

BEHAVIOUR OF METALAXYL AND MANCOZEB RESIDUES IN SOILS AND TOMATO



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BEHAVIOUR OF METALAXYL AND MANCOZEB RESIDUES IN SOILS AND TOMATO



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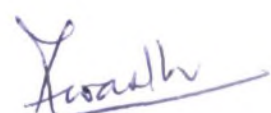
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This is to certify that the thesis entitled "BEHAVIOUR OF METALAXYL AND MANCOZEB RESIDUES IN SOILS AND TOMATO" submitted by Mr. T.H. HANUMANTHA RAJU, for the degree of DOCTOR OF PHILOSOPHY in SOIL SCIENCE AND AGRICULTURAL CHEMISTRY of the University of Agricultural Sciences, Bangalore is a record of research work done by him during the period of his study in this University under my guidance and supervision and the thesis has not previously formed the basis for the award of any degree, diploma, associateship, fellowship or other similar titles.

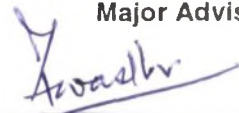
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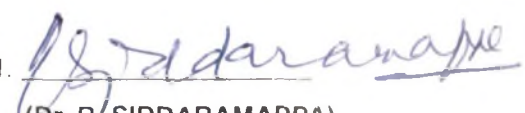
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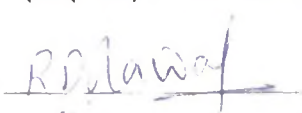
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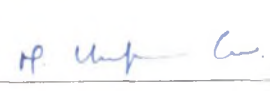
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CONTENTS

CHAPTER	TITLE	PAGE NO.
I	INTRODUCTION	1
II	REVIEW OF LITERATURE	3
III	MATERIAL AND METHODS	20
IV	RESULTS	38
V	DISCUSSION	79
VI	SUMMARY	94
VII	REFERENCES	98

LIST OF TABLES

SL. NO.	TITLE	PAGE NO.
1.	Analytical methods followed in soil analysis	21
2.	Physico-chemical properties of soils	22
3.	General properties of fungicides and metabolite under study	23
4.	Weather parameters of the experimental site	28
5.	Recoveries of metalaxyl, mancozeb and ethylenethiourea in soils and tomato	33
6.	Adsorption of metalaxyl in soils	39
7.	Adsorption isotherm constants of metalaxyl in soils	40
8.	Adsorption of ethylenethiourea in soils	42
9.	Adsorption isotherm constants of ethylenethiourea in soils	43
10.	Persistence of metalaxyl residues in different soil as affected by rate of application and moisture regime	45
11.	Persistence of metalaxyl residues in soils as affected by different temperatures and sterilization	46
12.	Equations explaining the kinetics of metalaxyl residues degradation in different soils	48
13.	Equations explaining the kinetics of metalaxyl residues degradation in different soils as affected by temperatures and sterilization	49
14.	Persistence of mancozeb residues in different soils as affected by rate of application and moisture regimes	51
15.	Persistence of mancozeb in soils as affected by different temperatures and sterilization	52
16.	Equations explaining the kinetics of mancozeb residues degradation in different soil affected by application rate and moisture regime	55

17. Equations explaining the kinetics of mancozeb residues degradation in different soils as affected by temperatures and sterilization	56
18. Persistence of ethylenethiourea (ETU) residues in different soils as affected by rate of application and moisture regimes	58
19. Persistence of ETU in soils as affected by different temperatures and sterilization	59
20. Equations explaining the kinetics of ETU residues degradation in different soils as affected by application rates and moisture regimes	61
21. Equations explaining the kinetics of ETU residues degradation in different soils as affected by temperatures and sterilization	62
22. Plant uptake and persistence of metalaxyl and ethylenethiourea (ETU) in tomato	64
23. Persistence of fungicides and its metabolite residues in Bangalore soil under tomato crop cover	66
24. Equations explaining the kinetics of fungicide residues degradation in Bangalore soil under tomato crop cover	67
25. Persistence of the residues of fungicides and metabolite in/on tomato foliage under field conditions	69
26. Persistence of the residues of fungicides and metabolite in/on tomato fruits under field conditions	70
27. Equations explaining the kinetics of fungicide residues dissipation and safety constants in/or tomato foliage and fruits	71
28. Persistence of the residues of fungicides and metabolite in soil following foliar spray of Ridomil-MZ on tomato crop	74
29. Equations explaining the kinetics of fungicide residues degradation in soil following foliar spray of Ridomil-MZ on tomato crop	75
30. Decontamination of fungicide residues from tomato fruits	77

