



UNIVERSITI PUTRA MALAYSIA

**SOME ASPECTS OF EFFICACY AND RESIDUES
OF MANEB AND MANCOZEB IN SELECTED VEGETABLES**

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**SOME ASPECTS OF EFFICACY AND RESIDUES
OF MANEB AND MANCOZEB IN SELECTED VEGETABLES**

By

S. RAMASAMY

**Thesis Submitted in Partial Fulfilment of the
Requirements for the Degree of Master of Agricultural Science
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LIST OF ABBREVIATIONS

ADI	- acceptable daily intake
a.i.	- active ingredient
b.a.	- <i>bahan aktif</i>
b.s.j.	- <i>bahagian sejuta</i>
ca.	- approximately
CMA	- corn meal agar
cv	- coefficient of variation
d	- day
EPPO	- European and Mediterranean Plant Protection Organization
FAO	- Food and Agriculture Organization of the United Nations
h	- hour
IUPAC	- International Union of Pure and Applied Chemistry
L	- litre
LSD	- least significant difference
mo	- month
MRL	- maximum residue limit
PDA	- potato dextrose agar
PHI	- preharvest interval
ppb	- parts per billion
ppm	- parts per million
R.H.	- relative humidity
S.E.	- standard error
ug	- microgram
ul	- microlitre
w	- week
WA	- water agar



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The use of the fungicides, maneb and mancozeb on selected leafy and fruit vegetables was studied with the view to ascertain their efficacy and degradation of their residues on these crops. The phenology of these vegetables and the disease problems encountered were also verified. Most of the test crops were prone to disease infection if not treated with suitable fungicides. Spinach was susceptible to downy mildew infection while spring onion was predisposed to leaf blight and leaf tip dieback. In leaf mustard and garland chrysanthemum, however, no serious disease infection was encountered. Fruit vegetables were susceptible to several serious diseases. Tomato had early blight, late blight, Fusarium wilt and anthracnose. Cucumber was infected by downy mildew and anthracnose while chilli was prone to anthracnose, Sclerotium wilt and mosaic virus infection.

Maneb and mancozeb when used weekly at recommended rates of 0.27% a.i. and 0.2% a.i. respectively were reasonably effective against diseases of leafy vegetables. In tomato, recommended rates of these fungicides, applied at 4-day intervals, gave reasonable control of late blight. In tomato, weekly applications were ineffective for late blight control especially during high disease pressure. However, reasonable control was achieved at 4-day intervals.



In cucumber, downy mildew infection was effectively reduced by the application of these fungicides. Similarly, anthracnose infection on chilli fruits was checked with these fungicides applied weekly at recommended rates.

Residues of maneb and mancozeb in spinach exceeded the maximum residue limit of 5 ppm at harvest even when last treated 2 weeks before harvest. In leaf mustard and spring onion, a 14 day PHI appeared a prudent recommendation with these fungicides. This was not a serious problem in tomato and cucumber as even after 3 days the amount detected was less than 1 ppm in all the treatments. Hence a 3 day PHI can be recommended for these crops. In chilli, however, 3-day intervals were insufficient as about 10 ppm was still detected. A 14 day PHI appeared more prudent. Other fungicides such as chlorothalonil, propiconazole or metiram could be used in place of maneb and mancozeb before harvest.



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**BEBERAPA ASPEK BERKENAAN DENGAN KEMUJARAPAN DAN SISABAKI
MANEB DAN MANCOZEB KEATAS SAYUR-SAYURAN YANG TERPILIH**

