *P. solanacearum* in natural (NS) and sterilized (S) soils adjusted to different percentages of moisture levels and determined as a function of days after inoculation.

at 28 C. Survival of the marker strain in soil and in the water phase was determined separately at intervals. Samples from the soil or from the standing water phase were drawn at zero time and on indicated days after inoculation. The results are presented in Table 17. The log number of surviving cells in soil and water phase are shown in Figures 6 and 7. The percentage distribution of the marker strain in soil and water phases are presented in Figures 8 and 9.

It is apparent from the results that flooding of the soil caused significant decrease in the population density irrespective of natural and sterilized soils. In general, the decline in population after flooding was more in natural soil than in sterilized soil. It was of interest that both in natural and sterilized soils a significant number of marker cells could be isolated from the water phase. Evidently, the marker strain became distributed in soil and water phase following flooding. The distribution pattern of the cells in water and soil phase indicated that at zero time 99.9% cells were in the soil phase, but with the increase in time of incubation the cells gradually became distributed more and more in the water phase. Thus, after 28 days of incubation the soil phase had little or no marker strain and 87 to 100% of the recoverable cells were obtained from the water phase only.

Effects of amendments of soils on survival. — Natural soil was amended with chemical fertilizers, organic manures, oil-cakes and host leaf residues separately. The survival of the marker strain in these amended soils was evaluated. Chemical fertilizers

Table 17. Effect of flooding of the soil with different depth of standing water on Pseudomonas solanacearum race 1.

Soil type	Depth of standing water (cm)	Number of cells ^a recovered per gram of soil or per ml of water (x 10 ⁶) after incubation for days									
		0		7		14		21		28	
		Soil ^b phase	Water ^c phase	Soil phase	Water phase	Soil phase	Water phase	Soil phase	Water phase	Soil phase	Water phase
Natural	0.8	70 (7.85) ^d	0.0001 (2.00)	330 (8.52)	0.2 (5.30)	38 (7.58)	33 (7.52)	2 (6.30)	110 (8.04)	0.08 (4.90)	900 (8.95)
Sterilized	0.8	90 (7.95)	0.0005 (2.70)	630 (8.80)	76 (7.88)	78 (7.89)	1500 (9.18)	5 (6.70)	130 (8.11)	4 (6.60)	29 (7.46)
Natural	1.0	60 (7.78)	0.0003 (2.48)	290 (8.46)	21 (7.32)	15 (7.18)	50 (7.70)	1 (6.00)	250 (8.40)	0.05 (4.70)	1800 (9.26)
Sterilized	1.0	90 (7.95)	0.00009 (1.95)	420 (8.62)	35 (7.54)	38 (7.58)	2200 (9.34)	2 (6.30)	150 (8.18)	2 (6.30)	33 (7.52)
Natural	1.2	50 (7.70)	0.0005 (2.70)	170 (8.23)	34 (7.53)	11 (7.04)	230 (8.36)	0.8 (5.90)	2 (6.30)	0.0005 (2.70)	0.5 (5.70)
Sterilized	1.2	60 (7.78)	0.0002 (2.30)	300 (8.48)	24 (7.38)	21 (7.32)	1800 (9.26)	1 (6.00)	130 (8.11)	0.5 (5.70)	60 (7.78)
Natural	1.5	80 (7.90)	0.00008 (1.90)	900 (8.95)	44 (7.64)	0.6 (5.78)	290 (8.46)	0.001 (3.00)	0.5 (5.70)	-	0.001 (2.00)
Sterilized	1.5	50 (7.70)	0.00005 (1.70)	150 (8.18)	22 (7.34)	12 (7.08)	2700 (9.43)	3 (6.48)	190 (8.28)	0.1 (5.00)	70 (7.85)

^a Population of Pseudomonas solanacearum expressed as number of cells/g of soil or per ml of water recorded after indicated days of intervals using PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml) following dilution plating technique

^b Soil samples in glass jars were flooded with water and portions of submerged soils were used

^c Soil samples were flooded with distilled water and the water phase was adjusted to 0.9, 1.0, 1.2, 1.5 cm respectively above the soil surface. Portions of these water phase were assayed for presence of Pseudomonas solanacearum. Samples of water phase were diluted serially and the population of the bacteria in such samples were determined by plate count method

^d Figures in parentheses indicate log transformation values.

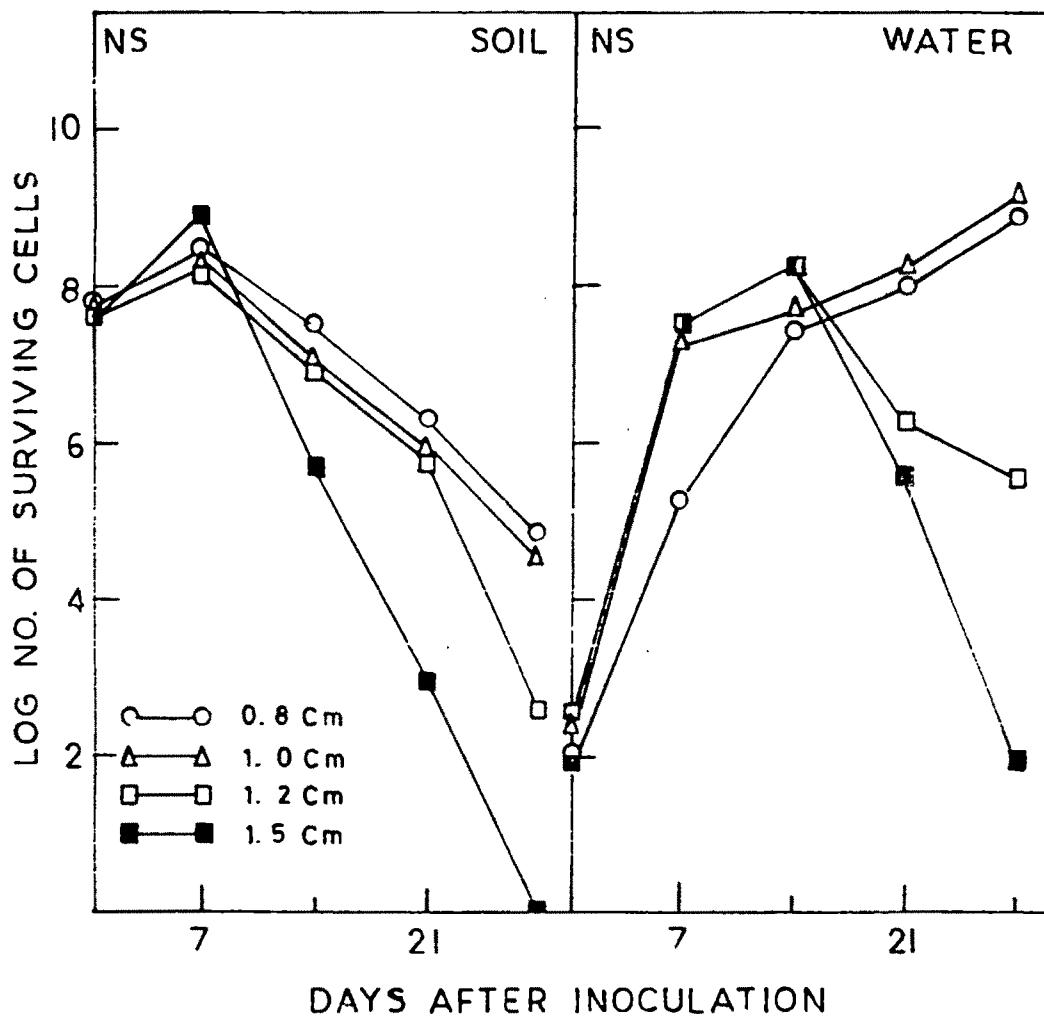


Figure 6. Effect of flooding of natural soil (NS) on the survival of the double marker strain of *P. solanacearum*. The population density in soil phase and water phase as a function of incubation days was determined.

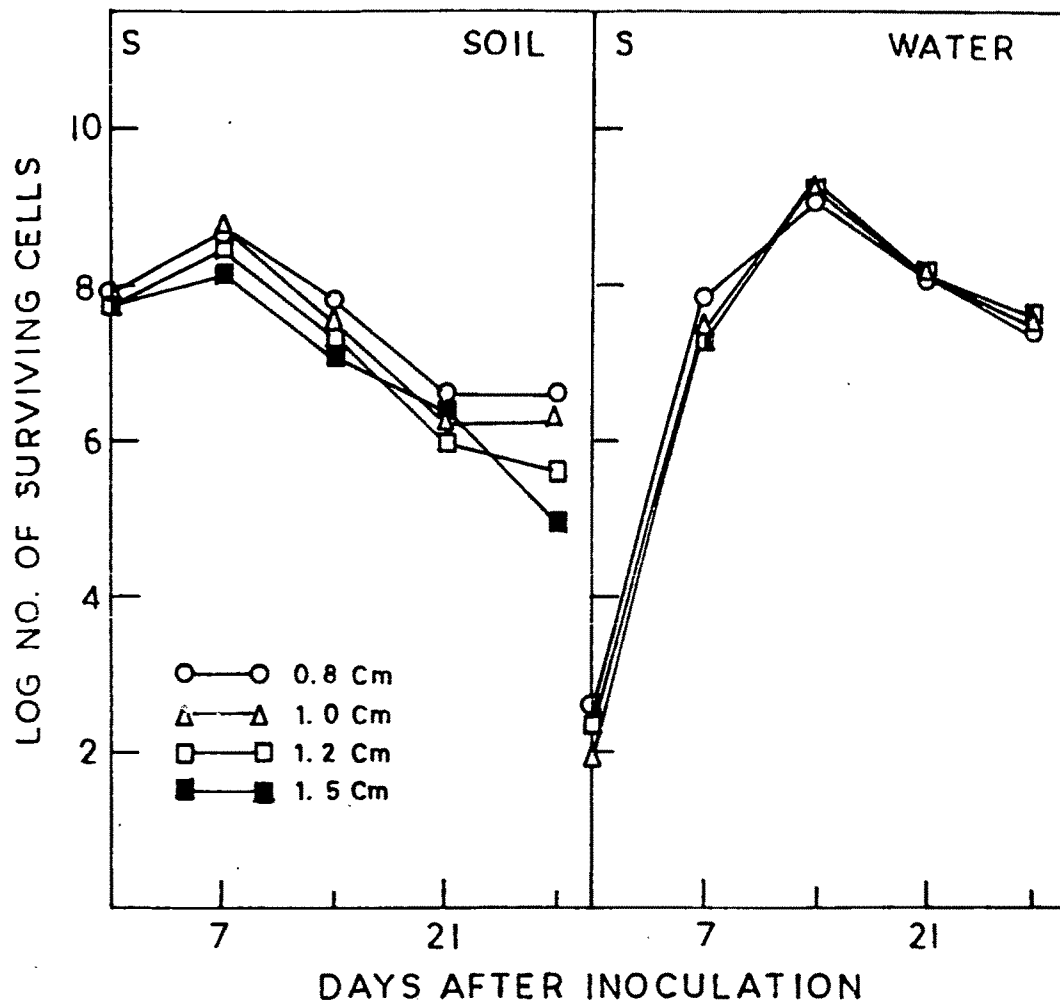


Figure 7. Effect of flooding of sterilized soil (S) on the survival of the double marker strain of *P. solanacearum*. The population density in soil phase and water phase as a function of incubation days was determined.

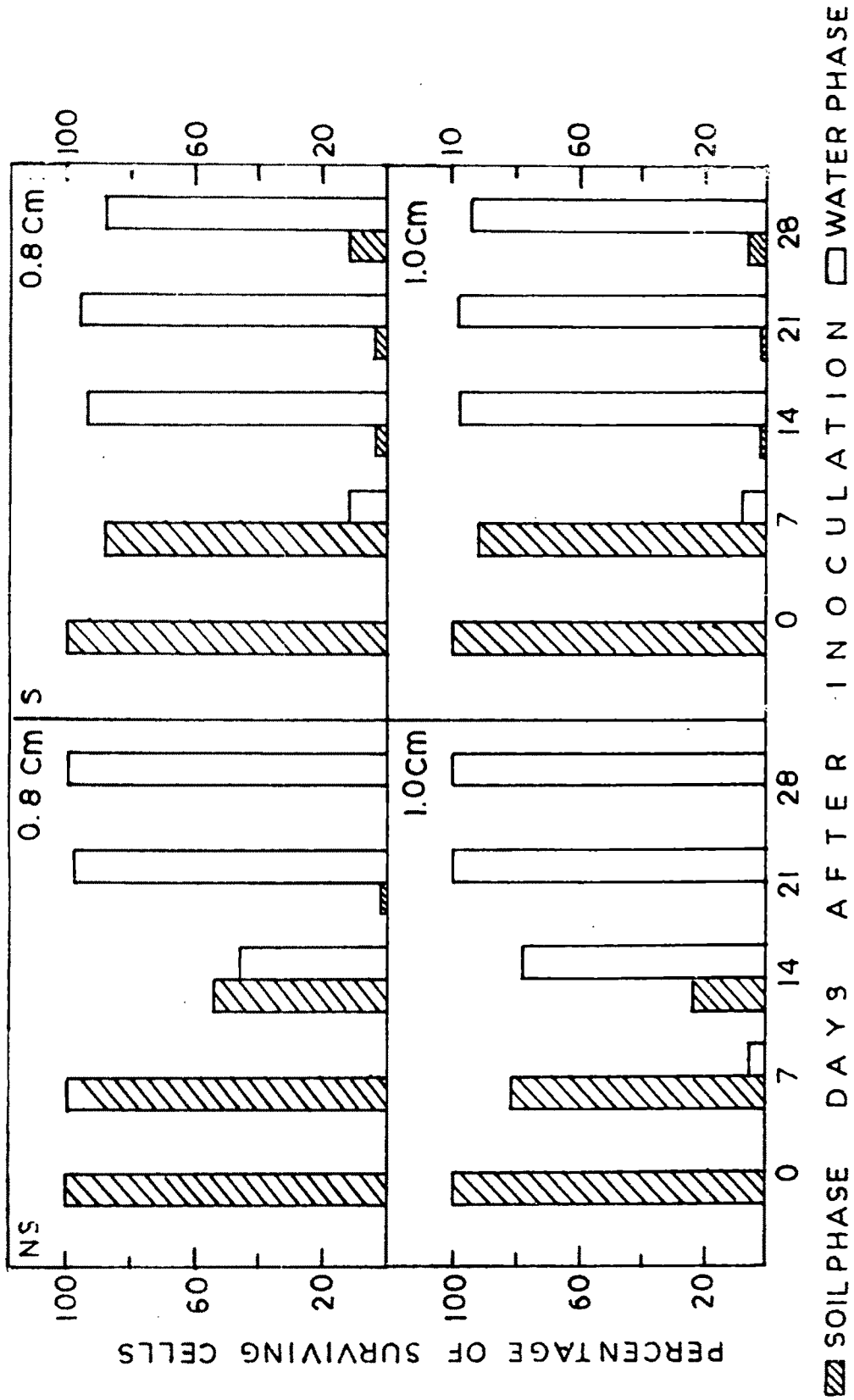


Figure 8. Comparison of the percentage distribution of surviving cells of the double marker strain of *P. solanacearum* in soil phase or water phase following flooding of natural soil (NS) and sterilized soils (S).

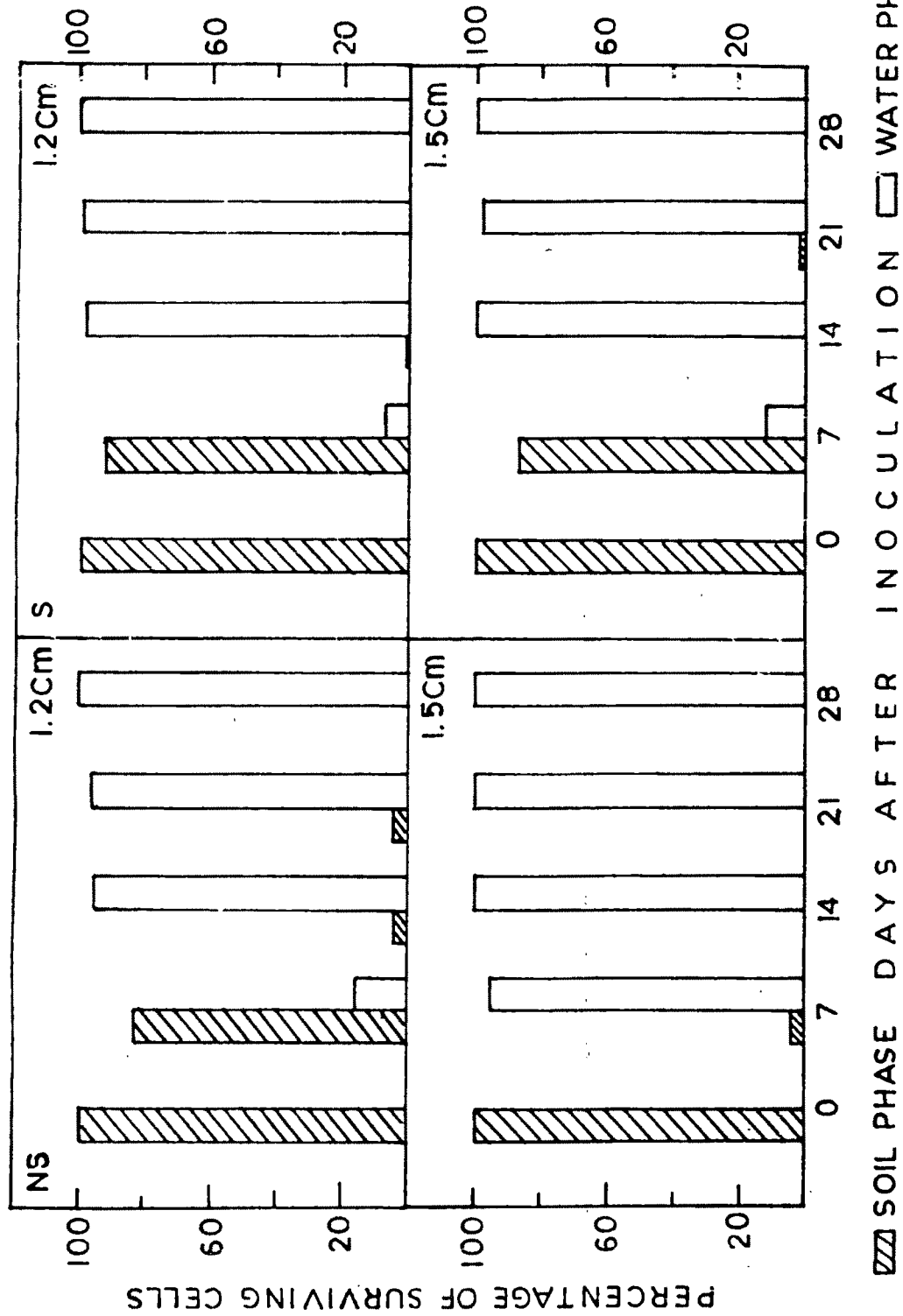


Figure 9. Comparison of the percentage distribution of surviving cells of the double marker strain of *P. solanacearum* in soil phase or in water phase following flooding of natural soils (NS) and sterilized soils (S).

used were potassium nitrate, ammonium sulphate, and urea. Organic manures used were cowdung and oil cakes of linseed, mahua, mustard and neem, and powdered leaves of eggplant was used as leaf residue amendments. Soils after amendment were inoculated with the marker strain and incubated at 28 C in the laboratory. Soil samples were drawn at 7-day interval and the surviving cells were determined by soil-dilution plate technique using PDA supplemented with streptomycin (2 mg/ml) and erythromycin (1 mg/ml). The data on survival of the marker strain in soils amended with chemical fertilizers are shown in Tables 18 to 20 and Figure 10.

It is apparent from the results that both potassium nitrate and urea were significantly inhibitory to the survival of the pathogen in the soil. In contrast, ammonium sulphate was rather conducive for the survival of the pathogen in natural soils. It was interesting that amendment of soil with urea irrespective of concentration used caused rapid decline of cell density of the marker strain within 7 days of amendment. Survival in soils amended with different concentrations of ammonium sulphate was more as compared to the control unamended soil. The population declined rapidly in unamended soils, but declined slowly and remained high even after 28 days of incubation in amended soils.