LIST OF FIGURES

SL. NO.	TITLE	BETWEEN PAGE NO.
1.	Flow chart showing the residue analysis of metalaxyl and ETU from tomato	30-31
2.	Chromatograms showing (a) metalaxyl (standard) (b) tomato (control) (c) tomato fortified with metalaxyl	31-32
3.	Decomposition distillation apparatus for determination of mancozeb residues	32-33
4.	Chromatograms showing (a) ETU (standard) (b) tomato (control) (c) tomato fortified with metalaxyl-mancozeb (Ridomil - MZ)	35-36
5.	Adsorption of metalaxyl in soils	39-40
6.	Adsorption of ethylenethiourea in soils	42-43
7.	Persistence of metalaxyl residues in different soils as influenced by application rate	46-47
8.	Persistence of metalaxyl residues in different soils as influenced by moisture regimes	46-47
9.	Persistence of metalaxyl residues in soils as influenced by temperature	46-47
10.	Persistence of metalaxyl residues in soils as influenced by sterilization	46-47
11.	Persistence of mancozeb residues in different soils as influenced by application rate	52-53
12.	Persistence of mancozeb residues in different soils as influenced by moisture regimes	52-53
13.	Persistence of mancozeb residues in soils as influenced by temperature	52-53

14.	Persistence of mancozeb residues in soils as influenced by sterilization	52-53
15.	Persistence of ETU residues in different soils as influenced by application rate	59-60
16.	Persistence of ETU residues in different soils as influenced by moisture regimes	59-60
17.	Persistence of ETU residues in soils as influenced by temperature	59-60
18.	Persistence of ETU residues in soils as influenced by sterilization	59-60
19.	Uptake of metalaxyl and ETU by tomato after the application of metalaxyl-MZ to soil	64-65
20.	Persistence of fungicide residues in Bangalore soil under tomato crop cover	66-67
21.	Persistence of fungicide residues in/on tomato foliage under field conditions	69-70
22.	Dissipation of fungicide residues in/on tomato fruits under field conditions	70-71
23.	Persistence of fungicide residues in soil after spraying of Ridomil-MZ on tomato crop	74-75
24.	Decontamination of fungicide residues from tomato fruits	77-78

INTRODUCTION

I. INTRODUCTION

The economic cultivation of field crops suffer heavy losses from the pest infestation in tropical climate due to relatively heavy carry over of pests. It has been estimated that annual crop losses in India due to pests amount to over Rs. 6000 crores. These losses are quantitative as well as qualitative in nature. The losses due to fungal infestations are gaining importance in addition to losses due to insects from the point of crop production. Accordingly, plant diseases were estimated to account for 20 per cent annual loss on food production that contribute to the extent of Rs.1560 crores to the total loss due to pests (Vasantharaj David, 1992). Among the various approaches employed for pest control, the use of pesticide chemicals has been established to be superior. Accordingly the pesticide consumption in India has reached to the level of 47074, 17770 and 4862 metric tonnes for insecticides, fungicides and weedicides respectively during 1990-91.

The release of vast variety of pesticides for pest control is polluting the ecosystem with their toxic residues. The beneficial effect of pesticides in protecting the crops get diluted due to the possible health hazards caused by their toxic residues left over after use through food chain and the biomagnification of such residues over a period of time. Further, the buildup of pesticide residues in soil, received following whatever their pattern of use, contaminate the soil system affecting adversely the soil health as well as polluting the subsoil water.

The above situation warrants detailed investigations on the behaviour of pesticides for their residues in plant and soil system from the point of safety to human being as well as protection of environmental degradation due to pesticide pollution. Pesticide residues on plant system are subjected to various forces immediately after their foliar application and dissipate or metabolise over a period of time. The fate of pesticide residues in soil is subjected to soil adsorption, plant uptake, movement and degradation depending on the simultaneous influence of physical, chemical and biological processes. Further soil is an ecosystem including mineral component, organic component and microorganisms. The behaviour of pesticides in soil, therefore, depends on

the characteristics and overall functioning of this ecosystem as well as physico-chemical properties of active ingredients and of its formulated product.

Metalaxyl and mancozeb are the two important fungicides extensively used both as foliar spray as well as soil drench for the control of various plant diseases in cereals, pulses, vegetable and fruits (Singh *et al.*, 1986; Zaki *et al.*, 1981; Rawal, 1986 and Calumpang *et al.*, 1993). Mancozeb is being consumed at a substantial quantity of 1760.79 tonnes per year, while metalaxyl is increased to 1.83 tonnes (Sarathy and Rangarajan, 1992). The combination formulations of these two fungicides are becoming popular due to simultaneous control of surface infection as well as from within plant tissues following the contact action of mancozeb and rapid systemic action of metalaxyl.

The tomato crop suffers heavily due to infestation of various diseases viz., damping-off (*Phythium* spp, *Phytophthora* spp, *Rhizoctonia solani*), buck eye spot (*Phytophthora nicotianae parasitica*), late blight (*Phytophthora infestans*) and early blight (*Alternaria solani*). For effective control of most of these diseases, metalaxyl and mancozeb fungicides are used either singly or in combination depending upon the severity of the diseases on the crop (Cabras *et al.*, 1985, Narasimha Rao *et al.*, 1993, Patil *et al.*, 1993 and Sharma *et al.*, 1994).