Oleh

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Penggunaan racun kulat, maneb dan mancozeb telah dikaji untuk menentukan kemujarapan dan kehilangan sisabaki racun-racun ini ke atas tanaman sayur-sayuran jenis berdaun dan buah. Fenologi dan jenis penyakit yang biasa didapati juga dipastikan. Sebahagian tanaman ujian didapati diserang oleh penyakit sekiranya tidak dirawat dengan racun-racun kulat yang sesuai. Bayam mudah diserang oleh penyakit kulapuk downy dan daun bawang biasanya diserang oleh penyakit hawar daun dan mati pucuk. Tiada sebarang penyakit yang serius didapati ke atas tanaman sawi putih dan 'tong ho'. Sayur-sayuran jenis buah seperti tomato sentiasa diserang oleh penyakit hawar, layu Fusarium dan antraknos. Timun diserang oleh penyakit kulapuk downy dan antraknos sedangkan cili didapati mudah dirosak oleh penyakit antraknos, layu Sclerotium dan mozek daun.

Maneb dan mancozeb didapati memberi kesan yang baik ke atas sayur-sayuran jenis berdaun apabila digunakan seminggu sekali dengan kadar 0.27% dan 0.2% bahan aktif. Rawatan ke atas tomato terpaksa dibuat 4 hari sekali kerana semburan seminggu sekali tidak dapat mengawal penyakit hawar dengan baik. Penyakit kulapuk downy ke atas tanaman timun dan antraknos ke atas tanaman cili dapat dikawal dengan racun kulat ini pada kadar yang disyorkan seminggu sekali.



Paras sisabaki racun kulat maneb dan mancozeb ke atas bayam didapati melebihi had maksimum iaitu 5 ppm pada masa dipetik walaupun rawatan terakhir dibuat 2 minggu sebelum mengambil hasil. Lat tempoh selama 14 hari sebelum mengambil hasil boleh disyorkan bagi tanaman sawi putih dan daun bawang. Tanaman cili yang biasanya dirawat 3 hari sekali juga didapati menghadapi masalah sisabaki yang berlebihan. Di sini juga 14 hari disyorkan sebagai tempoh masa sebelum mengambil hasil. Tanaman-tanaman lain seperti tomato, daun bawang dan tlmun, ini tidak mendatangkan sebarang kesulitan dan masa menunggu selama 3 hari selepas rawatan terakhir adalah mencukupi. Racun-racun kulat jenis lain seperti chlorothalonil, proplconazole atau metiram disyorkan sebagai gantian kepada maneb dan mancozeb pada masa sebelum mengambil hasil.



CHAPTER 1

INTRODUCTION

In Peninsular Malaysia, ethylene bis(dithiocarbamate) (EBDC) fungicides, e.g. maneb and mancozeb, are used widely in vegetable cultivation (Department of Agriculture, 1987). Their use on leafy vegetables such as spring onion (*Allium fistulosum* Linn.), spinach (*Spinacia oleracea* Linn.) or 'por choy' and garland chrysanthemum (*Chrysanthemum coronarium* Linn.) or 'tong ho' against specific diseases have not been given much attention. These crops which are usually grown in the cooler regions, and some, under transparent plastic shades are frequently treated with such fungicides, often with doses higher than those recommended by the manufacturers.

EBDC fungicides are also used extensively on fruit vegetables such as cucumber (*Cucumis sativus* Linn.), chilli (*Capsicum annum* Linn.) and tomato (*Lycopersicon esculentum* Mill.) (Department of Agriculture, 1984). Diseases like anthracnose, downy and powdery mildew have been reported on cucumber while tomato is highly prone to early and late blight infection (Johnston, 1959). Fungicides such as maneb and mancozeb are reasonably effective for the control of some of these diseases and are presently standard control recommendations (Department of Agriculture, 1981). However, many farmers claim that these fungicides are not as effective as they used to be (Department of Agriculture, 1987). Higher doses of these fungicides were applied and in some cases, more frequently. Farmers were also reluctant to switch to other suitable fungicides.