Survival data with respect to cowdung and oil-cakes are presented in Tables 21 to 25 and Figures 11 and 12. It may be noted that survival of the marker strain, in general, was more in soil amended with oilcakes as compared to control soils

Table 18. Effects of amendment of natural^a soil with different doses of potassium nitrate on the population dynamics of the marker strain of Pseudomonas solanacearum race 1

Days after inoculation	Unamended soil (KNO ₃ not added)	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with potassium nitrate at a concentration (%) ^c of			
		0.5	1.0	1.5	2.0
0	7 (7.85) ^d	6 (7.78)	9 (7.95)	6 (7.78)	9 (7.95)
7	314 (9.50)	9 (7.95)	0.01 (5.00)	0.003 (4.48)	-
14	23 (8.36)	0.4 (6.60)	0.002 (4.30)	-	-
21	15 (8.18)	-	-	-	-
28	0.7 (6.85)	-	-	-	-

^a Non-sterile natural soil as described in Materials & Methods

^b Number of colonies recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentration (wt/wt) of potassium nitrate amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

Table 19. Effects of amendment of natural^a soil with different doses of ammonium sulphate on the population dynamics of the marker strain of Pseudomonas solanacearum race 1

Days after inoculation	Unamended soil (NH ₄) ₂ SO ₄ not added	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with ammonium sulphate at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	7 (7.85) ^d	8 (7.90)	9 (7.95)	9 (7.95)	6 (7.78)
7	315 (9.50)	425 (9.63)	435 (9.64)	429 (9.63)	424 (9.63)
14	28 (8.45)	399 (9.60)	403 (9.61)	400 (9.60)	382 (9.58)
21	16 (8.20)	129 (9.11)	111 (9.05)	102 (9.01)	98 (8.99)
28	0.7 (6.85)	31 (8.49)	25 (8.40)	17 (8.23)	9 (7.95)

^a Non-sterile natural soil as described in Materials & Methods

^b Number of colonies recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentrations(wt/wt) of ammonium sulphate was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

Table 20. Effects of amendment of natural^a soil with different doses of urea on the population dynamics of the marker strain of Pseudomonas solanacearum race 1

Days after inoculation	Unamended soil (Urea not added)	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with urea at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	7 (7.85)	9 (7.95)	8 (7.90)	7 (7.85)	9 (7.95)
7	315 (9.50)	-	-	-	-
14	27 (8.43)	-	-	-	-
21	16 (8.20)	-	-	-	-
28	0.8 (6.90)	-	-	-	-

^a Non-sterile natural soil as described in Materials & Methods

^b Number of colonies recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentration (wt/wt) of urea amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

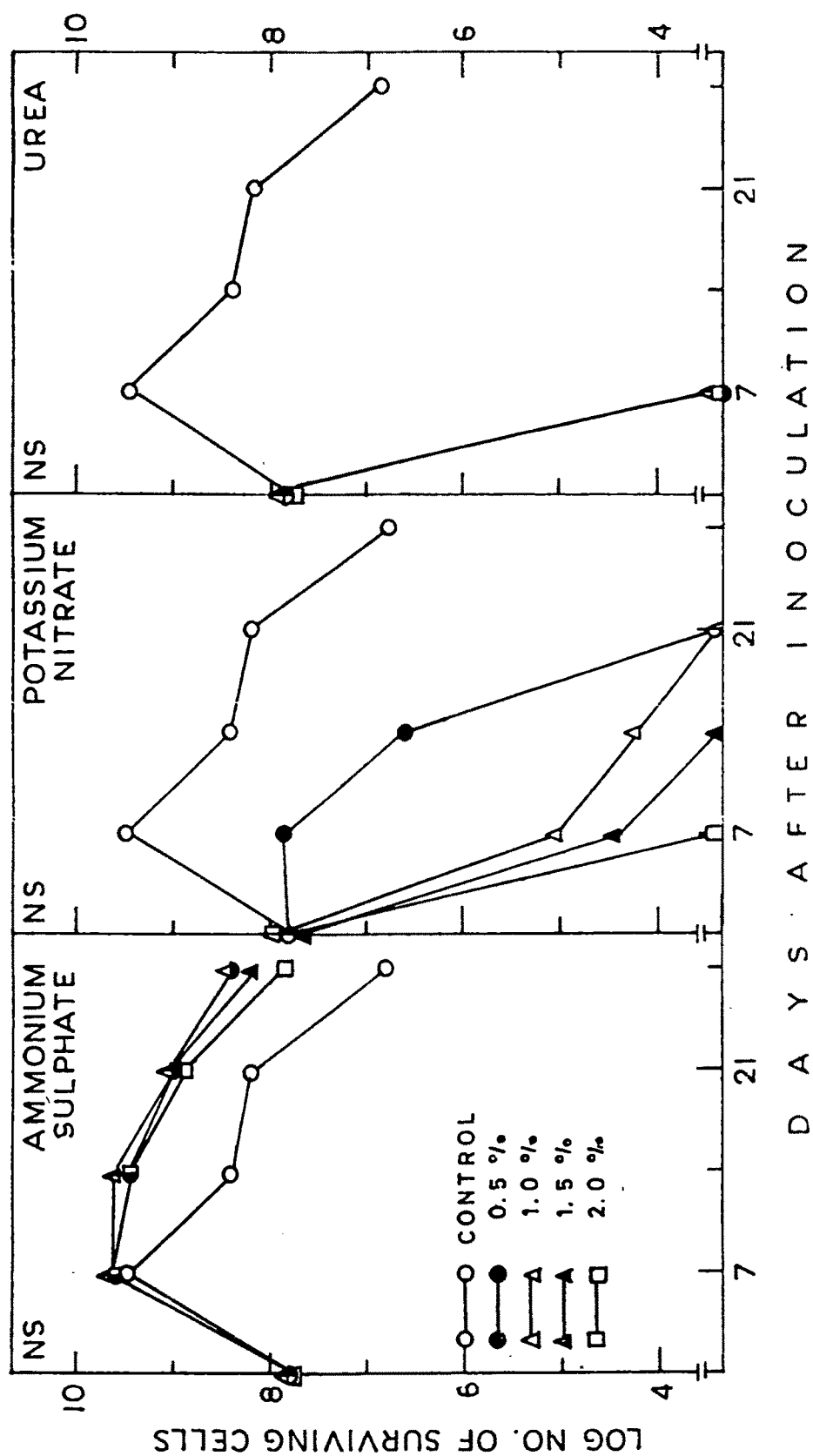


Figure 10. Effect of different doses of nitrogen fertilizers on the survival of the double marker strain of *P. solanacearum* in natural soils (NS) amended with fertilizers.

Table 21. Effects of amendment of natural^a soil with different doses of cowdung on the population dynamics of the marker strain of Pseudomonas solanacearum race 1.

Days after inoculation	Unamended soil cowdung not added	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with cowdung at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	7 (7.85)	4 (7.60)	5 (7.70)	4 (7.60)	5 (7.70)
7	315 (9.50)	600 (9.78)	521 (9.72)	483 (9.68)	190 (9.28)
14	28 (8.45)	370 (9.57)	220 (9.34)	199 (9.30)	25 (8.40)
21	21 (8.32)	13 (8.11)	25 (8.40)	3 (7.48)	1 (7.0)
28	0.9 (6.95)	0.9 (6.95)	1 (7.0)	0.3 (6.48)	0.1 (6.0)

^a Non-sterile natural soil described in Materials & Methods

^b Number of colonies were recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentration (wt/wt) of cowdung amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

Table 22. Effects of amendment of natural^a soil with different doses of mustard cake on the population dynamics of the marker strain of Pseudomonas solanacearum race 1

Days after inoculation	Unamended soil Mustard cake not added	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with mustard cake at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	7 (7.85) ^d	6 (7.78)	5 (7.70)	7 (7.85)	9 (7.95)
7	388 (9.48)	425 (9.63)	435 (9.64)	429 (9.63)	425 (9.63)
14	19 (8.28)	399 (9.60)	403 (9.61)	400 (9.60)	382 (9.58)
21	15 (8.18)	129 (9.11)	111 (9.05)	102 (9.01)	98 (8.99)
28	0.7 (6.85)	31 (8.49)	25 (8.40)	17 (8.23)	9 (7.95)

^a Non-sterile natural soil described in Materials & Methods

^b Number of colonies were recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml).

^c Percentage concentration (wt/wt) of mustard cake amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

Table 23. Effects of amendment of natural^a soil with different doses of mahua cake on the population dynamics of the marker strain of Pseudomonas solanacearum race 1

Days after inoculation	Unamended soil Mahua cake not added	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with mahua cake at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	7 (7.85)	4 (7.60)	5 (7.70)	6 (7.78)	4 (7.60)
7	315 (9.50)	448 (9.65)	539 (9.73)	541 (9.73)	550 (9.74)
14	23 (8.36)	480 (9.68)	610 (9.79)	600 (9.78)	600 (9.78)
21	17 (8.23)	452 (9.66)	572 (9.76)	578 (9.76)	582 (9.76)
28	0.8 (6.90)	416 (9.62)	516 (9.71)	519 (9.72)	519 (9.72)

^a Non-sterile natural soil as described in Materials and Methods

^b Number of colonies recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentration (wt/wt) of mahua cake amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate $\log$ transformation values.

Table 24. Effects of amendment of natural^a soil with different doses of linseed cake on the population dynamics of the marker strain of Pseudomonas solanacearum race 1

Days after inoculation	Unamended soil linseed cake not added	Number of colonies recovered ^b /g of soils ($\times 10^7$) amended with linseed cake at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	8 (7.90) ^d	7 (7.85)	7 (7.85)	6 (7.78)	7 (7.85)
7	312 (9.49)	290 (9.46)	435 (9.64)	435 (9.64)	759 (9.88)
14	23 (8.36)	255 (9.41)	405 (9.61)	432 (9.64)	739 (9.87)
21	19 (8.27)	140 (9.15)	235 (9.37)	279 (9.45)	499 (9.70)
28	0.7 (6.84)	35 (8.54)	99 (9.00)	124 (9.09)	365 (9.59)

^a Non-sterile natural soil as described in Materials and Methods

^b Number of colonies recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentration (wt/wt) of linseed cake amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

Table 25. Effects of amendment of natural^a soil with different doses of neem cake on the population dynamics of the marker strain of Pseudomonas solanacearum race 1

Days after inoculation	Unamended soil neem cake not added	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with neem cake at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	7 (7.85) ^d	9 (7.95)	7 (7.85)	5 (7.70)	4 (7.60)
7	312 (9.49)	84 (8.92)	93 (8.97)	402 (9.60)	421 (9.62)
14	27 (8.43)	82 (8.91)	85 (8.93)	103 (9.01)	95 (8.98)
21	19 (8.28)	59 (8.77)	67 (8.83)	76 (8.88)	72 (8.86)
28	0.7 (6.85)	36 (8.56)	41 (8.61)	53 (8.72)	63 (8.80)

^a Non-sterile natural soil as described in Materials and Methods

^b Number of colonies recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentration (wt/wt) of neem cake amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

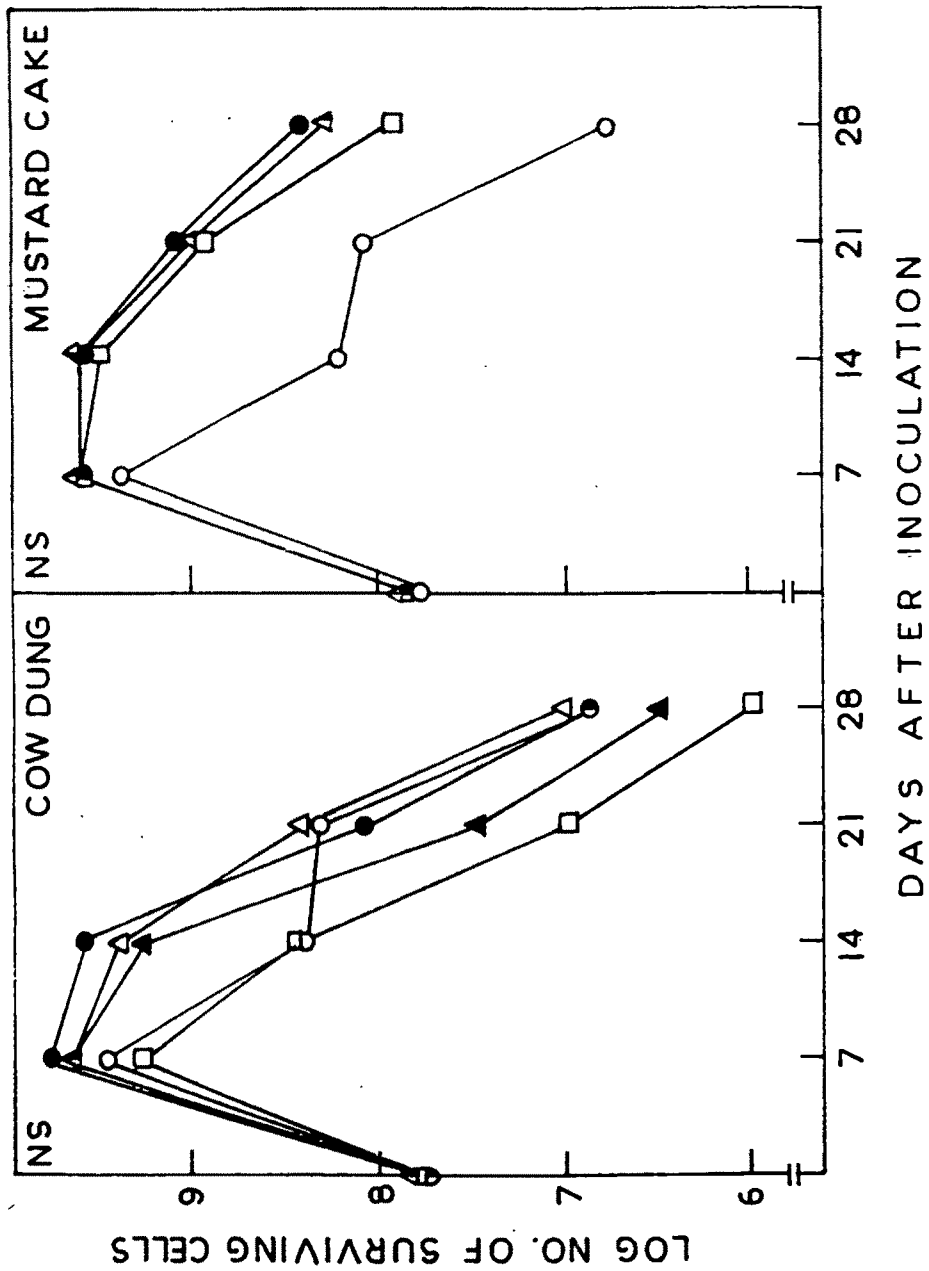


Figure 11. Effect of different doses of organic manures on the survival of the marker strain of *P. solanacearum* in natural soils (NS) amended with these manures.

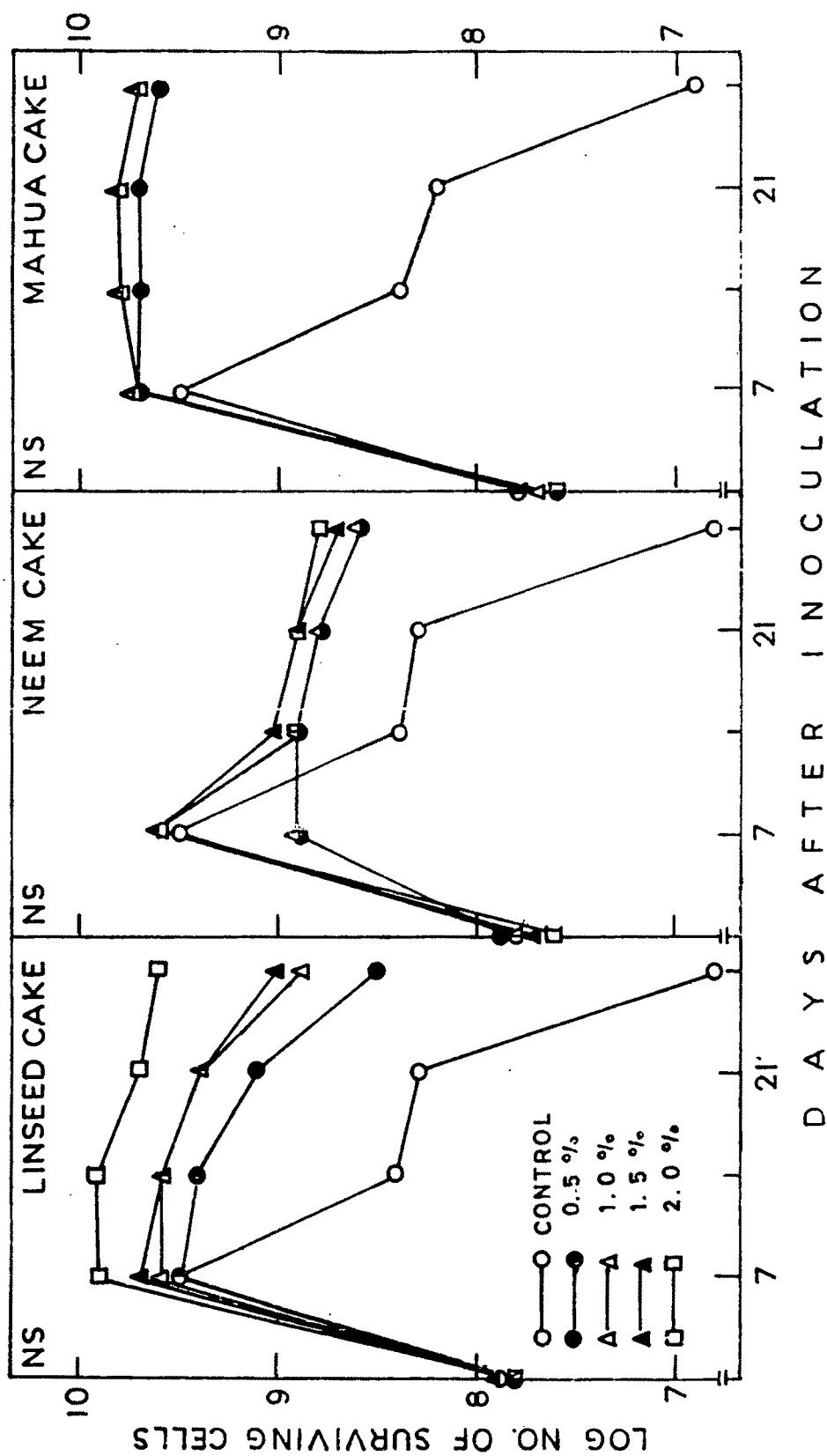


Figure 12. Effect of different doses of oilcakes on the survival of the marker strain of *P. solanacearum* in natural soils (NS) amended with these cakes.

receiving no amendments. In cowdung amended soils, the rate of decline of population density of the marker strain was comparable to that of the controlled soils (Figure 11). Of the oilcakes, Mahua and Neem caused good growth and survival of the marker strain as compared to linseed or mustard.

The data concerned with amendment of soil with host (eggplant) and non-host (cauliflower) leaf residues are presented in Tables 26 to 27 and Figure 13.

It is apparent from the results that amendment of soil with leaf residue irrespective of host and non-host origin, caused significant stimulation of growth and survival of the pathogen as compared to control soils.

Long-term survival in Soil. — In this series of experiments the long-term survival ability of the marker strain in natural and sterilized soil was investigated. The pathogen was inoculated to soil and the number of surviving cells were determined at monthly intervals using soil-dilution plate technique. The results are shown Table 28 and Figure 14.

It is evident from the results that under the experimental conditions employed, the marker strain could survive for 5 months in natural soil. In contrast, it survived for more than one year in sterilized soil.