The extensive use of the two fungicides may therefore have interactions of their residues with plant and soil. Accordingly present investigations were undertaken with following objectives:

- 1) to study the adsorption behaviour of metalaxyl and ethylenethiourea (ETU) in soils,
- 2) to study the effect of different factors on the persistence and degradation of metalaxyl, mancozeb and ETU in soils,
- 3) to study the plant uptake of metalaxyl and ETU by tomato from soil applied metalaxyl-mancozeb combined formulation,
- 4) to study the persistence and dissipation pattern of fungicide residues on tomato crop under field conditions and
- 5) to study the effect of decontamination processes on the reduction of fungicide residues from tomato fruits.

REVIEW OF LITERATURE

II. REVIEW OF LITERATURE

The economic cultivation of agricultural crops under tropical climate suffer heavy losses due to pest infestation. These losses multiply, if qualitative aspects are taken into account. Chemical control through the use of pesticides has proved efficient and economical over a period of time. Accordingly the consumption of pesticides in India has been growing rapidly in the last three decades due to the increased use of synthetic pesticides in agriculture. However, the importance of pesticides in increased agricultural production lies with their judicious use or else they can also be potential health hazards to the human being and the environment as a whole. Since, many of these pesticides are highly toxic, their continuous use is likely to contaminate soil, air, water, food stuffs and vegetation. With the environmental concern and awareness by everyone, the buildup of pesticide residues has gained considerable importance in recent years throughout the world. Whether pesticides are applied as aerial sprays, dusts to foliage or directly to the soil, large amounts of them ultimately find their way to reach soil, which finally acts as reservoir for most of the applied pesticides. Hence, it is important to know the pesticide-soil-water-plant interactions.

Metalaxyl and mancozeb are the two important synthetic organic fungicides which are being used both as soil application and as foliar spray for the control of diseases in cereals, pulses, vegetables and fruits (Zaki *et al.*, 1981 and Singh *et al.*, 1986). An attempt has been made to review the relevant and pertinent literature with respect to the adsorption, persistence and degradation of metalaxyl, mancozeb and ETU (prominent metabolite of mancozeb) in soil and their uptake and persistence in plants.

According to Bailey *et al.* (1968), the fate and behaviour of pesticides in soil system are dependent upon at least seven factors:

i) photo chemical reaction, ii) chemical decomposition, iii) microbial decomposition, iv) volatilisation, v) movement, vi) plant uptake and vii) adsorption. Among these factors, the phenomenon of adsorption-desorption appears to directly or indirectly influence the magnitude of the effects of other six factors. Adsorption appears to be

one of the major factors affecting pesticide soil colloid interaction. The process of adsorption could explain for more than 50 per cent of the loss of their bioactivity in soil (Kearney, 1970).

2.1 Adsorption of pesticides in soil

Adsorption may generally be described as the adhesion in an extremely thin layer of gas molecules, dissolved substance or liquid to the surface of the solids with which they are in contact (Boyd Ellis and Knenek, 1972). The process of adsorption involves the attraction between the electrical charges on the soil components and on the chemical and is regulated by the characteristics of soil solution (Frissel, 1961). The way in which the soil components are responsible for the adsorption depends upon the physical and chemical properties of both soil and adsorbate. A pesticide may be positively or negatively adsorbed on the soil surface.

Adsorption is due to the attraction between the solid surface and a vapour or solution. This attraction or repulsion is the resultant of interaction between the fields of force emanating from the surface of the adsorbent and the molecules of the adsorbate. Adsorption influences almost all the reactions of pesticide in soil. Firstly, as a result of this process the added toxicant loses its bioactivity, secondly adsorption restricts the movement and hence prevents the chemical from reaching its target and finally the adsorbed compound persists in soil for a longer time, thus creating problems of pollution. This phenomenon, therefore appears to be one of the major factors affecting pesticide-soil interactions.

There are four structural factors which determine the chemical character of the pesticide molecule and thus its adsorption on soil colloids. They are a) nature of functional groups such as alcohol (-OH), ketone ($>C=O$), carboxylic acid (-COOH) or amine (-NH₂), b) nature of the substituting groups, which tend to alter the behaviour of functional groups, c) position of the substituting groups, with respect to the functional groups, which may enhance or hinder intermolecular bonding and d) presence and magnitude of unsaturation in the molecule, which affects the lyophobic-lyophobic balance (Bailey *et al.*, 1968, and Bailey and White, 1970). More than one of these factors operate simultaneously.

Adsorption and interaction of pesticides with clays and soils merit consideration as the processes such as biocidal action, persistence, chemical and biodegradation, leachability and translocation depend to a great extent upon the nature of the adsorption of organic matter present in soil.