When such practices were found to be increasing, there were reasons to believe that some of the vegetables so treated could contain excessive residues of these fungicides at harvest. Studies conducted in 1986 indicated this to be true. Samples of spring onion, chinese celery, tong ho, spinach and sweet pea from the Cameron Highlands vegetable area were found to contain more than the maximum residue limit (MRL) permitted under the Food



Regulations, 1985 (Ministry of Health, 1986). Neighbouring importing countries have also been monitoring such residues and several consignments of vegetables from Peninsular Malaysia have been rejected since early 1987 after excessive levels of these fungicides were detected (Ministry of Environment, 1987).

Studies on pesticide residues in food, apart from being expensive and laborious, have not been given sufficient importance. It was only after the rejection of commodities such as cocoa and vegetables by the importing countries for reasons of excessive residues that it was viewed more seriously. Even then, local information on residues of such fungicides on these vegetables were rather limited. Available information was based mainly on studies conducted under temperate conditions. Preharvest intervals (PHI) recommended by the Department of Agriculture (1981), such as 14 days for maneb and mancozeb on vegetables, were based to some extent on studies conducted under temperate conditions. Many farmers viewed this as impractical here and were ignoring these standards (Department of Agriculture, 1987).

The Food Regulations, promulgated in 1985, were meant to control such residues in food by stipulating MRLs for the major food crops. The Pesticides Act, 1974 also has provisions for such control. These regulations have been formulated recognising the importance of protecting the health of the public. Moreover, regulatory authorities also realise the adverse effects such contaminated produce can bring about if exported. However, the limited information presently available locally has made it difficult to suggest practical measures that could be adopted to minimise or overcome these problems. This issue now requires practical solutions. The Department of Agriculture, on its part, has intensified its efforts to gather more information, particularly, the efficacy and use of EBDC fungicides, their residues and suitable preharvest intervals.

Other aspects such as the phenology of specific vegetables where fungicide use posed problems; the diseases encountered; whether these fungicides are less efficacious and how they can be used judiciously to obtain quality produce, also require attention. Hence, these studies are to understand better the use of two EBDC fungicides, maneb and mancozeb, with a view to minimising problems arising from their use. The specific objectives are:-

1. to study the phenology of selected vegetables such as spinach, spring onion, leaf mustard and garland chrysanthemum and verify the disease problems encountered in these vegetables which warrant the use of these fungicides;
2. to evaluate the efficacy of maneb and mancozeb as used on selected vegetables particularly with respect to dosage and frequency of application necessary for effective disease control and if there was any basis for increasing the dosages or the frequency of applications;
3. to study the residue levels and degradation pattern of these fungicides following their use on these crops and recommend suitable preharvest intervals which farmers could observe to ensure their produce do not contain excessive levels of these fungicides at harvest.

CHAPTER 2

REVIEW OF LITERATURE

Crop Phenology and Identification of Diseases

Spinach

Spinach (*Spinacia oleracea* Linn.) is a temperate crop which grows well in the cooler regions with temperatures between 12° and 25°C. This crop was reported to be sensitive to high temperature and daylength (Knott and Deanon, 1967). In temperate regions, this crop has been reported to be susceptible to downy mildew infection caused by *Peronospora spinaciae*, rust due to *Puccinia* sp. and smut (Heald, 1926). No local records are available on any disease infection on this crop.

Spring Onion

Spring onion (*Allium fistulosum* Linn.) is grown widely in Cameron Highlands particularly in the Tringkap and Kea Farm areas (Department of Agriculture, 1987a). In the lowlands, the red bulb variety, *Allium cepa* var. *ascalonium* Linn. Backer, is reported to be widely grown (Mohamed Zin and Hasmah, 1981). Soils with pH between 6.0-6.8 were found to be most suitable for spring onion cultivation. Proper drainage was essential for a good crop of spring onion as the bulbs tended to rot quickly under waterlogged conditions (Mohamed Zin and Hasmah, 1981).