Table 26. Effects of amendment of natural^a soil with different doses of eggplant leaf on the population dynamics of the marker strain of Pseudomonas solanacearum race 1

Days after inoculation	Unamended soil eggplant leaf not added	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with eggplant leaf at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	7 (7.85)	6 (7.78)	5 (7.70)	7 (7.85)	9 (7.95)
7	312 (9.49)	403 (9.61)	441 (9.64)	472 (9.67)	489 (9.69)
14	23 (8.36)	401 (9.60)	440 (9.64)	469 (9.67)	480 (9.68)
21	19 (8.28)	398 (9.60)	429 (9.63)	454 (9.66)	472 (9.67)
28	0.9 (6.95)	392 (9.59)	419 (9.62)	443 (9.65)	461 (9.66)

^a Non-sterile natural soil as described in Materials and Methods

^b Number of colonies recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentration (wt/wt) of eggplant leaf amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

Table 27. Effects of amendment of natural^a soil with different doses of cauliflower leaf on the population dynamics of the marker strain of Pseudomonas solanacearum race 1.

Days after inoculation	Unamended soil cauliflower leaf not added	Number of colonies recovered ^b /g of soil ($\times 10^7$) amended with cauliflower leaf at a conc. (%) ^c of			
		0.5	1.0	1.5	2.0
0	8 (7.90) ^d	6 (7.78)	7 (7.85)	5 (7.70)	6 (7.78)
7	315 (9.50)	336 (9.53)	381 (9.58)	390 (9.59)	393 (9.59)
14	23 (8.36)	399 (9.60)	423 (9.63)	427 (9.63)	452 (9.66)
21	19 (8.28)	95 (8.98)	97 (8.99)	92 (8.96)	115 (9.06)
28	0.8 (6.90)	70 (8.85)	82 (8.91)	80 (8.90)	99 (9.00)

^a Non-sterile natural soil as described in Materials and Methods

^b Number of colonies recovered after indicated days of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^c Percentage concentration (wt/wt) of cauliflower leaf amendment was calculated in terms of dry weight of soil

^d Figures in parentheses indicate log transformation values.

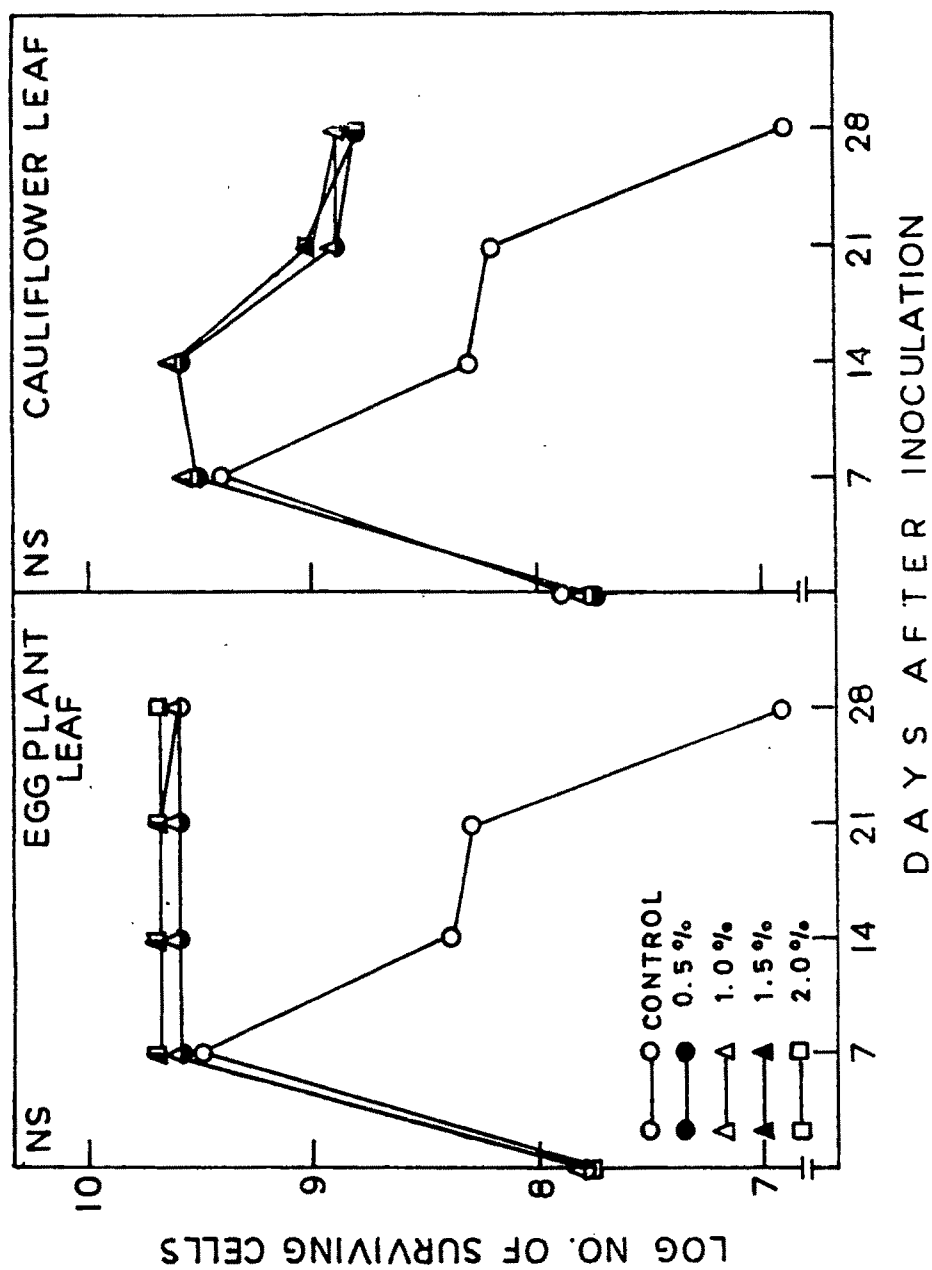


Figure 13. Effect of crop debris of host (eggplant) and nonhost (cauliflower) plants on the survival of the marker strain of *P. solanacearum* in natural soils (NS) amended with such debris.

Table 28. Long-term survival of marker strain of Pseudomonas solanacearum race 1 in natural and sterilized soils.

		Number of colonies recovered ^a /g of soil ($\times 10^7$) months after inoculation											
		0	1	2	4	6	8	10	12	13			
Natural ^b	5.00	8.00	0.006	0.00003	-	-	-	-	-	-			
	(7.70) ^d	(7.90)	(4.78)	(2.48)									
Sterilized ^c	7.00	96.00	82.00	40.00	1.0	0.05	0.0006	0.0002	-	-			
	(7.85)	(8.98)	(8.91)	(8.60)	(7.0)	(5.70)	(3.78)	(3.30)					

^a Number of colonies were recovered after indicated months of intervals on PDA containing streptomycin (2 mg/ml) and erythromycin (1 mg/ml)

^b Non-sterile natural soil as described in Materials and Methods

^c Soil samples were sterilized at 15 p.s.i. for 1 hr on three successive days

^d Figure in parentheses indicate log transformation values.

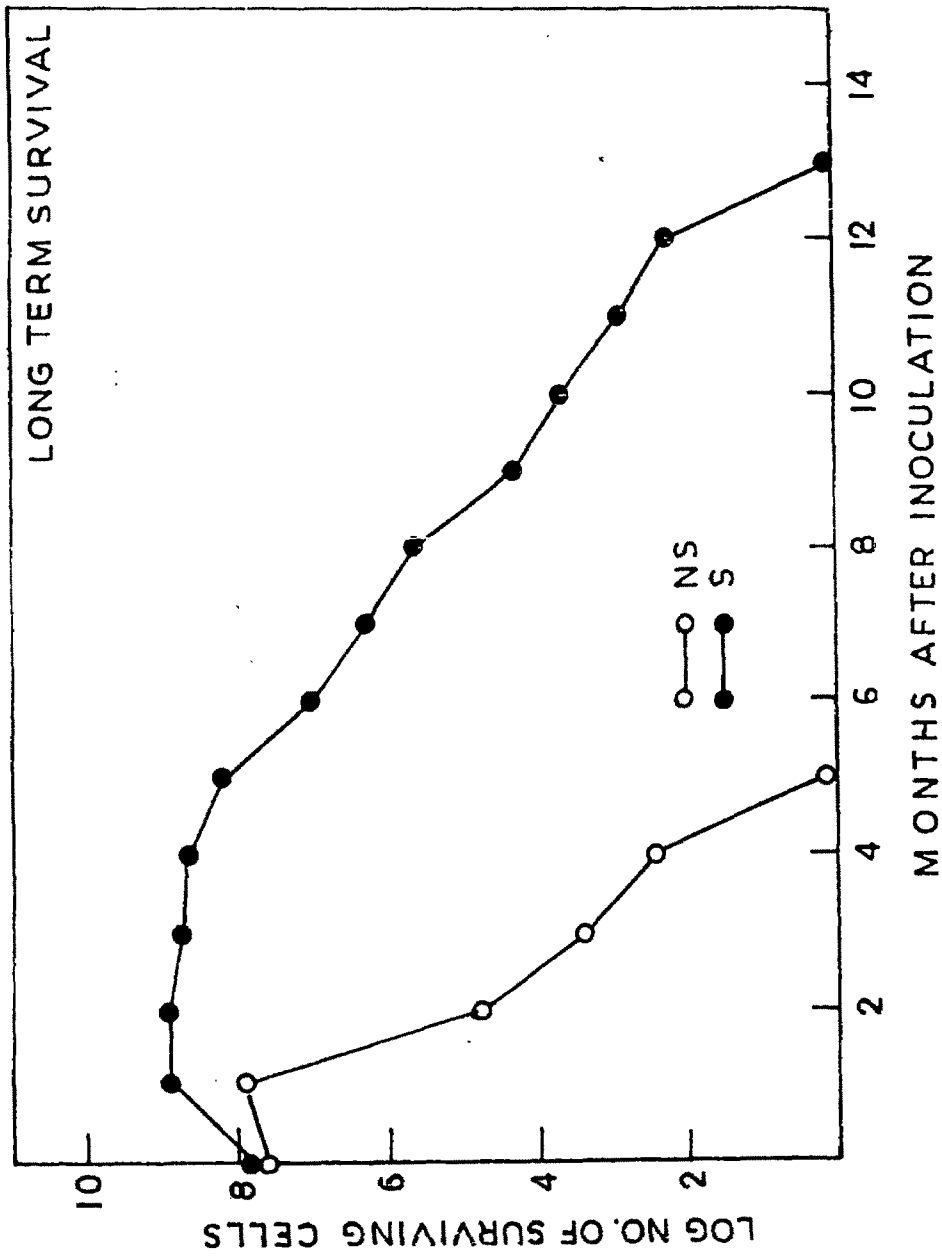


Figure 14. Comparison of the long-term survival of the double marker strain in natural (NS) and sterilized soils (S) determined as a function of months of incubation.

SECTION II : CONTROL OF EGGPLANT WILT

Control of vegetable wilt including eggplant caused by P. solanacearum is a serious problem all over the world. In this series of experiments attempts were made to control the wilt of eggplant caused by P. solanacearum employing varietal resistance, cultural and chemical approaches.

Varietal resistance. — Sixty three Indian germplasms including the highly susceptible Pusa Purple Long and 18 Bangladeshi germplasms were screened for their resistance to bacterial wilt under natural and artificial inoculation conditions in the field. Germplasms without any established commercial names were designated by the code numbers provided by the source authority. Indian germplasms were obtained from collections of IARI, New Delhi, G.B. Pant University of Agriculture and Technology, Pantnagar, U.P., Krishnagar Horticultural Research Station, West Bengal, Department of Horticulture, BCKV and Sutton & Sons Company, Calcutta. The Bangladeshi germplasms were supplied by BARI, Dhaka.

The natural wilt incidence of the germplasms were tested in experimental plots of Kalyani and Mondouri farms of BCKV. The germplasms were sown in replicate plots which had previous histories of eggplant cropping and significant incidence of bacterial wilt in three previous years. The percentage of wilted plants in each plot was determined after 80 days of transplantation. The results are presented in Table 29.

It is evident from the results that the cultivar Pusa

Table 29. Screening for varietal resistance of different germ-plasms of eggplant under natural conditions at Kalyani and Mondouri farms as kharif crop in 1986.

Code Number	Name of the germplasms	Percentage of plants showing wilt at		
		Kalyani	Mondouri	Average
(1)	(2)	(3)	(4)	(5)
IG-1 ^a	Pusa Purple Long	37.50 (37.75) ^b	20.00 (26.40)	28.75 (32.43)
IG-2	Banaras Giant Green	22.50 (28.29)	15.00 (22.50)	18.75 (25.66)
IG-3	Pusa Kranti	12.50 (20.61)	17.50 (24.68)	15.00 (22.79)
IG-4	Pusa Purple Cluster	-	2.50 (9.10)	-
IG-5	Improved Mukta-Keshi	-	2.50 (9.10)	-
IG-6	Nurki	15.00 (22.50)	22.50 (20.61)	13.75 (21.77)
IG-7	K-812	7.50 (15.68)	10.00 (18.43)	8.75 (17.21)
IG-8	Kanta Brinjal	25.00 (29.89)	17.50 (24.68)	21.25 (27.46)
IG-9	SM 65(1)	22.50 (28.00)	17.50 (24.22)	20.00 (26.57)
IG-10	SM 65(2)	7.50 (15.68)	-	-
IG-11	T-3	7.50 (15.68)	5.00 (9.22)	6.25 (14.48)
IG-12	KG Local	20.00 (26.57)	12.50 (20.61)	16.25 (23.77)

Table 29. Screening for varietal resistance of different germ-plasms of eggplant under natural conditions at Kalyani and Mondouri farms as kharif crop in 1986

(1)	(2)	(3)	(4)	(5)
IG-13	SM 50 (A)	22.50 (28.00)	15.00 (22.50)	18.75 (25.66)
IG-14	Long-(1)	2.50 (6.46)	-	-
IG-15	K-8(1)	2.50 (6.46)	-	-
IG-16	Pant Rituraj	22.50 (28.29)	25.00 (29.89)	23.75 (29.17)
IG-17	S-62(1)	5.00 (9.22)	7.50 (15.68)	6.25 (14.48)
IG-18	Karimpur	15.00 (12.92)	-	-
IG-19	Bhangar-1	7.50 (15.68)	-	-
IG-20	H-4(1)	20.00 (26.40)	17.50 (24.68)	18.75 (25.66)
IG-21	SM-227	-	5.00 (12.92)	-
IG-22	SM-48	17.50 (24.68)	15.00 (22.50)	16.25 (23.77)
IG-23	Salimakra	22.50 (28.29)	15.00 (22.50)	18.75 (25.66)
IG-24	Kalo Chaila	25.00 (29.89)	17.50 (24.68)	21.25 (27.46)
IG-25	SM-71	22.50 (28.29)	20.00 (26.57)	21.25 (27.46)
IG-26	Muktakeshi-1	25.00 (29.89)	20.00 (26.40)	22.50 (28.32)

Table 29. Screening for varietal resistance of different germ
 plasma of eggplant under natural conditions at Kalyani
 and Mondouri farms as kharif crop in 1986

(1)	(2)	(3)	(4)	(5)
IG-27	Garia	2.50 (6.46)	7.50 (15.68)	5.00 (12.92)
IG-28	Type 3(3) Long	-	5.00 (12.92)	-
IG-29	Gourigazar	22.50 (28.29)	12.50 (20.61)	17.50 (24.73)
IG-30	Pusakesar	20.00 (26.40)	15.00 (22.50)	17.50 (24.73)
IG-31	Type 3(2) Long	20.00 (26.57)	17.50 (24.68)	18.75 (25.66)
IG-32	Green Long	2.50 (6.46)	5.00 (9.22)	3.75 (11.17)
IG-33	Muktakeshi-II	20.00 (26.40)	15.00 (22.50)	17.50 (24.73)
IG-34	Black Round	15.00 (22.79)	22.50 (28.29)	18.75 (25.66)
IG-35	Katamakra	17.50 (24.22)	17.50 (24.68)	17.50 (24.73)
IG-36	Silimum	-	5.00 (12.92)	-
IG-37	PPR-7	15.00 (22.50)	20.00 (26.57)	17.50 (24.73)
IG-38	B-5	7.50 (15.68)	2.50 (6.46)	5.00 (12.92)
IG-39	Guzrat	22.50 (28.29)	17.50 (24.68)	20.00 (26.57)
IG-40	Long White	5.00 (12.92)	5.00 (9.22)	5.00 (12.92)
IG-41	Mysore	7.50 (15.68)	7.50 (11.40)	7.50 (15.89)
IG-42	Siliguri (A)	22.50 (28.29)	20.00 (26.57)	21.25 (27.46)

Contd..

Table 29. Screening for varietal resistance of different germ plasms of eggplant under natural conditions at Kalyani and Mondouri farms as kharif crop in 1986

(1)	(2)	(3)	(4)	(5)
IG-43	K-3	25.00 (29.89)	15.00 (22.79)	20.00 (26.57)
IG-44	Azad Kranti	25.00 (30.00)	22.50 (28.29)	23.75 (29.17)
IG-45	Dharampur Local	-	10.00 (18.43)	-
IG-46	K-2	17.50 (24.22)	17.50 (24.68)	17.50 (24.73)
IG-47	SM-31	-	7.50 (15.68)	-
IG-48	SM-273	17.50 (24.68)	15.00 (22.79)	16.25 (23.77)
IG-49	Bhangar-II	7.50 (15.68)	10.00 (17.86)	8.75 (17.21)
IG-50	White Brinjal	2.50 (6.46)	10.00 (13.29)	6.25 (14.48)
IG-51	Paichabani	-	7.50 (11.40)	-
IG-52	KNG-1	2.50 (6.46)	5.00 (9.22)	3.75 (11.17)
IG-53	Pant Samrat	27.50 (31.42)	17.50 (24.68)	22.50 (28.32)
IG-54	H-8	27.50 (28.29)	20.00 (26.57)	23.75 (29.17)
IG-55	Makra	15.00 (22.79)	20.00 (26.40)	17.50 (24.73)
IG-56	Monjorigota	-	7.50 (11.40)	-
IG-57	Purulia Purple	17.50 (24.68)	20.00 (26.40)	18.75 (25.66)
IG-58	PBR-91-2	15.00 (22.79)	22.50 (28.29)	18.75 (25.66)
IG-59	Muktakeshi-III	22.50 (28.29)	22.50 (28.29)	22.50 (28.32)

Table 29. Screening for varietal resistance of different germ
 plasm of eggplant under natural conditions at Kalyani
 and Mondouri farms as kharif crop in 1986

(1)	(2)	(3)	(4)	(5)
IG-60	KNG-II	32.50 (34.74)	25.00 (30.00)	28.75 (32.43)
IG-61	KT-4	22.50 (28.29)	17.50 (24.68)	20.00 (26.57)
IG-62	S-54	17.50 (24.68)	27.50 (31.61)	22.50 (28.32)
IG-63	Patakanta	25.00 (29.89)	25.00 (29.89)	25.00 (30.00)
BG-I ^c	D.R.Chowdhury	15.00 (22.79)	25.00 (29.89)	20.00 (26.57)
BG-2	Black Long	17.50 (24.68)	15.00 (22.79)	16.25 (23.71)
BG-3	Rangpur No.1	25.00 (29.89)	20.00 (26.57)	22.50 (28.32)
BG-4	Laljhuri	27.50 (31.42)	27.50 (31.61)	27.50 (21.63)
BG-5	Bhangar	17.50 (24.68)	17.50 (24.68)	17.50 (24.73)
BG-6	Black Acc	17.50 (24.22)	17.50 (24.22)	17.50 (24.73)
BG-7	Rajshahi No.6	2.50 (6.46)	-	-
BG-8	Dohazari	2.50 (6.46)	5.00 (12.92)	3.75 (11.17)
BG-9	Baromashi	17.50 (24.68)	22.50 (28.00)	20.00 (26.57)
BG-10	Rajshahi No.9	10.00 (17.89)	10.00 (17.86)	10.00 (18.43)

Table 29. Screening for varietal resistance of different germ plasms of eggplant under natural conditions at Kalyani and Mondouri farms as kharif crop in 1986

(1)	(2)	(3)	(4)	(5)
BG-11	Uttara	5.00 (9.22)	7.50 (15.68)	6.25 (14.48)
BG-12	Muktakeshi Round	22.50 (28.29)	17.50 (24.68)	20.00 (26.57)
BG-13	Khatkhatia	-	5.00 (12.92)	-
BG-14	Longla	25.00 (29.89)	22.50 (28.29)	23.75 (29.17)
BG-15	Islampuri	7.50 (15.68)	5.00 (9.22)	6.25 (14.48)
BG-16	Singnath	2.50 (6.46)	10.00 (18.43)	6.25 (14.48)
BG-17	Chittagong Local	5.00 (9.22)	7.50 (15.68)	6.25 (14.48)
BG-18	Sada Jhumki	22.50 (28.29)	25.00 (29.89)	23.75 (29.17)

C.D. at 5%		10.02	11.97	

^a Indian germplasms

^b Figures in parentheses indicate angular transformation values

^c Bangladeshi germplasms.