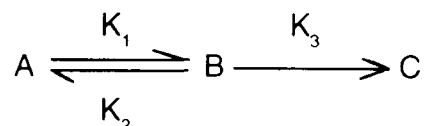
2.1.1 Adsorption of metalaxyl and ethylenethiourea in soil:

Sharom and Edgington (1982) conducted laboratory experiments to determine the adsorption of metalaxyl on four Ontario soils. The K values obtained from Freundlich's adsorption isotherms were 0.45, 0.89, 1.25 and 31.62 nanomoles per gram for Fox sandy loam, Honeywood loam, Guelph loam and Bradford muck (62.8 % organic matter) respectively. The Freundlich K values (relative measure of adsorption) for metalaxyl in different soil samples were very low and ranged from 0.20 to 0.60 nanomoles per gram as reported by Bailey and Coffey (1984). This may be due to low content of organic matter and clay in soil.

The ETU sorption was tested in three Hawaiian soils with various physical and chemical compositions. Ethylenethiourea was weakly sorbed to the three test soils and several selected clay minerals. Within three soils, ETU sorption increased with increasing organic carbon content, although other factors may be responsible for the effect observed. Relatively high sorption was also found with bentonite clay. Polar compounds such as ETU may bind to clay materials by mechanisms other than adsorption as concluded by Miles and Deorge (1991). The sorption coefficient (K_d) ranged from 0.03 to 0.12 for different soils.

2.2 Persistence of pesticides in soil

According to Kearney *et al.* (1969) persistence is defined as the time required to reduce the pesticidal concentration in soils to 75 to 100 per cent of the amount initially added. Some of these compounds persists in the soil for a longer time than others and the time required to degrade them is dependent upon the soil conditions and the nature of the compound. Once the biocide is applied to the soil, a dynamic equilibrium is established between adsorbed and solution phases and as the pesticide molecules are removed by decomposition, leaching or uptake by the plants desorption from the adsorbed phase occurs to re-establish the equilibrium. According to Furmidge and Osgerby (1967) this is explained by following equation as



where A- Soil-pesticide adsorption complex, B- pesticides in solution and C- decomposition products. The rate constant for the decomposition, K_3 is independent of soil type or its value is the same within the soil as that in a aqueous solution at the same pH and temperature in the absence of soil, if the degradation is only chemical.

Decomposition proceeds chemically by hydrolysis of the solvated pesticide molecule or from microbiologically and photochemically induced degradation. In all these processes the added pesticide initially metabolised through one of the four types of general reactions namely oxidation, reduction, hydrolysis or conjugation (Poinke and Chesters, 1973). The rates of degradation are determined by soil conditions such as pH, adsorption-desorption equilibria, moisture content, rate of application, temperature and the nature of the compound. In soils, both chemical and microbiological processes operate simultaneously.

2.2.1 Concept of half-life ($t_{1/2}$): The half-life concept has often been employed to compare the differences in the persistence between several pesticides in a given soil environment. Half-life is a measure of the time required for a pesticide to drop half of the original concentration. Although, this concept is useful in comparing the persistence of related compounds, it assumes that the rate of decomposition is logarithmic. However, in few cases this is not the situation and comparison has to be made on the basis of loss curves than on half-life.

2.2.2 Kinetics of degradation : The kinetics of pesticide degradation reaction must be considered in order to estimate the availability of a pesticide molecule for decomposition. While it is difficult to predict the order of the reaction for the decomposition of selected pesticides in a soil-water system, evidence has been given that first order kinetics is clearly a sound starting point in the consideration of the rates of breakdown. Many pesticide degradation reactions are closely approximated by this type of reaction which may be written as

$$C_t = C_o e^{-Kt}$$

where C_t - the concentration ($\mu\text{g g}^{-1}$) at time t (days), C_0 - the initial concentration ($\mu\text{g g}^{-1}$) and K - the degradation rate constant (day^{-1}). The value of K is shown to be negatively correlated and bears a definite relationship with the half-life of the pesticide in that environment (King and Mc Carty, 1968).

2.2.3 Persistence and degradation of fungicides or metabolite in soil

2.2.3.1. Metalaxyl : It is being used extensively as a seed treatment, foliar spray or soil drench in different formulation to control diseases caused by the peronosporales in many commercial crops (Zaki *et al.*, 1986).

The degradation of metalaxyl in soils has been examined by Sharom and Edgington (1982) at the rate of 4 μg per gram soil. In a loam soil containing 5.7 per cent organic matter, the half-life of metalaxyl was found to be 3 weeks. In contrast, a muck soil with 62.8 per cent organic matter, the metalaxyl half-life was about 8 weeks. This was attributed to the higher adsorptive capacity of the muck soil, due to its very high organic matter content, that may have made metalaxyl less available to its soil microorganisms.