Johnston (1958) reported that bulb rot caused by *Fusarium oxysporum* was one of the more common problems encountered in spring onion in the lowlands, while leaf blight due to *Alternaria porri* was the main disease problem in the highlands. Leaf tip dieback (also referred to as white or yellow tip) was another serious problem in onion cultivation. Mohamed Zin and Hasmah (1981) stated that the causal agent was yet to be confirmed. William and Liu (1976) had reported this symptom found on onion (*Allium cepa*) grown in Sabah to be caused by

Botrytis sp. The Tropical Development and Research Institute (1986) described similar symptoms where the leaves withered from the tip as neck rot caused by *Botrytis* sp.

Leaf Mustard

Leaf mustard (*Brassica juncea* Cosson) called 'kal choy' locally is another highland crop. It has been recorded in Peninsular Malaysia to be susceptible to leaf rot caused by *Sclerotium rolfsii*, club root disease associated with *Plasmodiophora brassicae* and soft rot due to a bacterium, *Erwinia carotovora* (Singh, 1980). Ko (1980) reported that this vegetable could be infected by downy mildew, due to *Peronospora parasitica*.

Garland Chrysanthemum (tong ho)

Garland chrysanthemum or tong ho is an annual herbaceous shrub-like plant. Its tender shoots were reported to be used as a vegetable (Burkill, 1966). No records of any disease infection on this crop in Peninsular Malaysia were available. In Sabah two fungi, *Cylindrotrichum* sp. and *Pithomyces chartarum* had been isolated from tong ho with leaf spot symptoms (Singh, 1980).

Tomato

Tomato is a herbaceous plant, procumbently branched and partly erect, bearing fruits in clusters. It grows best at 21-24°C day temperature and 15-20°C night temperature. High relative humidity, however, often causes serious losses in yield due to disease infection (Knott and Deanon, 1967). Being partly erect, it needed to be supported by staking or training using small stakes or strings to prevent the fruits from touching the ground surface. This also made easier, pruning which is necessary to obtain good yields (Department of Agriculture, 1982). The fruits may be harvested at three stages of maturity, namely, the mature green, the pink or breaker stage and the red-ripe stage (Knott and Deanon, 1967).

Tomato has been reported to be susceptible to several fungal diseases in Peninsular Malaysia. The most serious reported here is late blight caused by *Phytophthora infestans*

(Johnston, 1958; Ko, 1980). This was followed by others such as early blight due to *Alternaria solani*, collar rot due to *Pythium aphanidermatum*, powdery mildew due to *Oidium* sp., damping-off caused by *Corticium solani* and wilting due to *Sclerotium rolfsii* infection. There were also several fungi reported to cause leaf spot and fruit rot infection on tomato (Singh, 1980). Besides fungal diseases, wilt due to the bacterium, *Pseudomonas solanacearum* has been a major disease on lowland tomato (Department of Agriculture, 1981; William and Liu, 1976).

Cucumber

Cucumber is an annual herbaceous climber with tendrils. In Peninsular Malaysia it is grown widely in the lowlands, particularly in the southern state of Johore, in the districts of Kluang, Batu Pahat, Tangkak and Johor Bahru (Department of Agriculture, 1983). Other states which produced this vegetable were Kelantan and Selangor (Hasmah, 1982). Cucumber is a hairy, monoecious plant producing yellow bell-shaped flowers, 3-4 cm diameter. After pollination, the female flowers developed into fruits which were elongated and round with blunt ends. Optimum temperatures for cucumber were reported to be 29.4°C (Hasmah, 1982). Lower temperatures tended to retard growth while very low temperatures resulted in the seeds rotting. Well irrigated sandy soils with high organic matter content and a pH of 5 to 6.5 were reported to be most suitable for cucumber cultivation (Hasmah, 1982).