Purple Long was highly susceptible and exhibited a mean wilt percentage of 26.75. A total of 45 other germplasms showed more than 15% wilt incidence under natural conditions of growing the crop. The rest 35 germplasms showing less than 15% wilt incidence were considered as putative resistant. These were further subjected to field trials under artificial inoculation conditions. It may further be noted from the results (Table 29) that the resistance of the test germplasms were highly variable and the wilt incidence ranged from 2.50% to 37.50%. It is also of interest that the varieties Long-1 (IG-14), K-8(1) [IG-15_7, Garia (IG-27), Green Long (IG-32) and KNG-1 (IG-52) of India and varieties Rajshahi No. 6 (BG-7), Dohazari (BG-8) and Singnath (BG-16) of Bangladesh showed high degree of resistance (2.50% wilt) under natural conditions of cultivation.

For resistance screening, following artificial inoculations, 35 germplasms selected from the field screening (Table 29) were used. The cultivar Pusa Purple Long was also used as susceptible control.

Resistance or tolerance of these germplasms to P. solanacearum race 1 were tested in Kharif (rainy) and rabi(winter) seasons of cultivation. Seedlings were raised in seedbeds and 5-week-old seedlings were transplanted to replicate experimental plots following inoculation by root clipping method. All seedlings received a second inoculation by soil drenching method after 30 days of transplantation. Wilt incidence was recorded after 80 days of transplantation. Cultivars showing more than 20% of wilt incidence in either of the cropping seasons were considered as

susceptible. The results are presented in Table 3U.

It is evident from the data that none of the 35 germplasms including Pusa Purple Cluster which showed resistance in several earlier trials exhibited resistance under artificial inoculation conditions. The data further showed high susceptibility of Pusa Purple Long having 66.67% to 76.67% wilt incidence in both the seasons of cultivation. Cultivars Pusa Purple Cluster, Improved Muktakeshi, Khatkhatia and Type 3(3) Long, however, showed moderate resistance because the wilt incidence was, in general, lower than (less than 35%) other cultivars.

Cultural control. — Cultural control of the disease under field conditions was investigated. Effects of different doses of N:P:K fertilizers and oilcakes and different regimes of irrigation schedules on the incidence of wilt in experimental plots were examined. Wilt incidence was determined at pre-topdressing and final harvest time. The other growth and yield parameters were also evaluated.

Effects of N:P:K fertilizers on wilt incidence. — In this series of experiments the incidence of eggplant wilt and other parameters of loss assessment were investigated in the experimental plots amended with different doses of N, P and K fertilizers. Three doses of each N, P and K in combination comprising a total of 27 treatments were examined and the effect of different doses of fertilizers on disease severity under field conditions was investigated. The dose combinations of the fertilizers have been shown in Table 3 and the parameters evaluated were given

Table 30. Screening for resistance of selected germplasms of eggplant in field plots of Mondouri following artificial inoculation with race 1 of P. solanacearum

Code Number	Name of germplasms	Percentage of wilt incidence in growing seasons of		
		Kharif (June-Oct. '87)	Rabi (Nov. Mar. 87-88)	Mean values
IG-1 ^a	Pusa Purple Long	76.67 (61.22) ^b	66.67 (55.08)	71.67 (58.07)
IG-3	Pusa Kranti	56.67 (48.85)	50.00 (45.00)	53.34 (46.92)
IG-4	Pusa Purple Cluster	23.33 (28.78)	30.00 (33.00)	26.67 (31.07)
IG-5	Improved Mukta-keshi	16.67 (23.86)	23.33 (28.78)	20.00 (26.45)
IG-6	Nurki	60.00 (50.85)	63.33 (52.86)	61.67 (51.81)
IG-7	K-812	70.00 (57.00)	53.33 (46.92)	61.67 (51.76)
IG-10	SM 65(2)	53.33 (47.01)	40.00 (39.15)	46.67 (43.06)
IG-11	T-3	73.33 (59.21)	56.67 (48.93)	65.00 (53.73)
IG-14	Long-I	86.67 (68.86)	66.67 (54.78)	76.67 (61.22)
IG-15	K-8(1)	56.67 (48.85)	50.00 (45.00)	53.33 (46.92)
IG-17	S-62(1)	63.33 (52.78)	50.00 (45.00)	56.67 (48.84)
IG-18	Karimpur	40.00 (39.15)	46.67 (42.99)	43.34 (41.12)
IG-19	Bhanger-I	70.00 (57.00)	60.00 (50.85)	65.00 (53.76)

Table 30. Screening for resistance of selected germplasms of eggplant in field plots of Mondouri following artificial inoculation with race 1 of P. solanacearum

(1)	(2)	(3)	(4)	(5)
IG-21	SM-227	43.33 (41.15)	53.33 (46.92)	48.33 (44.04)
IG-27	Garia	66.67 (54.78)	60.00 (50.85)	63.34 (52.78)
IG-28	Type 3(3) Long	20.00 (26.67)	26.67 (30.79)	23.33 (28.54)
IG-32	Green Long	80.00 (63.43)	56.67 (48.85)	68.34 (55.77)
IG-36	Silimum	50.00 (45.00)	43.35 (41.07)	46.67 (43.06)
IG-38	B-5	60.00 (50.85)	56.67 (48.85)	58.34 (49.83)
IG-40	Long White	33.33 (35.22)	46.67 (43.08)	40.00 (39.21)
IG-41	Mysore	36.67 (37.22)	40.00 (39.15)	58.33 (38.19)
IG-45	Dharampur Local	76.67 (61.22)	53.33 (49.92)	65.00 (53.76)
IG-47	SM-31	53.33 (46.92)	40.00 (39.15)	46.67 (43.09)

Table 30. Screening for resistance of selected germplasms of eggplant in field plots of Mondouri following artificial inoculation with race 1 of P. solanacearum

(1)	(2)	(3)	(4)	(5)
IG-49	Bhangar-II	63.33 (52.78)	50.00 (45.00)	56.67 (48.84)
IG-50	White Brinjal	46.67 (43.08)	53.33 (47.01)	50.00 (45.03)
IG-51	Paichabani	80.00 (63.93)	63.33 (52.78)	71.6 (57.91)
IG-52	KNG-1	70.00 (57.00)	56.67 (48.85)	63.33 (52.80)
IG-56	Monjorigota	63.33 (52.78)	53.33 (47.01)	58.33 (49.82)
BG-7 ^c	Rajshahi No.6	33.33 (35.22)	40.00 (39.15)	36.67 (37.20)
BG-8	Dohazari	66.67 (54.78)	63.33 (52.86)	65.00 (53.76)
BG-10	Rajshahi No.9	60.00 (50.85)	53.33 (46.92)	56.67 (48.87)
Bg-11	Uttara	40.00 (39.15)	33.33 (35.01)	36.67 (37.12)
BG-13	Khatkhatia	26.67 (31.00)	33.33 (35.22)	30.00 (33.16)
BG-15	Islampur	46.67 (43.08)	56.67 (48.85)	51.67 (45.96)
BG-16	Singnath	43.33 (41.15)	56.67 (48.85)	50.00 (45.00)
BG-17	Chittagong Local	86.67 (68.86)	73.33 (59.00)	80.00 (63.55)

C.D.at 5%		12.76	9.48	6.77

^a Indian germplasm

^b Figures in parentheses indicate angular transformation values

^c Bangladeshi germplasms.

in Materials and Methods. The experiments were conducted in 1988 and repeated in 1989. For each treatment 3 replicate plots were used following Randomised Block Design technique. Eggplant seedlings of cultivar Improved Muktakeshi were inoculated with the pathogen by root clipping technique and transplanted to the treatment plots. All seedlings were reinoculated by soil drenching method 30 days after transplanting as described in Materials and Methods. Incidence of wilt and other parameters were assessed 45 days after transplanting and also at final harvest time. The results are presented in Tables 31 to 39. The results are expressed as mean of the 3 replicate plots for each treatment for the year 1988 and 1989. The mean values of two years' data for the year 1988 and 1989 are also included for comparison with the individual year.

It may be noted from the data that increase in nitrogen concentrations caused significant decrease in wilt incidence in both the years of experimentation. The mean values of two years' data also reflected the decreasing effect of increased doses of nitrogen on pre-topdressing wilt incidence. Increased doses of phosphorus showed significant decrease of wilt incidence during 1988 but not in 1989. All other treatment groups except N:K exhibited no significant effect on pre-topdressing wilt incidence. Increased doses of N:K showed significant reduction of wilt incidence during 1988 but not in 1989 (Table 31).

It may be noted from the final harvest time wilt incidence data that increase in nitrogen concentrations caused significant decrease in wilt incidence in both the years of 1988 and 1989.

Table 31. Effect of amendment of soil with different doses of fertilizers on pre-topdressing^a wilt incidence of eggplant inoculated with *Pseudomonas solanacearum* race 4.

Treatments Doses of N and P ₂ O ₅ kg/ha		Percentage of wilted plants in the field												
		Doses of K ₂ O (kg/ha)												
		1988				1989				Mean values of 2 years' data				
		40	60	80	Mean	40	60	80	Mean	40	60	80	Mean	
60	P ₂ O ₅	40	48.33 (44.06) ^b	35.00 (36.22)	31.67 (34.22)	38.33 (38.17)	51.67 (45.97)	48.33 (44.03)	41.67 (40.18)	47.22 (43.39)	50.00 (45.00)	41.67 (40.19)	36.67 (37.26)	42.78 (40.82)
		60	38.33 (38.25)	33.33 (35.25)	33.33 (35.25)	35.00 (36.25)	46.67 (43.08)	50.00 (45.00)	40.00 (39.21)	45.56 (42.43)	42.50 (40.68)	41.67 (40.20)	36.67 (37.25)	40.28 (39.38)
		80	41.67 (40.18)	33.33 (35.25)	31.67 (34.16)	35.56 (36.53)	45.00 (42.09)	36.67 (37.22)	33.33 (31.19)	40.00 (39.17)	43.33 (41.41)	35.00 (36.24)	35.00 (37.86)	37.78 (37.86)
		Mean	42.78 (40.83)	33.89 (35.57)	32.22 (34.54)	36.30 (36.98)	47.78 (43.71)	45.00 (42.08)	40.00 (39.19)	44.26 (41.66)	45.28 (42.27)	39.45 (38.88)	36.11 (36.91)	40.28 (39.35)
90	P ₂ O ₅	40	33.33 (35.25)	41.67 (40.17)	33.33 (35.22)	36.11 (36.86)	43.33 (41.12)	43.33 (41.12)	38.33 (38.19)	41.66 (40.14)	38.33 (38.22)	42.50 (40.65)	35.83 (36.72)	38.29 (38.53)
		60	36.67 (37.22)	35.00 (36.15)	30.00 (33.16)	33.89 (35.51)	43.33 (41.15)	38.33 (38.22)	38.33 (38.19)	40.00 (39.19)	40.00 (39.23)	36.67 (37.26)	34.17 (35.73)	36.95 (37.41)
		80	30.00 (33.16)	35.00 (36.24)	35.00 (36.24)	33.33 (35.21)	38.33 (38.19)	45.00 (42.09)	46.67 (43.08)	43.33 (41.12)	34.17 (35.76)	40.00 (39.23)	40.83 (39.69)	38.33 (38.23)
		Mean	33.33 (35.21)	37.22 (37.52)	32.78 (34.87)	34.44 (35.87)	41.66 (40.15)	42.22 (40.48)	41.11 (39.82)	41.66 (40.15)	37.50 (37.74)	39.72 (39.05)	36.95 (37.38)	38.06 (38.06)
120	P ₂ O ₅	40	31.67 (34.16)	40.00 (39.21)	38.33 (38.24)	36.67 (37.20)	40.00 (39.15)	38.33 (38.24)	41.67 (40.20)	40.00 (39.20)	35.83 (36.70)	39.17 (38.74)	40.00 (39.23)	38.33 (38.23)
		60	31.67 (34.16)	28.33 (32.14)	31.67 (34.23)	30.56 (33.51)	35.00 (36.24)	31.67 (34.23)	36.67 (37.26)	34.50 (35.91)	33.33 (35.22)	30.00 (33.21)	34.17 (35.75)	32.50 (34.73)
		80	31.67 (34.16)	28.33 (32.14)	30.00 (33.21)	30.00 (33.17)	31.67 (34.18)	30.00 (33.21)	36.67 (37.22)	32.78 (34.87)	31.67 (34.18)	29.17 (32.67)	33.33 (35.24)	31.39 (34.03)
		Mean	31.67 (34.16)	32.22 (34.50)	33.33 (35.23)	32.41 (34.63)	35.56 (36.52)	33.33 (35.23)	30.34 (30.23)	35.74 (36.66)	33.61 (35.37)	32.77 (34.87)	35.83 (36.74)	34.07 (35.66)
Sources of variation		C.D. of 1988 data at 5%				C.D. of 1989 data at 5%				C.D. of mean values of two years' data at 5%				
N		1.69				2.26				1.48				
P		1.69				NS				1.48				
K		NS				NS				NS				
N X P		NS				NS				NS				
N X K		2.92				NS				2.57				
P X K		NS				NS				NS				
N X P X K		NS				NS				NS				

^a Topdressing of experimental plots with urea at the rate of 45 kg/ha was done 45 days after transplanting

^b Figures in parentheses indicate angular transformation values.

The mean values of two years' data also reflected the decreasing effect of increased doses of nitrogen, phosphorus and potassium on final harvest incidence. Increased doses of phosphorus showed significant decrease of wilt incidence during 1988, but not in 1989. All other treatment groups exhibited no significant effect on the incidence of wilt at final harvest time (Table 32).

It was apparent from the data that increase in nitrogen concentrations caused significant increase in height of plants in both years of experimentation. The mean values of two years' data also reflected the increasing effect of increased doses of nitrogen, phosphorus and potassium on height (cm) of plants of eggplant. All other treatments exhibited no significant effect on height of plants of eggplant (Table 33).

It was also concluded that increase in nitrogen concentrations caused significant increase of the number of primary branches per plant in both years of experimentations. Increased doses of phosphorus showed significant increase on the number of primary branches per plant during 1989 but not in 1988. The mean values of two years' data also reflected the increasing effect of increased doses of nitrogen and phosphorus on the number of primary branches per plant. All other treatment groups exhibited no significant effect on the number of primary branches per plant (Table 34).

It was also evident from the data that increase in nitrogen concentrations caused significant increase of mean diameter of fruits (cm) in both years of experimentation. The mean values

Table 32. Effect of amendment of soil with different doses of fertilizers on final harvest time^a wilt incidence of eggplant inoculated with *Pseudomonas solanacearum* race 1

Treatments Doses of N and P ₂ O ₅ kg/ha		Percentage of wilted plants in the field												
		Doses of K ₂ O (kg/ha)												
		1988					1989					Mean values of two years' data		
N	P ₂ O ₅	40	60	80	Mean	40	60	80	Mean	40	60	80	Mean	
60	40	66.67 (54.78)	60.00 (50.82)	65.00 (53.75)	63.89 (53.12)	71.67 (57.91)	66.67 (54.78)	61.67 (51.84)	66.67 (54.84)	59.17 (56.33)	63.33 (52.78)	63.33 (52.74)	65.28 (53.95)	
	60	55.00 (47.88)	51.67 (45.96)	46.67 (43.07)	51.11 (45.64)	63.33 (52.74)	61.67 (51.0)	55.00 (47.88)	60.00 (50.84)	59.17 (50.29)	56.67 (48.85)	50.83 (45.48)	55.56 (48.21)	
	80	58.33 (49.83)	51.67 (45.96)	48.33 (44.04)	52.78 (46.61)	66.67 (54.89)	65.00 (53.76)	61.67 (51.90)	64.45 (53.52)	62.50 (52.25)	58.33 (49.80)	55.00 (47.91)	58.61 (49.99)	
	Mean	60.00 (50.83)	54.44 (47.58)	53.33 (46.95)	55.92 (48.45)	67.22 (55.18)	64.45 (53.48)	59.45 (50.54)	63.71 (53.07)	63.61 (52.96)	59.45 (50.48)	56.39 (48.71)	59.82 (50.72)	
90	40	48.33 (44.03)	50.00 (44.99)	43.33 (41.12)	47.22 (43.38)	65.00 (53.76)	61.67 (51.81)	53.33 (46.92)	60.00 (50.83)	56.67 (48.45)	55.83 (48.37)	48.33 (44.04)	53.60 (47.09)	
	60	50.00 (45.00)	48.33 (44.04)	45.00 (42.09)	47.78 (43.71)	53.33 (47.02)	58.33 (49.89)	51.67 (46.02)	54.44 (47.64)	51.67 (45.96)	53.33 (46.96)	48.33 (44.00)	51.11 (45.64)	
	80	40.33 (44.04)	45.00 (42.12)	41.67 (40.18)	45.00 (42.11)	56.67 (48.87)	55.00 (47.96)	48.33 (43.95)	53.33 (46.93)	52.50 (46.44)	50.00 (45.00)	45.00 (42.11)	49.17 (44.54)	
	Mean	48.89 (44.36)	47.78 (43.72)	43.33 (41.13)	46.67 (43.07)	58.33 (49.68)	58.33 (49.89)	51.11 (45.63)	55.92 (48.47)	53.61 (47.08)	53.05 (46.78)	47.22 (43.39)	51.29 (45.75)	
120	40	38.33 (38.24)	43.33 (41.16)	38.33 (38.22)	40.00 (39.21)	51.67 (45.96)	48.33 (44.06)	43.33 (41.10)	47.78 (43.71)	45.00 (42.12)	45.83 (42.60)	40.83 (39.66)	43.89 (41.46)	
	60	38.33 (38.22)	40.00 (39.23)	40.00 (39.15)	39.44 (38.89)	46.67 (43.08)	48.33 (44.06)	41.67 (40.14)	45.56 (42.43)	42.50 (40.67)	44.17 (41.63)	40.83 (39.71)	42.50 (40.67)	
	80	40.00 (39.21)	36.67 (37.26)	38.33 (38.22)	38.33 (38.23)	45.00 (42.03)	40.00 (39.04)	40.00 (39.03)	41.67 (40.03)	42.50 (40.56)	38.33 (38.20)	39.17 (38.73)	40.00 (39.16)	
	Mean	38.89 (38.56)	40.00 (39.22)	38.89 (38.53)	39.26 (38.78)	47.78 (43.69)	45.55 (42.39)	41.67 (40.09)	45.00 (42.06)	43.33 (41.12)	42.78 (40.81)	40.28 (39.37)	42.13 (40.43)	

Sources of variation		C.D. of 1988 data at 5%		C.D. of 1989 data at 5%		C.D. of mean values of two years' data at 5%	
N	P	2.22	2.22	3.70	3.70	2.05	2.05
K		NS	NS	NS	NS	2.05	2.05
N X P		NS	NS	NS	NS	NS	NS
N X K		NS	NS	NS	NS	NS	NS
P X K		NS	NS	NS	NS	NS	NS
N X P X K		NS	NS	NS	NS	NS	NS

^a Wilt incidence data taken just before the final harvest of the fruits

^b Figures in parantheses indicate angular transformation values.