Bailey and Coffey (1984) reported the degradation of metalaxyl (200 $\mu\text{g g}^{-1}$) in five soils and there was no evidence of biodegradation even after 10 weeks. In only one soil, there was extensive biodegradation of metalaxyl between 42 and 56 days. Metalaxyl was very stable in chemically sterilized soils with more than 90 per cent of the initial concentration remaining after 70 days. However, metalaxyl was degraded in some unsterilised soil which indicates that biological degradation was responsible for its loss. In three of five soils with a history of metalaxyl treatment, there was much more rapid degradation of fungicide compared with similar soils that had not received the chemical. However, one untreated soil with no history of prior exposure to metalaxyl was also capable of degrading it. The average half-life of metalaxyl in soils capable of degrading it was 28 days. The most active soil had a half-life of only 14 days. No difference was observed between soils treated previously with either 2.05 or 4.10 gram active ingredient per m^2 as concluded by Bailey and Coffey (1985).

Fourteen soil microorganisms were isolated, identified and tested for their relative ability to degrade ^{14}C -metalaxyl. With single microorganism, breakdown of

metalaxyl ranged from 36 to 52 per cent after 25 days and up to 75 per cent with mixtures of either fungi or bacteria. Four products of metalaxyl metabolism isolated by two dimensional thin layer chromatography (TLC) were tested for their anti-fungal activity using *Phytophthora boehmeriae* as a sensitive bioassay organism. All four break down products were inactive at concentration upto 2.5 µg per ml, contrasting with an effective concentration for 50 per cent inhibition of growth of *Phytophthora boehmeriae* for metalaxyl of 0.02 µg per ml (Bailey and Coffey, 1986).

Musumeci and Ruegg (1986) reported the degradation of metalaxyl in two soil types, sterilized or unsterilised and under flooded conditions or at 60 per cent water holding capacity. The rate of decomposition of the metalaxyl was influenced by soil type and mostly due to microbial activity. The distribution and dissipation of metalaxyl in the field was studied by Sharom and Edgington (1986). Metalaxyl was applied at 1.5 kg per hectare on 27th May 1982 and incorporated into the top 15 cm soil. The metalaxyl was not detected in the 45 to 60 cm zone and nearly 90 per cent of the applied amount was found in the top 30 cm of the soil throughout the growing season. Dissipation of the metalaxyl in the 15 to 30 cm zone was more rapid than in the 0 to 15 cm zone which may be attributed to degradation, plant uptake and leaching to below this layer. The half-life values of metalaxyl when sampled at depth of 15, 30 and 45 cm were approximately 30, 15 and 18 days respectively.

El-Yasery and Al-Adil (1991) conducted the dissipation of metalaxyl residues in soil treated with metalaxyl-mancozeb (Ridomil MZ 72 WP) at 2.5 gram per litre to field grown lettuce and green house grown cucumber plants. The residues reached a limit of 1.0 µg per gram after 20 days (fast) and 60 days (gradual) in greenhouse and field soils respectively. The metalaxyl degradation product in the soil with a molecular weight of 207 (compared with molecular weight 279 for metalaxyl) was identified in mass spectroscopy.

Zheng et al. (1989) studied the transformation of metalaxyl by the fungus *Syncephalastrum racemosum* (Cohn) Schroeter in pure culture. After 21 days of incubation in a basal medium amended with 5.0 µg of metalaxyl per ml more than 80 per cent of the compound was transformed by fungus and there was no noticeable

decrease in the amount of radioactivity from ring ^{14}C -metalaxyl in the growth medium which indicated that aromatic moiety of metalaxyl was not degraded. No mycelial growth could be found in a basal medium amended with metalaxyl and without an additional carbon source, there was also no observed transformation of metalaxyl. Apparently, *Syncephalastrum racemosum* could not use metalaxyl as the sole source for carbon, therefore it is assumed that mechanism of metalaxyl transformation by the fungus could be attributed to co-metabolism.

At metalaxyl concentration higher than 100.0 μg per ml, the fungus could not transform the fungicide, but its growth pattern was not changed as indicated by the mycelium (biomass) formation. This observation may suggest that the microbial enzyme system responsible for metalaxyl transformation was inhibited by higher metalaxyl concentration. Isolation and identification of metabolites indicated that hydroxylation on the aromatic ring or on either methyl side chain of the ring appeared to be the major transformation mechanisms of metalaxyl by *Syncephalastrum racemosum*. Hydroxylation on the methyl side chain of the ring, resulting in the formation of metalaxyl benzyl alcohol when treated with metalaxyl and the metalaxyl benzyl alcohol or its isomer occurred in the free form or conjugated with sugar or glucuronic acid. Benzylic hydroxylation of the methyl side chains and/or aromatic hydroxylation appeared to be the major reactions involved in the metabolism of metalaxyl.