Common fungal disease problems reported on cucumber were seedling blight caused by *Pythium* sp. and *Rhizoctonia solani*. The hypocotyl was usually damaged. Another serious disease was downy mildew caused by *Pseudoperonospora cubensis* (Department of Agriculture, 1983). Moist conditions and low temperatures (20 - 25°C) were found to be favourable for this disease to develop. The infected leaves have mycelia of this fungus on the lower surfaces. The upper surface tended to become chlorotic with yellow patches at the infected portions. Downy mildew was reported to be most prevalent during the fruit formation stage (Heald, 1926). Severe infection could cause the fruits to be stunted and become off-flavoured. Powdery mildew caused by *Erysiphe cichoracearum* was also a serious fungal

disease problem in cucumber. Rounded white patches of powdery layer could be seen on the older, infected leaves. These patches enlarged, coalesced and spread over the whole leaf surface. These leaves eventually turned yellow to brown and fruit yield was reduced considerably (Hasmah, 1982). Anthracnose caused by *Colletotrichum lagenarium* and seedling blight due to *Pythium* sp. were less common. Wet conditions were reported to favour these diseases and when present, anthracnose could attack all parts of the cucumber plant (Heald, 1926).

Chilli

Chilli is an annual cultivated all year round and a field crop in places where a distinct dry season followed harvest. This was because it permitted the fruit to be sun-dried. In Malaysia, it was only a garden crop as the climate was not dry enough for field cultivation (Burkill, 1966). Chilli is reported to be a perennial but usually grown as an annual (MARDI, 1986). It is an erect, branched plant with flowers occurring singly in the axil of branches. The flowers were small, usually single and white or yellow in colour. The fruits are pendulous or erect, elongated and tapering to the end and red in colour when ripe. The fruit is used as a vegetable. Fruit production can range from 2 to 12 months, depending on the variety. Varieties popularly grown locally included the Kulai, Kelantan, Melaka and C-10, a variety developed recently by MARDI.

Chilli can be planted in the hot and wet conditions of the tropics and sub-tropics. The most suitable time was that just after the wet season following which a dry spell prevailed (Looi, 1981). Planting should preferably be done during the wet season with harvesting targeted for the dry season. Edaphic conditions are not critical as most soils are suitable for chilli cultivation. However, it is necessary that these soils should be well-drained. A pH of about 5.5 was found to be ideal (Department of Agriculture, 1983a).

Chilli has been reported to be susceptible to diseases like anthracnose caused by *Colletotrichum capsici* and *Gloeosporium* sp., bloom blight (*Choanephora cucurbitatum*), leaf

curl virus, mosaic virus, bacterial wilt (*Pseudomonas solanacearum*), leaf spot (*Cercospora capsici*), powdery mildew (*Leveillula taurica*), wilting due to *Sclerotium rolfsii*, root rot due to *Fusarium* sp. and stem blight (*Phytophthora capsici*) (Johnston, 1959; Singh, 1980).

Efficacy of EBDC Fungicides

The fungicidal properties of the salts of dithiocarbamic acid and its N-alkyl derivatives were discovered by Tisdale and Williams of E.I. du Pont de Nemours Co. in 1934 (Tisdale and Williams, 1934). These compounds and their properties were further described by Tisdale and Flanner (1942) and then Hester (1943) of Rohm and Haas Company who discovered the ethylene bis(dithiocarbamate) fungicides. One of them, maneb was introduced in 1950 by du Pont under the trade name 'Manzate' and by Rohm and Haas as 'Dithane M-22'. Another fungicide, mancozeb was introduced in 1961 by Rohm and Haas under the trade mark 'Dithane M45'. Both fungicides had been acknowledged as one of the most widely used fungicides in the world (Worthing and Walker, 1987). In Malaysia, they have been recommended on a wide range of crops such as vegetables, cocoa, rubber, pepper, oil palm, coconut, fruit trees, potato, tobacco and ornamentals (Department of Agriculture, 1981).

Maneb, the common name for manganese ethylenebis (dithiocarbamate) had a chemical structure as in Fig. 1. It is a yellow crystalline solid which decomposes without melting or heating. It is virtually insoluble in water and common organic solvents but dissolves in chelating agents such as sodium salts of ethylene diamine tetraacetic acid (EDTA) (Worthing and Walker, 1987). However, it cannot be recovered from such solutions. It decomposes on prolonged exposure to air or moisture and rapidly on contact with acids.