Table 33. Effect of amendment of soil with different doses of fertilizers on height (cm) of plants of eggplant inoculated with *Pseudomonas solanacearum* race 1

Treatments Doses of N and P ₂ O ₅ kg/ha		Doses of K ₂ O (kg/ha)											
		1988				1989				Mean values of two years' data			
		40	60	80	Mean	40	60	80	Mean	40	60	80	Mean
60	P ₂ O ₅	73.37	74.38	76.22	74.66	61.67	62.17	64.00	62.61	67.52	68.29	70.11	68.64
	40	73.30	74.32	76.56	74.73	61.15	62.43	64.67	62.75	67.23	68.38	70.62	68.74
	60	73.87	77.60	88.04	79.84	61.63	66.35	75.10	67.69	67.75	71.99	81.57	73.77
	80	73.51	75.43	80.27	76.40	61.48	63.65	67.92	64.35	67.50	69.55	74.10	70.38
	Mean												
90		80.57	82.92	87.38	83.62	66.18	70.11	74.87	70.39	73.38	76.52	81.12	77.01
	40	84.90	86.45	87.23	86.19	69.72	71.45	73.74	71.64	77.31	78.95	80.48	78.91
	60	85.67	87.93	99.41	91.00	71.00	75.77	84.14	76.97	78.34	81.85	91.77	83.99
	80	83.71	85.77	91.34	86.94	68.97	72.44	77.58	73.00	76.34	79.11	84.46	79.97
	Mean												
120		97.53	97.92	101.07	98.84	80.76	81.27	85.93	82.65	89.15	89.60	93.50	90.75
	40	97.88	101.23	102.15	100.42	81.00	85.43	86.33	84.25	89.44	93.33	94.24	92.34
	60	100.00	102.25	113.26	105.17	85.47	90.72	98.20	91.46	92.74	96.49	105.73	98.32
	80	98.47	100.47	105.49	101.48	82.41	85.81	90.15	86.12	90.44	93.14	97.82	93.80
	Mean												
Sources of variation		C.D. of 1988 data at 5%				C.D. of 1989 data at 5%				C.D. of mean values of two years data at 5%			
N		5.87				7.77				5.70			
P		NS				NS				5.70			
K		NS				NS				5.70			
N X P		NS				NS				NS			
N X K		NS				NS				NS			
P X K		NS				NS				NS			
N X P X K		NS				NS				NS			

Table 34. Effect of amendment of soil with different doses of fertilizers on the number of primary branches per eggplant inoculated with *Pseudomonas solanacearum* race 1

Treatments Doses of N and P ₂ O ₅ kg/ha		Doses of K ₂ O (kg/ha)														
		1988					1989					Mean values of two years' data				
		40	60	80	Mean		40	60	80	Mean	40	60	80	Mean		
N	P ₂ O ₅															
60	40	6.88	7.04	6.91	6.94	5.23	5.30	5.40	5.31	6.06	6.17	6.16	6.13			
	60	7.11	6.87	7.32	7.10	5.30	5.50	5.60	5.47	6.21	6.19	6.46	6.29			
	80	7.22	6.84	7.25	7.10	5.53	5.57	5.67	5.59	6.38	6.21	6.46	6.35			
	Mean	7.07	6.92	7.16	7.05	5.35	5.46	5.56	5.46	6.22	6.19	6.36	6.26			
90	40	7.09	7.29	7.39	7.26	5.46	5.66	5.07	5.66	6.28	6.49	6.64	6.47			
	60	7.46	7.48	7.61	7.52	5.77	5.78	6.20	5.92	6.62	6.63	6.91	6.72			
	80	7.82	7.39	7.42	7.54	6.20	6.23	6.27	6.23	7.01	6.82	6.85	6.89			
	Mean	7.46	7.39	7.47	7.44	5.81	5.89	6.11	5.94	6.62	6.65	6.80	6.69			
120	40	7.64	7.50	7.61	7.58	6.16	6.10	6.10	6.12	6.90	6.80	6.86	6.85			
	60	7.63	7.58	7.63	7.61	6.20	6.27	6.26	6.24	6.92	6.93	6.95	6.93			
	80	7.78	7.84	7.76	7.79	6.40	6.47	6.50	6.46	7.09	7.16	7.13	7.13			
	Mean	7.68	7.64	7.67	7.66	6.25	6.28	6.29	6.27	6.97	6.96	6.99	6.97			
Sources of variation		C.D. of 1988 data at 5%					C.D. of 1989 data at 5%					C.D. of mean values of two years' data at 5%				
N		0.21					0.24					0.23				
P		NS					0.24					0.23				
K		NS					NS					NS				
N X P		NS					NS					NS				
N X K		NS					NS					NS				
P X K		NS					NS					NS				
N X P X K		NS					NS					NS				

of two years' data also reflected the increasing effect of increased doses of nitrogen on mean diameter of fruits (cm). All other treatments exhibited no significant effect on mean diameter of fruits (cm) of eggplant (Table 35).

It may be noted from the data that increase in nitrogen concentrations caused significant increase in mean fruit weight (g) of eggplant in both years of experimentation. The mean values of two years' data also reflected the increasing effect of increased doses of nitrogen on mean fruit weight (g) of eggplant. All other treatments exhibited no significant effect on mean fruit weight (g) of eggplant (Table 36).

It was apparent from the data that increase in nitrogen and phosphorus concentrations caused significant increase of yield (kg) per plant in 1988 and 1989. The mean values of two years' data also reflected the increasing effect of increased doses of nitrogen and phosphorus on yield per plant of eggplant. All other treatments exhibited no significant effect on yield per plant of eggplant (Table 37).

It was also evident from the data that increase in nitrogen and phosphorus concentrations caused significant increase in total yield (kg) per plot in both years of experimentation. Increased doses of potassium also showed significant increase in total yields (kg) per plot during 1989, but not in 1988. The mean values of two years' data further reflected the increasing effects of increased doses of nitrogen, phosphorus and potassium on total yield per plot. All other treatments

Table 35. Effect of amendment of soil with different doses of fertilizers on mean diameter of fruit (cm) of eggplant inoculated with Pseudomonas solanacearum race 1

Treatments Doses of N and P ₂ O ₅ kg/ha		Doses of K ₂ O (kg/ha)														
		1988					1989					Mean values of two years' data				
		40	60	80	Mean	Mean	40	60	80	Mean	Mean	40	60	80	Mean	
40	P ₂ O ₅															
	40	7.24	7.23	7.22	7.23	5.86	5.90	5.91	5.89		6.55	6.57	6.57	6.56		
	60	7.30	7.16	7.36	7.27	5.92	5.95	6.00	5.96		6.61	6.56	6.68	6.62		
	80	7.16	7.43	7.53	7.37	5.93	6.11	6.25	6.10		6.55	6.77	6.89	6.74		
	Mean	7.23	7.27	7.37	7.29	5.90	5.99	6.05	5.98		6.57	6.63	6.71	6.64		
90	P ₂ O ₅															
	40	7.73	8.03	8.00	7.92	6.40	6.49	6.56	6.48		7.07	7.26	7.28	7.20		
	60	8.16	8.33	8.20	8.23	6.70	6.77	6.85	6.77		7.43	7.55	7.53	7.50		
	80	8.36	8.46	8.33	8.38	6.90	6.94	6.96	6.93		7.63	7.70	7.65	7.66		
	Mean	8.08	8.27	8.18	8.17	6.67	6.73	6.79	6.73		7.38	7.50	7.49	7.45		
120	P ₂ O ₅															
	40	8.50	8.53	8.63	8.55	7.07	7.10	7.15	7.11		7.79	7.82	7.89	7.83		
	60	8.66	8.90	8.96	8.84	7.20	7.39	7.45	7.35		7.93	8.15	8.21	8.10		
	80	9.00	8.95	8.96	8.97	7.50	7.52	7.55	7.52		8.25	8.24	8.26	8.25		
	Mean	8.72	8.79	8.85	8.79	7.26	7.34	7.38	7.33		7.99	8.07	8.12	8.06		
Sources of variation		C.D. of 1988 data at 5%					C.D. of 1989 data at 5%					C.D. of mean values of two years' data at 5%				
N		0.44					0.37					0.40				
P		NS					NS					NS				
K		NS					NS					NS				
N X P		NS					NS					NS				
N X K		NS					NS					NS				
P X K		NS					NS					NS				
N X P X K		NS					NS					NS				

Table 36. Effect of amendment of soil with different doses of fertilizers on mean fruit weight (g) of eggplant inoculated with *Pseudomonas solanacearum* race 1

Treatments Doses of N and P ₂ O ₅ kg/ha	Doses of K ₂ O (kg/ha)									
	1988					1989				
	40	60	80	Mean		40	60	80	Mean	Mean values of two years' data
N										
P ₂ O ₅										
60	91.71	107.52	103.27	100.83	64.12	70.37	75.00	69.83	77.92	88.95 89.13 85.33
60	109.12	107.63	116.05	110.93	76.87	80.09	81.56	79.51	93.00	93.86 98.80 95.22
80	120.42	105.60	107.11	111.04	82.10	83.25	83.67	83.01	101.26	94.43 95.39 97.03
Mean	107.08	106.92	108.81	107.60	74.36	77.90	80.08	77.45	90.73	92.41 94.45 92.53
90	117.05	137.73	124.10	126.29	80.87	90.50	92.11	87.83	98.96	114.12 108.10 107.06
60	116.33	122.71	125.15	121.40	81.22	82.83	85.27	83.11	98.78	102.77 105.21 102.25
80	113.75	110.80	145.87	123.47	79.57	93.91	100.00	91.16	96.66	102.36 122.93 107.32
Mean	115.71	123.75	131.71	123.72	80.55	89.10	92.46	87.37	98.14	106.42 112.08 105.55
120	150.12	142.34	135.67	142.71	105.10	106.86	110.17	107.38	127.61	124.60 122.92 125.04
60	140.83	140.25	150.57	143.88	99.42	102.50	106.23	102.72	120.13	121.38 128.40 123.30
80	143.85	133.42	149.19	142.15	101.84	103.67	105.30	103.60	122.85	118.55 127.24 122.88
Mean	144.93	138.67	145.14	142.91	102.13	104.34	107.23	104.57	123.53	121.51 126.19 123.74
Sources of variation										
	C.D. of 1988 data at 5%				C.D. of 1989 data at 5%				C.D. of mean values of two years' data at 5%	
N	10.48				8.27				7.63	
P	NS				NS				NS	
K	NS				NS				NS	
N X P	NS				NS				NS	
N X K	NS				NS				NS	
P X K	NS				NS				NS	
N X P X K	NS				NS				NS	

Table 37. Effect of amendment of soil with different doses of fertilizers on yield per plant (kg) of eggplant inoculated with *Pseudomonas solanacearum* race 1

Treatments Doses of N and P ₂ O ₅ kg/ha		Doses of K ₂ O (kg/ha)														
		1988					1989					Mean values of two years' data				
		40	60	80	Mean		40	60	80	Mean	40	60	80	Mean		
60	P ₂ O ₅															
	40	1.80	1.92	1.90	1.87	0.97	1.00	1.04	1.00	1.39	1.46	1.47	1.44			
	60	1.84	1.95	2.34	2.04	1.00	1.09	1.21	1.10	1.42	1.53	1.78	1.58			
	80	2.20	2.01	2.20	2.14	1.18	1.22	1.21	1.20	1.69	1.62	1.71	1.67			
	Mean	1.95	1.96	2.15	2.02	1.05	1.10	1.15	1.10	1.50	1.54	1.65	1.56			
90	40	2.07	2.35	2.59	2.34	1.00	1.18	1.30	1.16	1.54	1.77	1.95	1.75			
	60	2.51	2.49	2.79	2.60	1.24	1.26	1.36	1.29	1.88	1.88	2.08	1.95			
	80	2.76	2.60	2.61	2.66	1.33	1.35	1.37	1.35	2.05	1.98	1.99	2.01			
	Mean	2.45	2.48	2.66	2.53	1.19	1.27	1.34	1.27	1.82	1.88	2.01	1.90			
120	40	2.70	2.58	2.70	2.66	1.41	1.39	1.40	1.40	2.06	1.99	2.05	2.03			
	60	2.73	2.84	2.62	2.73	1.41	1.48	1.57	1.49	2.07	2.16	2.10	2.11			
	80	2.96	2.91	2.94	2.94	1.55	1.53	1.60	1.56	2.26	2.22	2.28	2.25			
	Mean	2.80	2.78	2.76	2.78	1.46	1.47	1.52	1.48	2.13	2.11	2.14	2.13			
Sources of variation		C.D.of 1988 data at 5%					C.D.of 1989 data at 5%					C.D. of mean values of two years' data at 5%				
N		0.16					0.12					0.17				
P		0.16					0.12					0.17				
K		NS					NS					NS				
N X P		NS					NS					NS				
N X K		NS					NS					NS				
P X K		NS					NS					NS				
N X P X K		NS					NS					NS				

exhibited no significant effect on total yield per plot (Table 38).

It was also concluded that increase in nitrogen and phosphorus concentrations caused significant increase of projected yield (quintal/ha) in 1988 and 1989. The mean values of two years' data also reflected that increasing concentrations of nitrogen, phosphorus and potassium caused significant increase of projected yield (quintal/ha). Increase of potassium alone showed significant increase of projected yield (quintal/ha) during 1989 but not in 1988. All other treatment groups exhibited no significant effect on the projected yield (quintal/ha) (Table 39).

Effects of Oilcakes on wilt incidence. — Four oilcakes (Karanja, Mahua, Mustard and Neem) each at three doses (100, 200 or 400 gm/m²) were separately applied to experimental plots. Eggplant seedlings of cultivar Improved Muktakeshi were inoculated by root clipping technique with the pathogen and transplanted to the treatment and control plots. After one month all seedlings were inoculated for a second time by soil drenching method as described in Materials and Methods. Incidence of wilt and other parameters were assessed after 45 days of transplantation and also at final harvest time. The results are presented in Tables 40 to 43.

It may be noted from the data that amendment of soil with high doses of Karanja and neemcake caused significant decrease of wilt incidence recorded at final harvest time. In contrast, the application of mahua and mustard oilcakes caused no

Table 38. Effect of amendment of soil with different doses of fertilizers on total yield/plot (kg) of eggplant inoculated with *Pseudomonas solanacearum* race 1

Treatments Doses of N and P ₂ O ₅ kg/ha		Doses of K ₂ O (kg/ha)														
		1988					1989					Mean values of two years' data				
		N	P ₂ O ₅	40	60	80	Mean	40	60	80	Mean	40	60	80	Mean	
60	40	14.04	18.04	15.69	15.92	6.80	8.25	9.86	8.30	10.42	13.15	12.78	12.12			
	60	19.53	22.16	29.49	23.73	8.85	10.65	13.33	10.94	14.19	16.41	21.41	17.34			
	80	21.49	22.83	26.75	23.69	9.74	10.52	11.60	10.62	15.62	16.68	19.17	17.16			
	Mean	18.34	21.01	23.98	21.11	9.46	9.81	11.60	9.96	13.41	15.42	17.79	15.54			
90	40	25.10	26.59	34.70	28.80	8.75	10.52	14.73	11.33	16.92	18.56	24.72	20.07			
	60	29.57	29.76	36.16	31.83	13.91	12.92	16.84	14.56	21.74	21.34	26.50	23.19			
	80	33.61	35.49	37.47	35.52	14.32	14.98	17.83	15.71	23.97	25.23	27.65	25.62			
	Mean	29.43	30.61	36.11	32.05	12.33	12.81	16.47	13.87	20.88	21.71	26.29	22.96			
120	40	41.30	35.98	41.30	39.53	16.46	18.11	19.85	18.14	28.88	27.05	30.58	28.84			
	60	39.12	42.08	39.27	40.16	18.48	19.31	22.73	20.17	28.80	30.70	31.00	30.17			
	80	43.61	45.51	44.94	44.69	21.46	22.94	23.52	22.64	32.54	34.23	34.23	33.67			
	Mean	41.34	41.19	41.84	41.46	18.80	20.12	22.03	20.32	30.07	30.66	31.94	30.89			

Sources of variation		C.D. of 1988 data at 5%					C.D. of 1989 data at 5%					C.D. of mean values of two years' data at 5%				
N		3.73					2.51					2.66				
P		3.73					2.51					2.66				
K		NS					2.51					2.66				
N X P		NS					NS					NS				
N X K		NS					NS					NS				
P X K		NS					NS					NS				
N X P X K		NS					NS					NS				

Table 39. Effect of amendment of soil with different doses of fertilizers on projected yield (quintal/ha) of eggplant inoculated with *Pseudomonas solanacearum* race 1

Treatments Doses of N and P ₂ O ₅ kg/ha		Doses of K ₂ O (kg/ha)											
		1988				1989				Mean values of two years' data			
		N	P ₂ O ₅	40	60	80	Mean	40	60	80	Mean	40	60
60	40	110.47	142.00	123.43	125.30	50.93	61.74	73.76	62.14	80.70	101.87	98.60	93.72
	60	153.70	174.37	232.10	186.72	68.20	79.63	99.70	82.51	110.93	127.00	165.90	134.61
	80	169.10	179.63	210.50	186.41	72.83	78.70	86.77	79.43	120.98	129.17	148.64	132.93
	Mean	144.42	165.33	188.68	166.14	63.99	73.35	86.75	74.70	104.20	119.34	137.72	120.42
90	40	197.53	209.27	273.13	226.64	65.43	78.70	110.20	84.78	131.48	143.99	191.67	155.71
	60	232.20	234.20	284.57	250.50	104.03	96.63	125.90	108.85	168.37	165.43	205.24	179.68
	80	264.50	265.43	280.27	270.07	107.13	112.03	133.33	117.50	185.82	188.73	206.80	193.78
	Mean	231.58	236.30	279.32	249.07	92.20	95.78	123.14	103.71	161.89	166.04	201.24	175.39
120	40	308.93	269.13	308.93	295.66	123.13	135.50	148.47	135.70	216.03	202.32	228.70	215.68
	60	292.60	314.80	293.80	300.40	138.27	156.67	157.87	150.94	215.44	235.74	225.83	225.67
	80	326.23	340.43	336.10	334.25	160.50	171.60	175.93	169.34	243.37	256.01	256.02	251.80
	Mean	309.24	308.12	312.95	310.10	140.63	154.58	160.76	151.99	224.95	231.35	236.85	231.05

Sources of variation		C.D. of 1988 data at 5%				C.D. of 1989 data at 5%				C.D. of mean values of two years' data at 5%			
N		28.60				18.78				19.83			
P		28.60				18.78				19.83			
K		NS				18.78				19.83			
N X P		NS				NS				NS			
N X K		NS				NS				NS			
P X K		NS				NS				NS			
N X P X K		NS				NS				NS			

significant decrease of wilt as compared to controls during two years of experimentation. It is also evident from the data that only the highest dose (400 g./m^2) of karanja cake could effectively reduce the wilt incidence at final harvest time. The data on neem oilcake indicate that the highest dose (400 g./m^2) and the moderate dose (200 g./m^2) reduce wilt incidence as compared to controls (Table 40).

It is apparent from the data (Table 41) that application of high dose (400 g./m^2) of karanja, mustard and neem as well as the moderate dose (200 g./m^2) of neemcake could significantly increase the plant height as compared to control in both the years of experimentation and in mean values of two years' data. The same treatments could significantly increase the number of primary branches per plant in 1988 but not in 1989. Amendment of soil with the test doses of mahua had no significant effects with respect to plant height and the number of primary branches per plant as compared to unamended control in either of the years of experimentation.