The most active soil in degrading metalaxyl was a tobacco soil in which the half-life of fungicide was only 6 days, this highlights the high potential for specific soils to degrade metalaxyl. A decrease in the lag phase of metalaxyl degradation following single application of metalaxyl was demonstrated in the soil. Shortening of lag phase has been proposed as a mechanism for enhanced biodegradation for other pesticides using soil enrichment methods in which aqueous soil solutions were used. This enrichment phenomena probably reflects two different aspects of microbial behaviour, growth and adaptation. The main breakdown product of metalaxyl in all soils was the metalaxyl acid. Ring labeled ^{14}C -metalaxyl incubated for 4 weeks in 6 soils demonstrated a low rate of $^{14}\text{CO}_2$ evolution ranging from 2.1 to 11.3 per cent which was not related to the biodegradation properties of the soil. A single exposure of tobacco and corn soils to metalaxyl (100 μg ml^{-1} or 200 μg g^{-1} dry weight of soil)

significantly increased their subsequent capacity to degrade the fungicide (Droby and Coffey, 1991).

2.2.3.2 Mancozeb and ethylenethiourea (ETU): Blazquez (1973) studied the residues of mancozeb and ETU in soils when applied at the rate of 0.91 kg per 378.51 litre of water (2.0 lbs/100 gallon of water). Mancozeb (Dithane M-45) was detected from 0 ($360 \mu\text{g g}^{-1}$) to 6 ($125 \mu\text{g g}^{-1}$) days after application and no ETU was detected in soil extracts. Newsome (1975) conducted field experiment during summer 1974, to study the residues of maneb and ETU in tomato and beans. An aqueous suspension of 80 per cent active ingredient maneb formulation was applied by hand sprayer at a rate of 3.0 lbs per acre per 100 gallon. The ETU (0.35), ethylenethiuram monosulfide (ETM) (0.54) and ethylenediamine (EDA) ($0.12 \mu\text{g g}^{-1}$) residues were found in soil, 15 days after treatment with maneb.

Total ^{14}C -residues in ^{14}C -maneb treated soil (2.0 lbs/acre) under field conditions in Delaware (Keyport silt loam) had half-life of 4 to 8 weeks and the residues of ^{14}C -maneb leached much below the 5 inch soil depth (Rhodes, 1977). Nash and Beall (1980) observed half- concentration ($C_{1/2}$) times of 3, 36 and 23 days for ETU, maneb and zineb respectively in soil. Clearly little maneb/zineb or their possible degradation products moved within the soil and none was found below 1.0 cm. The samples treated with 10 mg mancozeb were completely eliminated after 3 months. At the same period 20 to 30 per cent of the active product remained in the sterile soils. The mancozeb degradation in soil is due to both chemical and biological factors. Specific bacteria (*Bacillus* spp.) were isolated from soil during *in vitro* degradation (Doneche, 1983).

Gupta and Bhattacharjee (1987) observed that mancozeb persisted longer in alkaline than in acid soils and half-life recorded for zineb in soils was 2 weeks. Calumpang *et al.* (1993) noticed the rapid loss of mancozeb in soil with an initial half-life of 2.9 days. After 15 days, mancozeb was virtually undetectable when mancozeb applied to each pipe (8.5 cm diameter x 36 cm long) at 0.00393 g corresponds to 5.5 kg a.i per hectare. The mancozeb recorded in these pipes were $3.01 \mu\text{g CS}_2$ per gram as an initial deposit and $0.14 \mu\text{g CS}_2$ per gram after 15 days application. The ETU residues in mancozeb treated plots were 1.59 and $0.004 \mu\text{g}$ per gram soil at 0 and 21 days after application.

2.2.3.3 Ethylenethiourea (ETU) : The ETU drenched (2 lbs/acre) on soil surface (Immokalee fine sand) was easily detected at $86 \mu\text{g g}^{-1}$ 13 days after application, but only a trace amount could be found after 27 days (Blazquez, 1973). Rhodes (1977) observed the total ^{14}C -analysis of soil treated with ^{14}C -ETU at 2 lbs per acre by surface application showed ^{14}C -ETU and its degradation products have an overall half-life in soil (Keyport silt loam) of less than 4 weeks under field conditions in Delaware. Only about 58 per cent of the ^{14}C -remaining in the ^{14}C -ETU treated soil was extractable after one week exposed to field conditions.

A short half-life of 3 days has been reported in soil at pH 6.7 containing 5.2 per cent organic matter under temperate conditions (Nash and Beall, 1980). Miles and Doerge (1991) studied the fate of ETU in Hawaiian soils, ETU degraded rapidly in all the three soils. Degradation experiments at three temperatures (15, 25 and 35°C) showed reaction rate approximately doubled for every 10°C increase ($E_{\text{act}} = 15.7 \text{ K Cal mol}^{-1}$) of temperature. Soils sterilised by gamma radiation and autoclaving to differentiate between biotic and abiotic degradation observed the degradation rates of 0.354 and 0.077 day^{-1} respectively when compared to control soil (0.484 day^{-1}). ETU degraded rapidly with 78 per cent reduction at 3 days after application. Degradation in soil had a half-life of 2.5 days (Calumpang *et al.*, 1993).