Mancozeb (Fig. 1) is a complex of zinc and maneb containing 20% of manganese and 2.55% zinc, the salt present (e.g. mancozeb chloride) being stated. The chemical (International Union of Pure and Applied Chemistry, IUPAC) name is manganese ethylenebis (dithiocarbamate) (polymeric) complex with zinc salt (Worthing and Walker, 1987). It is a greyish-yellow powder which is stable under normal storage conditions. It is rather insoluble

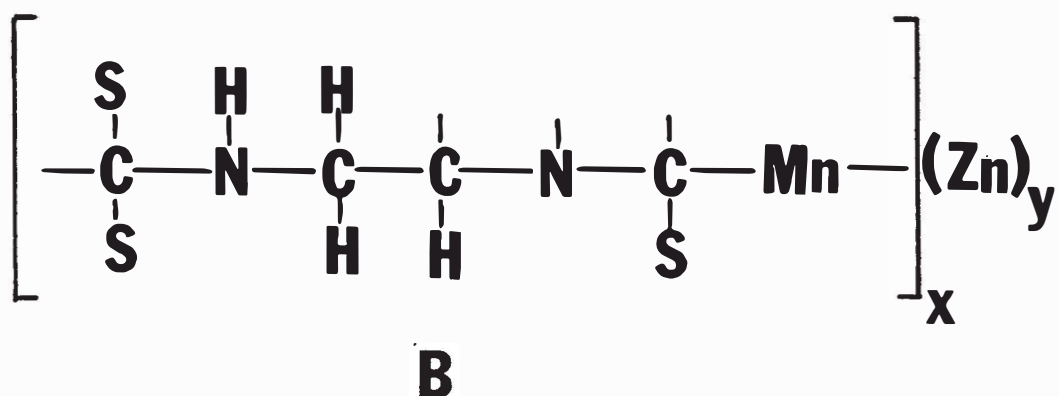
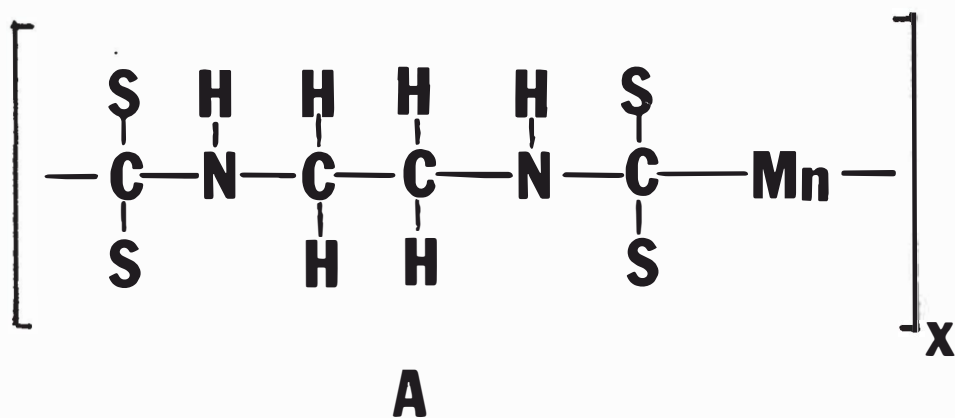


Figure 1: Chemical Structure of Maneb and Mancozeb.

A - Maneb - Manganese Ethylenebis (dithiocarbamate)
(Polymeric)

B - Mancozeb - Manganese Ethylenebis (dithiocarbamate)
(Polymeric) Complex with Zinc Salt.

in water and most organic solvents but is decomposed at high temperatures by moisture, and by acids. It is a protective fungicide, compatible with most other commonly used pesticides. However, Worthing and Walker (1987) reported that it might cause greasing with some emulsifiable concentrates.