It may be noted from the data (Table 42) that application of high dose (400 g./m^2) of karanja, mustard and neemcake caused significant increase in length, diameter and weight of fruit as compared to control during the years of experimentation as well as in mean values of two years' data. Mahua cake at its highest dose (400 g./m^2) elongated the fruit length significantly in both the years of study and in mean values of two years' data. However, this cake could not increase significantly the diameter and weight of fruit as compared to control during the period of

Table 40. Effect of amendment of soil with different oilcakes on wilt incidence of eggplant inoculated with race 1 of *Pseudomonas solanacearum* and determined at pre-top dressing and final harvest time

Treatments ^c	Percentage of wilted plants in the field					
	Pre-top dressing ^a			Final harvest time ^b		
	1988	1989	Mean values of 2 years' data	1988	1989	Mean values of 2 years' data
(1)	(2)	(3)	(4)	(5)	(6)	(7)
Karanja						
100	23.33 (28.86) ^a	28.33 (32.14)	25.83 (30.53)	45.00 (42.11)	48.33 (44.03)	46.67 (43.07)
200	20.00 (26.26)	26.67 (30.79)	23.33 (28.79)	38.33 (38.22)	41.67 (40.17)	40.00 (39.22)
400	16.67 (24.05)	21.67 (27.71)	19.17* ^f (25.93)	30.00* (33.16)	35.00* (36.15)	35.00* (36.26)
Mahua						
100	23.33 (28.78)	30.00 (33.16)	26.67 (31.03)	45.00 (42.09)	50.00 (45.01)	47.50 (43.55)
200	21.67 (21.71)	28.33 (32.09)	25.00 (29.95)	43.33 (41.15)	48.33 (44.04)	45.83 (42.60)
400	20.00 (26.45)	26.67 (31.07)	23.33 (28.84)	38.33 (38.19)	41.67 (40.17)	40.00 (39.23)
Mustard						
100	25.00 (29.80)	31.67 (34.15)	28.33 (32.03)	51.67 (45.94)	51.67 (45.97)	51.67 (45.96)
200	26.67 (31.07)	30.00 (33.16)	28.33 (32.10)	53.33 (46.95)	53.33 (46.95)	53.33 (46.96)
400	26.67 (31.00)	30.00 (33.16)	28.33 (32.13)	55.00 (48.09)	51.67 (45.96)	53.33 (47.00)
Neem						
100	23.33 (28.86)	28.33 (32.14)	25.83 (30.53)	45.00 (42.09)	46.67 (43.08)	45.83 (42.60)
200	18.33 (25.00)	25.00 (29.93)	21.67 (27.58)	31.67* (34.15)	35.00* (36.24)	33.33* (35.22)
400	15.00 (22.79)	20.00 (26.57)	17.50* (24.73)	25.00* (29.74)	28.33* (32.02)	26.67* (30.90)

Table 40. Effect of amendment of soil with different oilcakes on wilt incidence of eggplant inoculated with race 1 of Pseudomonas solanacearum and determined at pre-top dressing and final harvest time

Treatments ^c	Percentage of wilted plants in the field					
	Pre-top-dressing ^a			Final harvest time ^b		
	1988	1989	Mean values of 2 years' data	1988	1989	Mean values of 2 years' data
(1)	(2)	(3)	(4)	(5)	(6)	(7)
Control ^d	25.00 (30.00)	31.67 (34.23)	28.33 (32.10)	50.00 (45.00)	53.33 (46.95)	51.67 (45.96)
C.D. at 5%	NS	NS	5.18	9.96	8.91	6.89

^a Topdressing of experimental plots with urea @ 45.0 kg/ha was done 45 days after transplanting

^b Data were taken just before the final harvest of the fruits

^c Doses of oilcakes were 100, 200 or 400 g per m² for each in combination with N : P : K fertilizers @ 90:60:60 kg/ha

^d No oilcake but only N:P:K fertilizers were used

^e Angular transformation values are given in parentheses

^f Single asterisk mark indicates significance at P=0.05 level with respect to control.

Table 41. Effect of amendment of soil with different oilcakes on plant height and number of primary branches/plant of eggplant inoculated with race 1 of Pseudomonas solanacearum

Treatments ^a	Plant height (cm)			Number of primary branches per plant		
	1988	1989	Mean values of 2 years' data	1988	1989	Mean values of 2 years' data
Karanja						
100	73.30	59.43	63.37	5.13	4.43	4.78
200	81.17	62.35	71.76	5.47	4.50	4.99
400	94.00* ^c	68.10*	81.05*	5.97*	5.17	5.57*
Mahua						
100	71.93	57.20	64.57	4.95	4.40	4.68
200	73.13	60.65	66.89	5.27	4.60	4.94
400	80.33	63.90	72.12	5.37	4.70	5.04
Mustard						
100	74.50	59.83	67.17	5.10	4.47	4.79
200	83.03	63.10	73.07	5.30	4.80	5.05
400	97.53*	69.25*	83.39*	6.10*	5.33	5.72*
Neem						
100	76.17	62.15	69.16	5.35	4.60	4.98
200	90.40*	66.75*	78.58*	5.90*	5.10	5.50*
400	100.93*	70.77*	85.85*	6.20*	5.43*	5.82*
Control ^b	70.83	56.45	63.64	4.60	4.23	4.42

C.D.	16.58	7.93	9.83	1.16	1.11	0.71

^a Doses of oilcakes were 100, 200 or 400 g./m² for each in combination with N:P:K fertilizers @ 90:60:60 kg/ha.

^b No oilcake but only N:P:K fertilizers were used.

^c Single asterisk mark indicates significance at P=0.05 with respect to control.

Table 42. Effect of amendment of soil with different oilcakes on some fruit characters of eggplant inoculated with race 1 of *Pseudomonas solanacearum*

Treatments ^a	Mean length of fruit(cm)			Mean diam. of fruit (cm)			Mean weight of fruit (g)		
	1988	1989	Mean values of 2 years' data	1988	1989	Mean values of 2 years' data	1988	1989	Mean values of 2 years' data
Karanja									
100	9.41	7.37	8.39	4.93	3.40	4.17	95.33	81.33	88.33
200	10.00	8.03	9.02	5.57	3.93	4.75	109.78	98.02	103.90
400	11.00 ^{*c}	8.91 [*]	9.96 [*]	6.50 [*]	4.80	5.65 [*]	138.21 [*]	122.68 [*]	130.45 [*]
Mahua									
100	9.23	7.25	8.24	4.90	3.37	4.14	93.30	80.60	86.95
200	9.80	7.73	8.77	5.37	3.80	4.59	101.00	93.30	97.15
400	10.90 [*]	8.80 [*]	9.85 [*]	6.03	4.45	5.24	113.43	109.52	111.48
Mustard									
100	9.33	7.30	8.32	4.95	3.43	4.19	98.67	81.44	90.06
200	9.80	7.87	8.84	5.60	4.00	4.80	116.84	94.55	105.70
400	10.93 [*]	8.85 [*]	9.89 [*]	6.55 [*]	4.83	5.69 [*]	140.10 [*]	124.06 [*]	132.08 [*]
Neem									
100	9.43	7.33	8.38	5.00	3.45	4.23	99.84	81.85	90.85
200	9.97	8.00	8.99	5.70	4.10	4.90	122.13	106.52	114.33 [*]
400	11.25 [*]	9.00 [*]	10.13 [*]	6.63 [*]	4.90	5.77 [*]	150.00 [*]	125.38 [*]	137.69 [*]
Control ^b	9.17	7.15	8.16	4.87	3.33	4.10	92.86	80.11	86.49
C.D. at 5%	1.43	1.36	1.07	1.25	NS	1.22	35.81	33.41	27.34

^a Doses of oilcakes were 100, 200 or 400 g/m² for each in combination with N:P:K fertilizers @ 90:60:60 kg/ha.

^b No oilcake but only N:P:K fertilizers were used.

^c Single asterisk mark indicates significance at P 0.05 with respect to control.

Table 43. Effect of amendment of soil with different oilcakes on some yield characters of eggplant inoculated with race 1 of *Pseudomonas solanacearum*

Treatments ^a	Yield per plant (kg)			Total yield/plot (kg)			Projected yield (quintal/ha)		
	1988	1989	Mean values of 2 years' data	1988	1989	Mean values of 2 years' data	1988	1989	Mean values of 2 years' data
Karanja	0.98	0.81	0.90	10.40	8.40	9.40	81.87	66.10	73.99
	1.17	1.00	1.09	14.90	12.00	13.43	117.27	94.43	105.85
	1.54 ^c	1.37*	1.46*	21.76*	18.97*	20.37*	171.30*	149.30*	160.30*
	0.95	0.80	0.88	10.17	8.19	9.19	80.03	64.50	72.27
Mustard	1.07	0.87	0.97	12.26	9.46	10.87	96.50	74.47	85.49
	1.15	1.00	1.08	14.09	11.43	12.77	110.93	89.97	100.45
	0.09	0.83	0.91	9.50	8.30	8.91	74.80	65.33	70.07
	1.19	1.03	1.11	11.70	10.53	11.12	92.10	82.90	87.50
N neem	1.50*	1.33*	1.42*	12.90	13.40	13.15	101.53	105.47	103.50
	1.02	0.87	0.95	11.33	9.17	10.26	89.20	72.13	80.67
	1.48*	1.31*	1.40*	20.37*	15.67*	18.02*	160.30*	123.30*	141.80*
	1.67*	1.43*	1.55*	24.93*	20.63*	22.79*	196.20*	162.37*	179.29*
Control ^b	0.90	0.79	0.85	8.80	7.50	8.15	69.27	59.03	64.15
C.D.	0.46	0.44	0.41	7.12	6.53	5.35	55.81	51.42	42.23

^aDoses of oilcakes were 100, 200 or 400 g/m² for each in combination with N:P:K fertilizers @ 90:60:60 kg/ha

^bNo oilcake but only N:P:K fertilizers were used.

Single asterisk mark indicates significance at P, 0.05 with respect to control.

investigation and when data were averaged.

Evaluation of yield parameters, such as, yield per plant, total yield per plot and projected yield following amendment of soil with different oilcakes indicate that high dose (400 g/m^2) of karanja and moderate (200 g/m^2) and high (400 g/m^2) doses of neemcake caused significant increase of these yield parameters as compared to control in both the years of experimentation. The mean values of two years' data also reflected this increase in yield. Application of high dose of mustard oilcake, although showed increase in yield per plant but did not increase the total yield per plot. Amendment of soil with mustardcake did not reduce wilt incidence (Table 40). Consequently there was heavy wilting and high percentage of mortality of plants. The few surviving plants, however, showed more yield per plant as compared to control. Application of low, moderate, or high doses of mahua oilcake did not reduce wilt incidence (Table 40) and consequently did not have any increasing effect on the yield parameters evaluated in the study.

Effects of irrigation schedules on wilt incidence. — In this series of experiments, wilt incidence as affected by different irrigation schedules was examined. Four irrigation schedules, such as, watering of plots at intervals of 5, 10, 15 or 20 days were used. Eggplant seedlings of cultivar Improved Muktakeshi were transplanted to the fields following inoculation and exposed to above mentioned irrigation regimes. The seedlings were inoculated for a second time after 30 days of

transplantation by soil drenching method as stated in Materials and Methods. Incidence of wilt and other parameters of growth and yield were ascertained after 45 days of transplanting and at final harvest time. The data obtained were subjected to Duncan's multiple range test and presented in Tables 44 to 47.

It is evident from the data (Table 44) that different regimes of irrigation had little or no effects on the wilt incidence prior to topdressing except the irrigation schedule of watering at an interval of 20 days. Wilt incidence was less in this dry soils. Analysis of final harvest time data, however, indicates, that both watering at an interval of 5 or 20 days resulted in less incidence of the wilt as compared to the plots receiving irrigation at an interval of 10 or 15 days. The maximum wilt incidence was observed in plots receiving irrigation at an intervals of 10 days. The plots receiving water at an interval of 15 days showed intermediate level of wilt incidence.

Analyses of the results concerning plant height and number of primary branches per plant is presented in Table 45. It is evident from the data that more frequent irrigation schedule was significantly favourable for these two growth parameters of eggplants. In soils receiving frequent watering such as at an interval of 5 or 10 days showed more height and more number of primary branches per plant as compared to other irrigation schedules.

It is evident from the results (Table 46) that length, diameter and weight of the fruit were significantly higher in plants

Table 44. Effect of different irrigation schedules on bacterial wilt incidence of eggplant under artificial inoculation conditions in the field and determined at pre-topdressing and final harvest time

Irrigation at intervals of	Percentage of wilted plants in the field					
	Pre top-dressing**			At final harvest time***		
	1987-88	1988-89	Mean values of 2 years' data	1987-88	1988-89	Mean values of 2 years' data
5 days	31.67 *(33.86) ^a	20.00 (26.42) ^a	25.83 (30.30) ^a	35.00 (35.63) ^a	27.50 (31.07) ^a	31.25 (33.78) ^a
10 days	35.00 (35.87) ^a	25.83 (30.34) ^a	30.42 (33.36) ^a	63.33 (53.38) ^b	52.50 (46.54) ^b	37.92 (49.77) ^b
15 days	29.17 (32.27) ^a	20.83 (26.77) ^a	25.00 (29.88) ^a	49.17 (44.49) ^{ab}	38.33 (38.12) ^{ab}	43.75 (41.32) ^c
20 days	13.33 (21.24) ^b	11.67 (19.60) ^b	12.50 (20.62) ^b	30.00 (32.90) ^a	23.33 (28.70) ^a	26.67 (30.96) ^a
C.D. at 5%	9.50	5.94	5.09	11.83	9.52	7.99

* Angular transformation values are presented in parentheses

** Topdressing of experimental plots with urea at the rate of 45 kg/ha was done 45 days after transplanting

*** Data were taken just before the final harvest of the fruits

N.B. Data with similar letters were not significantly different.

Table 45. Effect of different irrigation schedules on plant height (cm) and number of primary branches per plant of egg-plant under artificial inoculation conditions in the field

Irrigation at inter- vals	Plant height (cm)			No. of primary branches/per plant		
	1987-88	1988-89	Mean values of 2 years data	1987-88	1988-89	Mean values of 2 years data
5 days	92.39 ^a	74.81 ^a	93.60 ^a	5.12 ^a	4.48 ^a	4.80 ^a
10 days	89.71 ^{ab}	70.72 ^{ab}	80.22 ^{ab}	4.78 ^{ab}	3.92 ^{ab}	4.35 ^{ab}
15 days	83.82 ^{bc}	65.78 ^{bc}	74.80 ^{bc}	3.98 ^{bc}	3.47 ^{bc}	3.73 ^{bc}
20 days	80.82 ^c	61.62 ^c	71.22 ^c	3.62 ^c	2.78 ^c	3.20 ^c
C.D. at 5%	7.99	8.53	5.99	0.91	0.92	0.64

N.B. Data with similar letters were not significantly different.

Table 46. Effect of different irrigation schedules on some fruit characters of eggplant under artificial inoculation conditions in the field

Irrigation at inter- vals of	Mean length of fruit(cm)		Mean diameter (cm)		Mean fruit weight (g)				
	1987-88	1988-89	Mean values of 2 years data	1987-88	1988-89	Mean values of 2 years data			
5 days	12.52 ^a	9.92 ^a	11.22 ^a	6.33 ^a	6.97 ^a	6.65 ^a	161.18 ^a	135.6 ^a	145.4 ^a
10 days	11.92 ^a	9.63 ^a	10.78 ^a	5.83 ^a	6.50 ^a	6.17 ^a	165.33 ^a	135.18 ^a	157.7 ^a
15 days	9.58 ^b	7.18 ^b	8.38 ^b	4.28 ^b	4.82 ^b	4.55 ^b	89.08 ^b	88.69 ^b	88.89 ^b
20 days	9.18 ^b	7.08 ^b	8.13 ^b	4.01 ^b	4.60 ^b	4.31 ^b	81.92 ^b	74.50 ^b	78.21 ^b
C.D.at 5%	2.14	2.37	1.44	1.54	1.62	1.18	64.65	45.14	43.42

N.B. Data with similar letters were not significantly different.

Table 47. Effect of different irrigation schedules on some yield characters of eggplant under artificial inoculation conditions in the field

Irrigation at inter- vals of	Yield/Plant (kg)		Yield/plot (kg)		Projected yield (q/ha)				
	1987-88	1988-89	Mean values of 2 years data	1987-88	1988-89	Mean values of 2 years data			
5 days	2.08 ^a	1.59 ^a	1.84 ^a	25.85 ^a	22.65 ^a	24.25 ^a	203.47 ^a	178.20 ^a	190.84 ^a
10 days	1.70 ^a	1.29 ^a	1.50 ^a	12.54 ^b	12.09 ^b	12.31 ^b	98.67 ^b	95.13 ^b	96.90 ^b
15 days	0.75 ^b	0.70 ^b	0.73 ^b	7.58 ^b	9.03 ^b	8.30 ^b	59.62 ^b	71.07 ^b	63.35 ^b
20 days	0.50 ^b	0.57 ^b	0.54 ^b	7.60 ^b	8.44 ^b	8.01 ^b	59.67 ^b	66.38 ^b	63.03 ^b
C.D. at 5%	0.75	0.54	0.42	8.85	7.66	6.59	69.66	60.27	51.84

N.B. Data with similar letters were not significantly different.

growing in drier soils i.e. receiving water less frequently yielded smaller fruits in general.

When the yield parameters were considered it was apparent that plants grown in plots receiving water at 10 days interval had significantly poor yield per plot due to more number of wilted plants and this was reflected also in projected yield data. It was of interest that yield per plant of plants which received water at an interval of 5 or 10 days was similar, although, the latter watering regime caused the highest incidence of wilt (Table 44). The highest yield per plot was obtained in plots which received watering at an interval of 5 days only due to low incidence of wilt in such plots as well as better performance of growth and yield parameters. The plants which received water less frequently (15 or 20 days interval) showed poor yield. Apparently, drought was the limiting factor in latter cases.

Chemical control. — Since the pathogen has been found to be persisting on seeds of eggplant, the possibility of controlling the wilt of eggplant by treatment of seeds, with different toxic chemicals was investigated in this series of experiments.

Seed treatment in vitro. — The efficacy of different concentrations of captan, vitavax and streptomycin for eradication of the pathogen from the seed surface was attempted and the results are presented in Table 48.

It may be noted from the data that out of the three chemicals, only streptomycin could effectively eradicate the pathogen from the surface of the seeds. Seeds exposed to 250 ppm of streptomycin for 30 minutes could effectively reduce the percentage of infections

Table 48. Effects of seed treatments on eradication of the pathogen from the seeds of eggplant as determined by seed pathological study

Treatments	Treatment conc.	Treatment time (minutes)	Total per cent seed infected	Per cent seed infected with		
				Bacteria	Fungi	Actinomyces
Control ^a (Untreated)	-	-	100	95	5	-
Sterile distilled water ^b	-	15	100	92	8	-
	-	30	100	89	11	-
Streptomycin ^c	125 mg/L	15	45	45	-	-
		30	17	17	-	-
	250 mg/L	15	33	33	-	-
		30	4	4	-	-
	500 mg/L	15	15	15	-	-
		30	-	-	-	-
Captan	0.3%	15	100	100	-	-
		30	100	100	-	-
	0.4%	15	100	100	-	-
		30	100	100	-	-
	0.5%	15	97	97	-	-
		30	95	95	-	-
Vitavax	0.3%	15	100	100	-	-
		30	100	100	-	-
	0.4%	15	100	100	-	-
		30	100	100	-	-
	0.5%	15	100	100	-	-
		30	100	100	-	-

^a Unwashed dry seeds were directly plated on PDA

^b Seeds (5 g) were washed with 10 ml of sterile distilled water for indicated period and then plated on PDA

^c Seeds (5g) were treated with different concentrations of test chemicals separately and then plated on PDA.

upto 4% only as compared to 100% infection in control seeds. captan and vitavax treatments had no reducing effect on seed contamination at any one of the concentration tested.

Effect of seed treatment on wilt incidence in field.— In this series of experiments, seedlots of Improved Muktakeshi were treated with the test chemicals separately, and the seedlings were raised in seedbed as described in Materials and Methods. These seedlings were transplanted to replicated plots in Mondouri farm and the wilt development was monitored under natural conditions of infection. Seedlings raised from untreated seeds were used as controls. Wilt incidence was determined at pre-topdressing and final harvest time. The other growth and yield parameters were also evaluated. The results are presented in Tables 49 to 52.

It may be noted from the data that plants from streptomycin-treatment group exhibited significantly less wilt incidence as compared to controls. Similar results were obtained at pre-topdressing and final harvest time. Plants grown from seeds treated with captan or vitavax showed percentage of wilt incidence comparable to controls.

The results concerning the plant growth and yield parameters of eggplants raised from seeds treated with test chemicals are presented in Tables 50 to 52.

It may be noted that plant height, number of primary branches per plant and yield parameters including fruit length, diameter,

Table 49. Effect of pre-sowing seed treatment on bacterial wilt incidence of eggplant under natural infection conditions of field and determined at pre-topdressing and final harvest time

Treatment	Percentage of wilted plants in the field at					
	Pre-topdressing			Final harvest time		
	1987	1988	Mean values of 2 years' data	1987	1988	Mean values of 2 years' data
Control	15.00 ^a (22.44)	23.33 ^a (28.73)	19.17 ^a (25.85)	35.00 ^a (36.01)	38.33 ^a (38.18)	36.67 ^a (37.18)
Captan	12.50 ^a (20.32)	21.67 ^a (27.56)	17.08 ^a (24.25)	31.67 ^a (33.86)	35.83 ^a (36.57)	33.75 ^a (35.39)
Strepto- mycin	5.83 ^b (13.84)	11.67 ^b (19.40)	8.75 ^b (16.97)	14.17 ^b (21.52)	18.33 ^b (24.79)	16.25 ^b (23.49)
Vitavax	11.67 ^a (17.60)	19.17 ^a (25.73)	15.42 ^a (22.99)	29.17 ^a (32.35)	33.33 ^a (34.97)	31.25 ^a (33.77)
C.D. at 5%	5.06	6.17	3.82	8.90	7.76,	6.80

^a Topdressing of experimental plots with urea @ 45.0 kg/ha was done 45 days after transplanting

^b Data were taken just before the final harvest of the fruits

^c Transformed values are given in parentheses

N.B. Data with similar letters were not significantly different.