2.3 Uptake of metalaxyl and ETU by plants

The information on uptake and translocation of fungicides is essential for better understanding of the mode of action in controlling plant diseases. Uptake, translocation and inter and intra plant part distribution considerably affect the *in-vivo* efficacy of a systemic fungicide and are affected by plant species involved.

2.3.1 Metalaxyl : Metalaxyl has been found to be systemic inside the plants. Its translocation is mainly apoplastic but trace amount of downward translocation has also been reported. Cohen *et al.* (1979) reported that ridomil (metalaxyl 50 WP) at the rate of 175 mg per plant provided full protection of tomato leaves against *Phytophthora infestans* 1 hour after soil drench application. However, when the fungicide was applied as a foliar spray systemic translocation occurred neither upward from older leaves, downward from younger leaves, nor from sprayed leaves into fruits.

In *Persia indica*, the labelled fungicide was readily taken up by the roots and the radioactivity was translocated uniformly to the above ground parts of the plant. Metalaxyl concentrations ranging from 67 to 106 μg per gram of dry *Persia indica* root tissue were observed after the application of 1.0 mg of the fungicide as a soil drench, the amount appeared to provide a partial protection of roots of *Persia indica* against diseases. More than 50 per cent of the recovered radioactivity was found in the leaves. The portion of the recovered radioactivity found in the stem ranged from 5 to 8 per cent of the total, although the amount of the labelled fungicide that was not taken up by the plant and presumably had remained in the soil was high (89-97% of applied fungicide). These findings could suggest that for the control of root pathogens, it is more efficient to apply metalaxyl to the soil than to apply it to the foliage (Zaki et al., 1981).

The efficacy of metalaxyl, a systemic fungicide specific for peronosporales was tested by Rowe (1982) for the control of *Phytophthora infestans* on potatoes grown under field and greenhouse conditions. In greenhouse, *Phytophthora infestans* was controlled up to 90 days with soil application at the rate of 68 mg per plant (equivalent 3.4 kg/ha), whereas in field when metalaxyl was applied in furrows the diseases were controlled up to 60 days after application.

The results reported by Marucchini et al. (1983) shows that metalaxyl was rapidly absorbed and translocated by sunflower (*Helianthus annuus* L.) either when applied as a seed dressing or incorporated into the soil and active principle effectively controlled the diseases caused by *Plasmopora helianthi*. Metalaxyl reached the maximum concentration on the leaves 11 days after planting when applied as a seed dressing and 20 days after planting when applied to soil. Then, there was a progressive decrease and 34 days after planting the concentration of metalaxyl in the leaves was 70 per cent lower than the maximum concentration regardless of the treatments and its presence in the leaves was still detected 90 days after planting. The metalaxyl concentration in the leaves decreased between 3 and 5 to 0.1 and 0.2 $\mu\text{g g}^{-1}$ in the range of 11 to 90 days, the weight of the leaves however increased about 25 fold in the same period. For this reason, it believed that the decreased concentration of the fungicide in the plant organs has to be attributed to a variation of the ratio between weight of metalaxyl and weight of organs (leaves, stem, root, fruit) rather than to a degradation of the active principle.

Seed treatment of metalaxyl was recommended for the control of downy mildew of pearl millet caused by *Sclerospora graminicola* (Sau.) Schroet where primary inoculum is either soil or seed borne and infection takes place at every stage through crown or underground plant parts. Metalaxyl accumulated at the apex and the margins of leaves on root application. This is because where venation is convergent palmate parallel type, a imobiotic condition will be drawn much strongly through veins towards tip by transpirational pull (Singh et al., 1986).

Carries and Bristo (1987) noticed that the metalaxyl was readily absorbed by roots and translocated throughout young cabbage, red raspberry and strawberry plants, movement in the transpiration stream appears to account for the distribution of metalaxyl within the plants. This is indicated by the accumulation of radioactivity in older leaves and leaf margins. The increased uptake in strawberry plant parts when water was withhold from receptor and the accumulation of radioactivity at sites of injury to leaves. The concentration of systemic pesticides at sites where the epidermis has been broken is attributed to enhanced evaporation at that location. Conversely decreasing the rate at which water moves through the plant reduced uptake of metalaxyl as happened when the leaves and cotyledons of cabbage plants were wrapped in plastic and aluminium foil.