Maneb is recommended for the control of many foliage diseases, particularly the blights of potato and tomato, downy mildew of lettuce and vines, and many fungal diseases of field crops, fruits, nuts, vegetables, ornamentals and turf. It is frequently used in combination with other fungicides to prolong the duration of protection they would otherwise provide (Worthing and Walker, 1987).

Mancozeb, introduced as a protective fungicide, was found effective against a wide range of foliar fungal diseases, including those caused by *Phytophthora infestans* on potato, *Fulvia fulva* on tomato and *Venturia* sp. on apples and pears (Alexopoulos, 1962). As with maneb, it is used in combination with other systemic fungicides to extend the duration of protection given to the crop (Worthing and Walker, 1987).

In temperate conditions, maneb has been proven to be effective for the control of stem anthracnose of lima bean caused by *Cercospora truncatum* (Chambers, 1966). Mancozeb has been reported to control bean rust caused by *Uromyces phasioli typica* (Lewis, 1971) and early leaf spot caused by *Cercospora arachidicola* on groundnut (Littrell and Lindsay, 1981). Phipps (1981) found that mancozeb was suitable for the control of late leaf spot caused by *Cercosporidium personatum*. White (1980) in his studies on the control of leaf blight disease on corn noted that mancozeb was effective in controlling both the northern corn leaf blight caused by *Dreschlera turcicum* and southern corn leaf blight caused by *Dreschlera maydis*. Maneb and mancozeb were both found effective in controlling anthracnose, downy mildew and soil rot of cucumber when applied weekly at 0.2 % active ingredient (a.i.) (Jones and Everett, 1966). Wade (1978) found that mancozeb at 1.12 kg a.i./ha applied weekly gave good control of early and late blight on potato. Lewis (1974) in his studies on the control of downy mildew

on broccoli caused by *Peronospora parasitica* found that mancozeb at 1.8 kg a.i./ha gave good control. Similar rates were also effective for the control of tomato anthracnose caused by *Colletotrichum phomoides*. Lewis (1978) and Abdel-Rahman (1978) had also shown that mancozeb at 1.8 kg a.i./ha was effective for the control of powdery mildew caused by *Erysiphe cichoracearum* and anthracnose (*Colletotrichum lagenarium*) on cucumber. Similar results were obtained by them in the control of anthracnose on pepper (*Capsicum annum*) and early blight on tomato.

The efficacy of maneb and mancozeb under local conditions has also been well documented. Ko (1979), Ko and Zabedah (1982) and Nordin *et al.* (1986) have found that maneb was suitable for the control of late blight of tomato caused by *Phytophthora infestans*. Ko's studies indicated that application of maneb at 0.15-0.3% a.i. during the middle of the crop growth was necessary and critical as tomato was most susceptible to late blight infection during this stage. Ko and Zabedah (1982) evaluated four fungicides including mancozeb for the control of late blight on tomato and found that weekly application of mancozeb at 0.25% a.i. was needed to achieve effective control. It was not possible to control this disease with less frequent sprays even at doses as high as 0.4% a.i. Nordin *et al.* (1986) did similar studies on tomato in Cameron Highlands and found that mancozeb in combination with metalaxyl (Ridomil MZ) at 0.2% a.i. applied weekly gave reasonable control of tomato late blight there. However, it was less efficacious during the wet season.

Besides tomato, maneb and mancozeb have been found to be used extensively on crops like cucumber, spinach, garland chrysanthemum and spring onion. These crops have been reported to be susceptible to specific foliar diseases. Cucumber was infected by downy mildew caused by *Pseudoperonospora cubensis*, powdery mildew caused by *Erysiphe cichoracearum* and anthracnose caused by *Colletotrichum lagenarium* (Johnston, 1959; Mah, 1982). Spring onion was believed by Johnston (1958) to be predisposed to the same pathogens found on leek (*Allium porrum* L.), i.e., *Peronospora* sp. causing downy mildew and *Alternaria porri* causing purple blotch. Maneb and mancozeb have been reported to be