Table 50. Effect of pre-sowing seed treatment on plant height and number of primary branches per plant of eggplant exposed to natural infection by race 1 of P. solanacearum under field conditions

Treatment	Plant height (cm)			No. of primary branches/plant		
	1987	1988	Mean values of 2 years' data	1987	1988	Mean values of two years' data
Control	71.60 ^a	64.80 ^a	68.20 ^a	3.60 ^a	2.60 ^a	3.10 ^a
Captan	72.33 ^a	65.35 ^a	68.84 ^a	3.80 ^a	2.90 ^a	3.35 ^a
Strepto-mycin	80.50 ^b	74.95 ^b	77.73 ^b	5.20 ^b	4.60 ^b	4.90 ^b
Vitavax	73.04 ^a	67.90 ^a	70.47 ^a	4.00 ^a	3.10 ^a	3.55 ^a
C.D. at 5%	6.37	5.96	3.65	1.12	1.18	0.90

N.B. Data with similar letters were not significantly different.

Table 51. Effect of pre-sowing seed treatment on some fruit characters of eggplant exposed natural infection by race 1 of P. solanacearum under field conditions

Treatment	Length of fruit (cm)		Diameter of fruit (cm)		Mean fruit weight (g)				
	1987	1988	Mean values of 2 years' data	1987	1988	Mean values of 2 years' data			
Control	10.20 ^a	9.35 ^a	9.78 ^a	6.00 ^a	5.10 ^a	5.55 ^a	77.33 ^a	51.23 ^a	64.28 ^a
Captan	12.80 ^a	12.40 ^a	12.60 ^a	6.20 ^a	5.30 ^a	5.75 ^a	81.40 ^a	50.55 ^a	65.98 ^a
Strepto- mycin	14.00 ^b	12.79 ^b	13.49 ^b	7.20 ^b	7.00 ^b	7.10 ^b	122.37 ^b	81.04 ^b	161.71 ^b
Vitavax	13.60 ^b	12.52 ^b	13.06 ^b	6.30 ^b	5.70 ^b	6.00 ^b	90.45 ^b	61.28 ^b	75.87 ^b
C.D.at 5%	2.42	2.61	2.04	0.83	1.17	0.55	26.28	19.20	15.44

N.B. Data with similar letters were not significantly different.

Table 52. Effect of pre-sowing seed treatment on some yield characters of eggplant exposed to natural infection by race 1 of P. solanacearum under field conditions

Treatment	Yield/plant (kg)			Total yield/plot (kg)			Projected yield (quintal/ha)		
	1987	1988	Mean values of 2 years, data	1987	1988	Mean values of 2 years, data	1987	1988	Mean values of two years, data
Control	0.93 ^a	0.75 ^a	0.84 ^a	11.89 ^a	9.33 ^a	10.61 ^a	93.52 ^a	73.45 ^a	83.49 ^a
Captan	1.08 ^a	0.83 ^a	0.96 ^a	14.67 ^a	10.42 ^a	12.54 ^a	115.42 ^a	81.97 ^a	98.70 ^a
Strepto-mycin	1.92 ^b	1.70 ^b	1.81 ^b	33.35 ^b	27.48 ^b	30.41 ^b	262.47 ^b	216.30 ^b	239.39 ^b
Vitavax	1.27 ^a	0.95 ^a	1.11 ^a	18.15 ^a	11.67 ^a	14.90 ^a	142.85 ^a	91.52 ^a	117.34 ^a
C.D.at 5%	0.51	0.52	0.34	9.29	7.35	6.28	73.11	57.82	49.42

N.B. Data with similar letters were not significantly different.

weight and yield per plot and hectare, all these parameters were significantly more in plants raised from seeds treated with streptomycin. Treatment of seeds with Captan and Vitavax failed to control wilt and consequently growth and yield parameters were also comparable to control plants.

DISCUSSION

The identity of the causal organism of a particular disease is a necessary prerequisite for planning and initiation of effective control measures. It is now well established that there are several pathogenic variants of P. solanacearum in nature. Buddenhagen et al. (1962) reported presence of at least 3 races, whereas Hayward (1964, 1976) considered these as biovars and proposed the existence of a fourth biovar (= race) of the pathogen. The three races (Sensu Buddenhagen et al., 1962) of the pathogen, however, can be distinguished by their cultural, biochemical and pathogenic properties categorized by Buddenhagen et al. (1962) and Hayward (1976). Lack of definitive knowledge of the distribution pattern and the exact identity of the race resulted in much confusion regarding survival pattern of the wilt pathogen and control of the disease in the past (Buddenhagen and Kelman, 1964).

The results of the present investigation clearly indicate that the isolate of the pathogen causing eggplant wilt in West Bengal is race 1 of P. solanacearum. The isolate could utilize hexose alcohols, mannitol and sorbitol, and produced acid from disaccharides like maltose and lactose. These are characteristic biochemical features of race 1. In contrast, the races 3 ($\equiv$ potato isolate) and 2 ($\equiv$ banana isolate) could not utilize sugar alcohols (Hayward, 1976). Previously the eggplant isolate of the pathogen

from West Bengal was considered as P. solanacearum var. asiaticum (Das and Chattopadhyay, 1955). Most systematists working on P. solanacearum, however, considered that the pathovar asiaticum is either non existent (Hayward, 1976) or it is a variant of the race 3 (Buddenhagen and Kelman, 1964). Widespread occurrence of race 1 in the plains of India including West Bengal and the presence of race 3 only in the hills has been reported by several Indian workers (Rao, 1976; Shekhawat, 1976).

Knowledge of the ecology of P. solanacearum in seed or in soil is lacking because adequate techniques are not available to monitor the pathogen in soil (Sequeira and Kelman, 1976). Survival of P. solanacearum in soil has been determined indirectly with the use of indicator plants to measure inoculum densities in soil (Kelman, 1953; Buddenhagen and Kelman, 1964; Sequeira and Kelman, 1976). Serological techniques were used to detect cells in soils (Jenkins et al., 1967), but live cells were not distinguished from dead cells, or virulent from avirulent ones. Several media have been developed for isolation and quantification of P. solanacearum from soil (Okabe, 1969; Karganilla and Buddenhagen, 1972; Tanaka and Noda, 1973; Ramos, 1976, 1978) but none has gained wide acceptance. A major limitation of these media is that low population densities of the pathogen are not detectable. None of these media was satisfactory in preliminary tests for selective isolation of P. solanacearum from North Carolina soil (Nesmith and Jenkins, 1976).

In this study of survival ecology of P. solanacearum in seed and in soil, the population dynamics and persistence of race 1

cells as affected by diverse abiotic factors of the soil were investigated by using streptomycin and erythromycin resistant marker strain. The double marker strain was used because it is much quicker and easier to carryout than the other methods (Obaton, 1971). Moreover, the change in population dynamics of the pathogen can be measured directly by this method. Development of antibiotic resistant strain of race 1 following the two steps method of Obaton (1971) is quite easy, but critical appraisal of the stability of the marker characters, biochemical properties and pathogenic ability and virulence of the marker strain is necessary before it can be used for survival studies (Obaton, 1971; Schwinghamer and Dudman, 1973).

In preliminary experiments the wild type cells of race 1 of P. solanacearum were found to be highly sensitive to streptomycin and erythromycin and failed to grow in presence of 200 $\mu\text{g/ml}$ of streptomycin or erythromycin in the medium. All the soil microflora, bacteria, actinomycetes and fungi of the sandy loam soil of Kalyani used in this study were completely inhibited by 200 $\mu\text{g/ml}$ of streptomycin or 400 $\mu\text{g/ml}$ of erythromycin (results not shown in Table 5). Soil dilutions plated on 600 $\mu\text{g/ml}$ of streptomycin or erythromycin did not develop any colony. The marker strain of race 1 capable of withstanding 2 mg/ml streptomycin and 1 mg/ml erythromycin was, therefore, considered to be highly suitable for the study of population dynamics in soil.

Comparison of morphological and biochemical properties of the double marker strain of race 1 with that of the wild type

strain further indicated that the double marker strain used in this study retained all characters of the wild type cells except tolerance to streptomycin and erythromycin. The stability of the double marker strain was further tested by examining the pathogenicity and recovery of cells from soil.

The virulence and infectivity of the double marker strain developed in this study were comparable to the wild type strain. Both the wild type and double marker strains caused high percentage of wilt of seedlings of tomato cultivars Marglobe Supreme and Best of All within 4 days of inoculation. Apparently, this double marker strain retained its pathogenicity. The results concerned with the recovery study (Table 13) clearly indicated that the cells of the marker strain could be successfully recovered from the soil. The morphological, biochemical and pathogenic properties of the recovered cells were comparable to that of wild type cells.

Results concerned with the seed pathological study clearly indicated that the eggplant strain of P. solanacearum could be isolated from the seeds collected from the local markets. A large number of seed samples of eggplant obtained from the germ plasm collection of BCKV also yielded P. solanacearum as one of components of seed surface microflora when plated on TZC medium (Kelman, 1954; Husain and Kelman, 1958). Surface sterilization of the seeds could effectively eradicate the bacterium from the seeds. The results suggest that the pathogen is possibly externally seedborne. Externally seedborne nature and persistence

of the pathogen on the seeds of tomato have been reported previously (Remadevi and Menon, 1980; Moffett et al., 1981). Several authors, however, failed to detect persistence and transmission of the pathogen in seeds of tomato and chilli previously (Labrousse, 1932; Shekhawat et al., 1979). The apparent discrepancy in the results could be due to variability of the isolates or races used by these authors. The eggplant isolate used in this study were all identified as race 1 of P. solanacearum on the basis of their morphological, biochemical and pathogenic properties and the results indicated their survival on the seed surface.

It is evident from the study that the double marker strain of race 1 of P. solanacearum could successfully survive on the seeds of eggplant. When seeds, following inoculation with the marker strain were incubated under laboratory conditions, significant number of cells of the marker strain could be isolated after 4 weeks of incubation. Furthermore, the population density of the double marker strain on the seeds of eggplant increased during the 1st week, but declined thereafter. It was of interest that high population density of the pathogen persisted on seeds incubated at low temperature and at low percentages of relative humidity of the ambient atmosphere. The use of double marker strain provided direct proof of the persistence of race 1 on the surface of eggplant seeds for a prolonged period.

The results concerned with the recovery study (Table 13) clearly indicated that the cells of the marker strain of race 1 could be successfully recovered from the soil. It was of interest

to note that the recovery of the introduced race 1 cells was dependent on the initial population density added to soil. Very low or high initial population density resulted in less recovery, whereas 50 to 75 per cent of the introduced cells could be recovered at zero time when the initial population density was 8×10^7 cells per g of soil. It is evident from the results concerned with survival of the marker cells in sterilized and non-sterilized soil samples that race 1 multiplied more in sterilised soil than in unsterilized soils. Moreover, the increased level of population in sterilized soils persisted better than the natural soil. The rate of decline in sterilized soil was slower than in natural soil. In contrast, in natural soils the population of race 1, although multiplied initially, declined rapidly and become 1/100 of the population. The results clearly indicated influence of soil biota on the population density and persistence of introduced cells of race 1 in soil environment. Large number of microorganisms antagonistic to P. solanacearum race 1 have been detected in soils suppressive to the pathogen (Nesmith and Jenkins, 1985). The suppressive soils had several bacteria and actinomycetes which strongly inhibited race 1 both in vitro and in soil. The roles of different biotic factors especially antagonistic microflora of Kalyani soil on the survival of the test marker strain of race 1 have not been included in this study.

The data concerned with the effects of incubation temperature on the survival of the marker strain in soil clearly indicated that the population of the organism increased during the first week of incubation irrespective of incubation temperature and sterile or

nonsterile nature of the soils. It is also evident that the population increased more and persisted for longer period in sterilized soils except at 40°C. At high temperature (40°C), the population declined very rapidly after the second week of incubation and after four weeks the organism was completely eliminated from the soil. At lower temperatures ranging from 15 to 30 C the marker strain multiplied and survived well in sterilized soil. The maximum growth was obtained in soils kept at 25 C. Apparently, the optimum soil temperature for the pathogen was in the range of 15 to 25 C, and at higher temperatures, the survival was poor. In non-sterilized natural soil the organisms survived poorly under identical conditions of temperature conditions. The results are in conformity with the earlier works done by several workers (Gallegly and Walker, 1949; Tanaka, 1976).

In the experiments concerned with the effects of different soil moisture levels on the survival of race 1, it was observed that survival of race 1 was better in soils with high moisture. After three weeks of incubation the population density was more in soils kept at WHC ranging from 40 to 50 per cent. Apparently drier soils were not conducive for the growth and survival of the race. Furthermore, it was observed that the test organism exhibited better survival in sterilized soils irrespective of the moisture levels. Relationship between the soil moisture level and survival of P. solanacearum races has been reported previously by several authors (Buddenhagen and Kelman, 1964; Tanaka and Noda, 1973; Okabe, 1971; Moffett et al., 1983; Nesmith and Jenkins, 1985). There are several reports that race 1 survive poorly in dry soils

(Buddenhagen and Kelman, 1964; Tanaka and Noda, 1973). In contrast, poor survival of race 1 in relatively more moist soils has also been reported (Okaba, 1971; Moffett et al., 1983). Nesmith and Jenkins (1985), however, observed that neither dry nor wet soils were conducive for survival of race 1. The organisms survived best in soils with moisture level ranging from -0.5 to -1 bars. The present results with double marker strain suggested good survival of the pathogen with high moisture levels.

It is evident from the results of the study that flooding significantly decreased the population of race 1 in non-sterilized soil. It was of interest that the population decrease in flooded soil was correlated with the depth of the free water above the soil level. Thus, with free water depth of 1.5 cm above the soil, population declined much more rapidly than soil samples with standing free water of 0.8 cm or 1 cm. It was also evident that flooding of the soils caused distribution of more cells in water phase than in soil phase. In unflooded soils most of the cells of race 1 remained in soil phase, but in flooded soils increasing in degree of flooding resulted in more and more cells distributed in water phase with time. After four weeks in incubation, natural soils flooded under 1.5 cm depth of standing water had no cells in soil phase and only a few cells in water phase. In contrast most of the surviving cells in sterilized soil were in water phase. The interaction of other soil organisms with race 1 of P. solanacearum following flooding of natural soil needs investigation. Eradication of the pathogen by flooding of the soils has been reported previously (Kelman, 1953; Van Schreven, 1948; Nesmith and Jenkins, 1985).

The results of the present study are in confirmation of these early works.

It is apparent from the results of fertilizer amendment of soils that ammonium sulphate caused significant stimulation of growth and survival of race 1 in soil. In contrast, urea and potassium nitrate were significantly inhibitory to the growth and survival of the test marker strain in soil. In soils amended with urea, the test organism did not grow at all and after 7 days of incubation could not be detected by soil plating technique. The results suggested successful eradication of the pathogen by the application of high dose (1.5%) of urea to soil. In soils amended with nitrate, population of the pathogen did not increase and the initial population introduced declined rapidly to zero level after two to three weeks. Moreover, increase in concentration of nitrate caused progressively rapid decline of the population of race 1 in soil. Reduction of wilt of tomato supplied with nitrate fertilizers (Kelman, 1950) and that of tobacco by urea (Smith and Clayton, 1943; Kincaid, 1950) have been reported. The results of the present investigation are in confirmation of these earlier works.

Laboratory experiments (results not shown here) revealed that the growth of the pathogen in nutrient cultures was inhibited by high concentration of nitrate and urea. The decrease of disease by these nitrogen fertilizers in field could be due to the inability of race 1 to utilize nitrate or urea (Kelman, 1953). The preferential utilization of ammonium nitrogen by the organism has been reported (Kelman, 1953). The nitrate reductase potential and

the operation of urea cycle in race 1 of P. solanacearum warrant investigation. An alternative explanation of inhibition of population of race 1 in soil by urea or nitrate could be stimulation of antagonists microbes in soil and has been proposed earlier (Kelman, 1953).

Amendment of soil with organic manures like oilcakes, cow-dung and crop residues caused significant stimulation of growth and survival of the marker strain in soil. The application of eggplant leaf residues was highly conducive for growth and stimulation of the marker strain in soil. The results also indicated good survival of the pathogen in presence of crop debris. The results also exhibited non-specific stimulation of the growth of the pathogen by organic residues and consequent long-term survival in soil. The survival of the pathogen in crop debris under field conditions has also been reported previously (Kelman, 1953; Graham et al., 1979).

A comparison of the long term survival of the marker strain of race 1 in natural and sterilized soil indicated that the organisms could survive more than a year in sterilized soil but could survive about 4 months only in natural soil. The survival in natural soil in the absence of hosts for 4 months suggested carryover of the pathogen from one crop to the next through soil. Eggplant in West Bengal is cultivated at least 3 times in a year: as summer crop, as kharif crop, and as rabi crop in the same field. Apparently the pathogen surviving in soil could easily infect the next crop. The situation under field conditions is more conducive for survival because the debris of the previous crop are available

and favours growth and multiplication of the pathogen for a long time. The results of this study clearly indicated that race 1 of P. solanacearum can survive for a long time in seed as well as in soil and the primary source of inoculum in nature is provided by either the seed or the soil or both. There are also several reports of the survival of the pathogen in solanaceous weed hosts (Kelman, 1953; Buddenhagen, 1965; Buddenhagen and Kelman, 1964), but this aspect of survival was not considered in this study.

It is now well established that the control of bacterial wilt caused by P. solanacearum by such practices as crop rotation, manipulation of cultural methods, and chemical treatment of infested soils has been only partially successful or prohibitively expensive (Kelman, 1953; Kelman, 1976; Thurston, 1976). It is also known that control through the development of disease resistance is the only method that offers a practical solution to most of the diseases caused by P. solanacearum (Kelman, 1976; Thurston, 1976). The remarkable success of wilt resistant tobacco varieties in the USA and peanut varieties in Indonesia has provided the hope for finding sources of and breeding for resistance to bacterial wilt of solanaceous vegetables including eggplant. The search for varieties of eggplant resistant to bacterial wilt in different countries, however, has not been as successful as has been done on tobacco and peanut. Although, the occurrence of resistant cultivars has been reported in the USA, Puerto Rico, the Philippines and Ceylon, conflicting information is available with respect to Indian cultivars and the performances of certain

recognised resistant cultivars in tropical countries including India (Srinivasan et al., 1969; Rao et al., 1976; ~~Thurston, 1976~~; Thurston, 1976; Sitaramaiah et al., 1981).

In the present investigation sixtythree Indian and eighteen Bangladeshi cultivars were screened for resistance under natural conditions of infection in the field. Twentyseven Indian cultivars including previously recognised resistant cultivars like Pusa Purple Cluster, Improved Muktakeshi, Green Long, and experimental lines of Sm, K, B and KNG showed wilt incidence less than 15% and were considered as resistant. Similarly, eight Bangladeshi cultivars including Rajshai No. 6 and 9, and Khatkhatia exhibited good resistance to bacterial wilt. The bacterial strain isolated from the wilted plants or from soils of Kalyani and Mondouri farms was always race 1 of P. solanacearum. When these cultivars which showed resistance to wilt under natural conditions were exposed to heavy load of artificial inoculation by race 1, none showed resistance to the pathogen. Only Improved Muktakeshi exhibited a low level of resistance.

It was of interest to note that cultivars Pusa Purple Cluster, Improved Muktakeshi, Green Long, Banaras Giant and experimental lines such as Sm, K or KNG which were considered as resistant to P. solanacearum by previous workers (Rao et al., 1976; Sitaramaiah et al., 1981; Khan, 1974; Mukherjee and Mukhopadhyay, 1982; Gopalkrishnan and Gopalkrishnan, 1985) have been found to be susceptible under the conditions of West Bengal. Failure of resistance of Pusa Purple Cluster, Improved Muktakeshi, SM lines

and several other germplasms to P. solanacearum, under agro-climatic conditions of Kerala (Srinivasan et al., 1969, Sheela et al., 1984; Jessykutty and Peter, 1986, Karnataka (Siddarama Gowda et al., 1974) and Orissa (Das and Panda, 1985) has been reported previously. The results of this work are in confirmation of these earlier works and suggested differential response of the Indian cultivars under different agro-climatic conditions of screening.

Failure of resistance of recognised foreign resistant cultivars under Indian conditions of field testing has been reported previously. In this respect, Rao et al. (1976) reported that the Philippino varieties Dingras Multiple Purple and Sinampiro were resistant, but the cultivars of Dumagneta Long Purple (Philippino), Kopek (Javanese) and Matle (Ceylonese) were susceptible under Bangalore conditions. The reported resistant lines Dingras Multiple Purple and Sinampiro, however, wilted under the warm humid climate of Kerala (Sheela et al., 1984; Jessykutty and Peter, 1986). Similar results were obtained with tomato; all the 22 resistant cultivars collected from Hawaii, Philippines, North Carolina and Ceylon were found to be susceptible under Indian condition (Rao and Tikoo, 1975; Bedekar, 1977). It is evident from these results that varieties developed with high levels of resistance at a given location may not survive in different geographic areas because of the presence of specific strains of the pathogen or because resistance is not expressed under certain environmental conditions. It is now well established that races of the pathogen differ markedly in their

virulence to different breeding lines of tomato, eggplant and potato (Kelman, 1976). Apparently, screening of large number of eggplant germplasms for stable resistance to different races of the pathogen singly or in combination under different agro climatic conditions should be initiated through International co-operation.

Since suitable resistant cultivars could not be identified in screening tests, an attempt was made to control the wilt of eggplant by manipulation of cultural practices. The results of this investigation clearly indicated that application of high doses of N:P:K fertilizers could significantly reduce the wilt incidence of eggplant caused by inoculation of race 1 under field conditions. The nitrogen used in the present study was in the form of urea and the highest dose applied (120 kg/ha) decreased wilt and was also stimulatory to vegetative growth and fruit yield parameters of the test eggplant cultivar Improved Mukta-keshi. The highest dose of P (80 kg/ha) significantly reduced wilt incidence and stimulated vegetative growth and certain yield parameters, but not all. Increased doses of K (80 kg/ha) although reduced wilt incidence, had little or no effect on vegetative growth and yield parameters tested.

The results suggested significant reduction of wilt of eggplant by the application of high doses of N, P and K to soil. Decrease in disease incidence with high nutrition level and especially of N-nutrition has been reported previously (Gallegly, 1948; Gallegly and Walker, 1949). Significant reduction of the wilt of tomato, potato, chilli and eggplant by the application

of high doses of urea has also been reported previously (Smith and Clayton, 1943; Kincaid, 1950; Clairon, 1984). The results of the present investigation also confirmed these earlier reports. Control trials of eggplant cultivars against race 1 of P. solanacearum by the application of heavy doses of urea, phosphorus and potassium under different agroclimatic zones warrant serious consideration. Effects of different forms of N-fertilizers on the incidence of wilt of eggplant also warrant further investigation.

Experiments concerned with the amendment of fields with different oilcakes indicated significant control of the wilt of eggplant by some, but not by others. Thus, application of non-edible neem and karanja oilcakes could significantly reduce the incidence of eggplant wilt as compared to control plants which received no oilcake amendments. The edible oilcakes like mahua and mustard failed to give any significant control of the disease. Besides reduction of wilt incidence, both neem and karanja cakes were stimulatory to vegetative growth and fruit yield parameters of the eggplant cultivar Improved Muktakeshi. In contrast, mahua and mustard cakes although not inhibitory to wilt incidence, caused stimulation of growth and yield parameters of the test cultivar. Significant reduction of tobacco and tomato wilt caused by P. solanacearum by the application of peanut meal (Van der Poel, 1938; Van Eek and Thung, 1950; Tanaka, 1976) and rubber seedmeal (Jayaprakash and Rajan, 1975) has been reported previously. It is apparent from the present study that high doses of neem or karanja caused a low but significant reduction of the wilt of

eggplant under field conditions. Evidence for reduction of wilt by application of urea has been mentioned previously in this report. The combined effects of amendment of soil with urea and neemcake warrant immediate investigation.

The mechanism of reduction of eggplant wilt by application of organic matters like oilcakes is poorly understood. An organic substrate added to soil is a source of food for the micro-biota. Reductions in root disease by organic amendments are mainly through stimulation of antagonists and consequent reduction in inoculum density or suppression of growth and survival of the pathogen in soil. Stimulation of population of Bacillus subtilis and production of an antibiotic which suppresses Streptomyces scabies in soils amended with soybean hay has been reported (Weinhold and Bowman, 1968). The effects of oilcake fertilization on the rhizosphere microflora of eggplant in relation to growth and survival of race 1 of P. solanacearum warrant investigation for a better understanding of the biology and control of the disease. The results with in vitro soil experiments, using the marker strain, however, indicated stimulation of growth (population density) and persistence of the pathogen in oilcake or host residue amended soils. The apparent discrepancy between the laboratory and field data could be due to disturbances in textural profile of the soils kept in bottles as well as breakdown of ecological equilibrium between soil microflora and the introduced pathogen in isolated soil samples. A critical investigation is necessary in this respect.

It is apparent from the results that field irrigation regimes could significantly affect the incidence of wilt of eggplant caused by race 1 of P. solanacearum. It was of interest that both frequent (5 days interval) and delayed (20 days interval) irrigation schedules could significantly decrease the wilt incidence under field conditions. It is also evident that in soils receiving more irrigation water (5 days interval), the vegetative growth and fruit yield of eggplant were significantly higher than the plants receiving less water. Apparently, the high moisture level of the soil increased the vigor of eggplant and probably rendered the plants more resistant to infection by race 1 of P. solanacearum (Rotem and Palti, 1969). Plants grown in drier soils with less irrigation were less vigorous and possibly more susceptible to bacterial wilt. Predisposition of plants to infection by varying irrigation schedule of fields has been reported by several previous workers (Rotem and Palti, 1969; Griffin, 1969; Cook and Papendick, 1972).

In experiments concerned with behaviour of the marker strain in soil environment, it was observed that the pathogen could grow and survive well in high soil moisture conditions. Both too dry and too wet soils were not conducive for its growth. The low incidence of wilt in drier soils could be due to the direct effect of drying on the pathogen or due to indirect effect on the host vigor. The data of field irrigation experiments further supported the laboratory data and suggested poor wilt incidence in both high and low moisture levels of the soils. Although a low level of

control was achieved by variation in irrigation schedules, the manipulation of irrigation water along with other cultural practices such as N-fertilizer and organic amendments of soil should be exploited for controlling eggplant wilt by race 1 under field conditions.

The results of this investigation and that compiled in other laboratories of the world clearly indicated that race 1 of P. solanacearum, the causal agent of eggplant wilt, can persist in seed, in soil, in crop debris and in association with diverse solanaceous weed hosts. According to Van der Plank (1963) the bacterial wilt disease is a "Simple interest disease" and the amount of disease or epiphytotics depends mainly on the amount of inoculation present at the beginning of the season. Epidemics of a simple interest disease within a single season is represented by the equation :

$$dx/dt = QR(1-x)$$

where, X = the proportion of diseased tissue on plants at time t ; Q = the initial inoculum; and R = the infection rate. The infection rate R for most soil-borne diseases has been found to be low or negligible (Van der Plank, 1963; Garrett, 1970; Fry, 1977). Reduction of initial inoculum (Q) therefore, would strategically decrease the epiphytotics of bacterial wilt of eggplant.

Eradication of the pathogen from the seeds by simple surface sterilization procedures has been adopted in the present study to

test the effects of seed dressing with chemicals on subsequent decrease of the pathogen in vitro and in the field. It is evident from the results that treatment of eggplant with streptomycin could successfully eradicate race 1 from the seed surface without affecting the viability of the seed. Seedlings raised from streptomycin treated seeds exhibited significantly less incidence of wilt due to race 1 as compared to untreated control or seeds treated with captan or vitavax.

Seedling raised from streptomycin treated seeds showed good vegetative growth and high yield of fruits as compared to other treatments and non-treated controls. The results further suggested that streptomycin concentrations used in this study could eradicate the pathogen without any phytotoxicity. Several early workers have reported that race 1 of P. solanacearum is a gram-negative rod and is highly sensitive to broad-spectrum antibiotic streptomycin (Winstead and Kelman, 1960; Perez, 1961; El-Goorani et al., 1978). Control of the wilt of tomato (Mondal and Mukherjee, 1978) and eggplant (Dutta and Verma, 1969) by the application of streptomycin to the seedling roots has been reported previously.

The data of this present work provided further evidence of significant control of eggplant wilt by pre-sowing treatment of seeds with streptomycin. Dutta and Verma (1969) reported reduction of wilt incidence of eggplant by treatment of the seedlings with streptomycin prior to transplantation. While seed dressing decreases seed-borne pathogens, the seedling treatment is aimed at combating the pathogen present in the rhizosphere soil. The

efficacy and cost benefit ratio of seed treatment before sowing and seedling treatment before transplantation, on the eggplant wilt caused by race 1 of the pathogen need further investigation. Search for low cost newer antibiotics which are more effective than streptomycin is another approach which should be continued (Katsuyoshi et al., 1978). Several chemicals applied to soil as drench are known to reduce the wilt of potato and tomato (El-Goorani et al., 1978; Enfinger et al., 1979). Efficacy of such or other nonphytotoxic chemicals, standardization of the doses and application schedules are worth trying for reducing the inoculum density of the pathogen in soil.

It is apparent from the above discussion that race 1 of P. solanacearum is a very successful pathogen with respect to its survival efficiency from one season to another. It is also apparent from the present findings that no single method can effectively control the disease under field conditions. Apparently, integrated disease management approaches should be adopted for the effective control of the disease. Use of available resistant sources, disease-free seeds, seed and seedling treatment with suitable antibiotics or chemicals, application of high doses of N:P:K with urea as N-source, amendment of soil with neemcake and proper management of irrigation schedules in combination should be tested in different agroclimatic conditions against different races of the pathogen. Well planned integrated disease management approach only can control the eggplant wilt and provides unhampered supply of this nutritious vegetable to the millions of people in Africa, Asia and Europe.

SUMMARY AND CONCLUSION

Wilt of solanaceous vegetables including eggplant incited by Pseudomonas solanacearum causes enormous yield loss to the growers in many parts of the world including India. Despite heavy economic loss, the control of the disease in many less developed countries is poorly achieved because of inadequate knowledge of the nature of race, primary source of infection and mode of survival of the pathogen in the absence of host plants. In this work attempts have been made to identify the race of the eggplant pathogen under West Bengal conditions and to determine the survival and transmission of the pathogen in eggplant seeds and in soil using a marker strain. Attempts were also made to control the disease in the field through cultural practices like varying doses of N:P:K fertilizers, organic manures and irrigation. A large number of Indian and Bangladeshi germplasms were screened for selection of resistant genotypes as source of breeding materials.

For identification of the race, the morphological, biochemical and pathogenicity characteristics of the eggplant isolates were investigated. All isolates of eggplant utilized hexose alcohols like mannitol and sorbitol, and produced acids from disaccharides such as maltose and lactose. Moreover, the eggplant isolates were highly virulent to tomato cultivars Marglobe Supreme and Bony Best. It was concluded that the pathogen causing eggplant wilt in West Bengal was P. solanacearum race 1.

For survival study, a double marker strain of race 1 resistant to 2 mg/ml streptomycin and 1 mg/ml erythromycin was developed. The stability of the mutant nature of the selected strain was checked periodically by growing it on PDA containing the respective doses of the two antibiotics. Experiments concerned with characterization of morphological and biochemical properties of wild type cells and the mutant isolate indicated that the latter was comparable to wild type cells except antibiotic resistance ability. The pathogenicity potential of the marker strain was also found to be comparable with that of the wild type cells.

Seed pathological studies clearly indicated that the pathogen was present on the surface of eggplant seeds. Seed samples of germplasms collected from India and Bangladesh yielded P. solanacearum race 1 along with other microorganisms when plated on PDA in petridishes. Experiments concerned with the survival of the double marker strain on the seeds of eggplant clearly indicated that the pathogen could survive for prolonged period on the surface of seeds. Survival on the seed surface, however, was poor when the inoculated seeds were incubated at high temperature (40 C) and high relative humidity (34% or above) conditions of storage.

Recovery of double marker mutant isolate of race 1 from soil at zero hour of introduction was investigated by soil dilution plate technique using PDA containing 2 mg/ml streptomycin and 1 mg/ml erythromycin. The results indicated that very high or very low initial cell density caused low recovery of cells from soils. When the initial cell density of 8×10^7 cells/g of soil was used, 50 to 70% of the cells could be recovered. This

initial cell density was used in studies concerned with survival ecology of the pathogen in soil.

Experiments designed for examining the survival of the marker strain in soil indicated that the population of the marker strain survived better in sterilized soils kept in bottles with lids closed. It was noted that the rate of decline of the population was very slow in sterile soils as compared to natural soil. Slow drying of soils by opening the lids of the bottles significantly reduced the survival of the marker strain.

The effects of soil temperature on the survival ability of the marker strain was examined by exposing the inoculated soils to different temperature conditions, such as, 15, 25, 30 or 40 C adjusted to 30% or 60% W.H.C. It was observed that the marker strain was very sensitive to 40 C and the population declined very rapidly. At lower temperatures (15 to 30 C), the population declined slowly and after about a month the population density was one log unit less than the initial inoculum added to natural soil. Similar temperature effects were obtained irrespective of the adjustment of the soils to 30% or 60% W.H.C.

Effect of soil moisture on the population dynamics of the double marker strain of race 1 was determined. The percentages of soil water content used were 10, 14, 17, 20, 23 and 25. It was evident that the marker strain survived well in high moisture. Dry soils were not conducive for the survival of the pathogen. Furthermore it was observed that the marker strain exhibited better survival in sterilized soil.

Effect of soil flooding on the survival of the marker strains

of race 1 was investigated. Inoculated soil samples were flooded with water so that water levels remained 0.8, 1.0, 1.2 and 1.5 cm above the surface of the soil. It was noted that the flooding caused significant decline in population density. The rate of decline of the population density was more in flooded natural soil than in flooded sterilized soils. However, increase of the depth of standing water above the surface of the soil caused significantly more decrease of the population. It was also noted that in flooded soils progressively more percentages of the marker cells could be isolated from the water phase than the soil phase.

The effects of inorganic fertilizers and organic manures on the population of the marker cells of race 1 were examined. Soils were amended with different doses of KNO_3 , $(\text{NH}_4)_2\text{SO}_4$, urea, cowdung, and oilcakes of linseed, mahua, mustard and neem prior to introduction of the marker strain. The results indicated that amendment of soils with $(\text{NH}_4)_2\text{SO}_4$ stimulated the survival whereas urea (1.5%) and KNO_3 (2%) caused rapid declination of the population density. The rate of decline of the marker strain in unamended soil was higher than that of the decline in soils amended with $(\text{NH}_4)_2\text{SO}_4$. Soils amended with cowdung or different organic manures, were moderately stimulatory to the growth and survival of the marker strain of race 1.

Experiments concerned with the amendment of soil with leaf residues of host and non-host plants clearly indicated that the populations of the marker strain was significantly stimulated after amendment with host-leaf residues.

Results concerned with long-term survival of the pathogen in soil using the marker strain of race 1 indicated that the organism can survive for more than one year in sterilized soil, but in natural soil it could not be detected after 5 months of incubation. The studies concerned with the ecology of survival of the pathogen clearly indicated that the pathogen can survive on seeds of eggplant and also in soil during the inter-cropping period.

Sixtythree Indian and eighteen Bangladeshi cultivars of eggplant were screened to identify the resistance source under field conditions. In the final year, the seedlings were transferred to field plots with previous history of eggplant wilt due to race 1 and the disease incidence due to natural infection was recorded. All cultivars showing natural infection incidence more than 15% under natural field conditions were discarded. The results showed that the varieties Long-1, K-8(1), Garia, Green Long and KNG-1 from India and varieties Rajshahi No. 6, Dohazari and Singnath from Bangladesh showed 2.50% of wilt incidence under natural conditions of infection. The results also showed that the Indian cultivar Pusa Purple Long was highly susceptible and cultivars Pusa Purple Cluster, Improved Muktakeshi and Type 3(3) Long were moderately resistant (5.0% wilt) under natural infection condition of the field.

All cultivars which showed resistance under natural field conditions were further tested for their resistance in the following rabi season by inoculation with race 1 under field conditions. Seedlings were inoculated by root-clipping method

before transplantation and again at the age of 1 month by soil drenching. The results showed that all cultivars including Pusa Purple Cluster which exhibited resistance under natural conditions were highly susceptible to the pathogen. The cultivar Improved Muktakeshi with a mean wilt percentage of 20 showed a moderate level of resistance in this experiment.

The effect of different doses of fertilizers on the wilt incidence of eggplant caused by race 1 was determined in field trials using Improved Muktakeshi as test cultivar. The doses used were 60, 90 or 120 kg N/ha in the form of urea and 40, 60 or 80 kg/ha for each of P (in the form of single superphosphate) and K (as muriate of potash) in 27 combinations. The results showed that wilt incidence was reduced with the increase in nitrogen concentration. Besides, the increased doses of nitrogen caused enhanced vegetative growth and increased fruit length, fruit diameter and total yield of fruit of eggplant significantly. Increased doses of phosphorus caused reduction of wilt and also caused increase of growth and fruit yields. Increased doses of K were found to have little or no effect on wilt incidence as well as growth and yield characters.

Effect of amendment of soil with oilcakes of karanja, mahua, mustard or neem in combination with basal dose (90:60:60 kg/ha) on the wilt incidence of cultivar Improved Muktakeshi caused by race 1 was investigated in field plots. It was noted that the non-edible oilcakes, neem and karanja caused significant reduction of wilt incidence. These cakes also increased the growth and yield parameters. It was evident that the edible oilcakes mahua

and mustard did not cause any appreciable control of the wilt incidence.

The effect of varying frequency of irrigation in the field on the incidence of wilt of the eggplant cultivar Improved Muktakeshi caused by race 1 of the pathogen was investigated. The inoculated seedlings were transplanted to fields and irrigated at an interval of 5, 10, 15 or 20 days. The results showed that irrigation at 5 days interval significantly reduced wilt incidence. Moreover, with high frequency of irrigation vegetative growth and fruit yield per plant were significantly higher than the plants receiving less water. Irrigation schedule of 20-day interval also caused significant control of the wilt, but vegetative growth and fruit yield was reduced. Other irrigation schedules had little or no controlling effect on the wilt caused by race 1 of P. solanacearum. It was evident that too dry or too wet soils could significantly reduce wilt incidence.

Effects of treatment of seeds with different doses of captan, vitavax and streptomycin on the eradication of the pathogen from the seed surface was investigated under laboratory condition. Treatment of seeds with 250 µg/ml of streptomycin for 30 minutes could effectively reduce the percentage of infection upto 4% only as compared to 100% infection in untreated control seeds.

Effect of pre-sowing treatment of seeds of Improved Muktakeshi with captan, vitavax and streptomycin on the incidence of wilt of eggplant caused by race 1 was investigated under field conditions. The results showed that the seedlings raised from streptomycin-treated seeds exhibited significantly less incidence

of wilt as compared to untreated control or seeds treated with captan or vitavax. Seedling raised from streptomycin-treated seeds showed good vegetative growth and high yield of fruits as compared to other treatments and non-treated controls. It was concluded that streptomycin concentration used in the present study could eradicate the pathogen without any phytotoxicity.

It was apparent from this investigation that race 1 of P. solanacearum can persist in seed, soil or crop-debris and is carried over from season to season. It was also evident that integrated disease management approaches should be adopted for the control of wilt disease of eggplant caused by race 1 of P. solanacearum. Use of available resistant sources, disease-free seeds, seed or seedling treatment with suitable antibiotics, application of high doses of N:P:K with urea as N-source, amendment of soil with neemcake and proper management of irrigation schedules in combination should be adopted for the control of bacterial wilt of eggplant caused by P. solanacearum.

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