2.3.2 Ethylenethiourea : Ethylenethiourea may also be taken up by plants from the soil following the breakdown of ethylene bis dithiocarbamate fungicides (EBDC) viz., maneb, zineb, mancozeb, ziram, thiram etc. Hoagland and Frear (1976) showed that the radioactivity from ^{14}C -ETU was rapidly taken up from an aqueous solution by the roots of seedlings of corn, lettuce, pepper and tomato plants and transported to leaves. It is also metabolised and/or degraded to at least four methanol soluble components and incorporated into a methanol insoluble residue fraction in several plant species. However, complete ETU degradation to CO_2 appears to be slow and minor catabolic pathway in higher plants. The plant species tested can readily degrade ETU once it is absorbed and translocated in the plant.

The radioactivity was readily taken up and translocated throughout soybeans from ^{14}C -ETU and ^{14}C -EBDC treated soils. Leaf and stem residues from ETU surface soil treatments increased as the treatment rate increased, however the parent EBDC

added was considerably less than that for ETU subsurface treatment. Plant uptake was probably dependent upon EBDC degradation products whereas ETU could be absorbed directly. ETU content in soybeans from EBDC fungicide soil treatments remained low ($<0.2 \mu\text{g g}^{-1}$) throughout the 15 day monitoring period when applied at $10.0 \mu\text{g g}^{-1}$ soil (Nash, 1976).

Rhodes (1977) noticed that the tomato plants grown outdoors in soil treated with ^{14}C -maneb at 2.0 lbs per acre and ^{14}C -ETU at 1.1 lbs per acre surface applied or incorporated (0-4 inch), took up only trace ^{14}C labelled residues. The total ^{14}C -labelled residues in the plants based on combustion analysis but calculated as ETU ranged from only 0.1 to $0.4 \mu\text{g g}^{-1}$ after 1 week of treatment. These residues decreased at 0.01 to $0.03 \mu\text{g g}^{-1}$ at maturity (10 weeks after treatment). No detectable ^{14}C -ETU ($< 0.01 \mu\text{g g}^{-1}$) was found in any sample. Smith *et al.* (1988) carried out the uptake studies of ETU from soil treated with aqueous samples of ETU (2.0 or 10.0 mg in 10 ml), zineb (2.5 g containing 4.7 mg ETU in 50 ml) or maneb (1 g containing 0.58 mg ETU in 50 ml) were applied evenly to the soil in the pots containing the lettuces. A study carried out in which single high dose of ETU (10 mg) was applied to soil in each pot and lettuces were harvested and analysed at daily intervals. The level of ETU within the leaves rose rapidly over 4 days and then remained virtually constant for 3 to 4 days and finally becoming undetectable after 20 days. The rate of loss was not as rapid as after direct spraying with ETU. In a second experiment, repeated doses of ETU (2 mg) were applied at daily intervals to simulate the breakdown of EBDC's to ETU in the soil. In this case, the levels of ETU in the leaves rose more slowly and after 5 days was much less than following application of the same quantity of ETU as a single dose. If the daily addition of ETU (2 mg) continued, the level of ETU in the lettuces stabilised at about $0.36 \mu\text{g g}^{-1}$. At this level, the rate of loss from the leaves must balance the uptake from the soil. However, the concentration in the leaves never reached the level ($0.7 \mu\text{g g}^{-1}$) found in the first study suggesting that the level in the lettuces depend on the concentration rather than the amount of ETU available in the soils and additional ETU is not cumulative.

In another study, Smith *et al.* (1988) added zineb and maneb containing known levels of ETU to the soil pot grown lettuce. On monitoring the ETU levels in the lettuce, the levels increased rapidly during the first 3 days reaching a maximum level after 10

to 12 days. Once the maximum level had been reached, the decline was much slower than found for ETU alone and did not fall to an undetectable level until 20 to 35 days after the initial application. It therefore, appears that additional ETU was being formed in the soil by the breakdown of fungicides and the same was being absorbed by the plants. The rate of ETU formation was not however sufficient to produce a steady level, presumably as the fungicides themselves declined.

2.4 Persistence of pesticide residues in crops

Pesticides have become indispensable to agriculture and some of these chemicals persist in food for extended periods of time and pose a threat to the health of the consumers. Accordingly the persistence pattern of all the pesticides are being studied from the safety point of view.

2.4.1 Metalaxyl : The residual behaviour of acylanilidic group of fungicides was studied in greenhouse grown tomatoes by Cabras *et al.* (1985) at the rate of 380 and 760 g per hectare in Italy. The disappearance of residues from acylanilide (metalaxyl, furalaxyl and benalaxyl) did not seem to be linear or a pseudo-first order kinetics. The data showed that furalaxyl, metalaxyl and benalaxyl had different persistence pattern in tomatoes even if their pre-harvest times was 21 days. The values of furalaxyl residues 13 days after application were very low ($<0.04 \mu\text{g g}^{-1}$) while those of metalaxyl and benalaxyl were detectable up to 20 days (0.16 to $0.21 \mu\text{g g}^{-1}$) after the last spray.

El-Yasery and Al-Adil (1991) noticed the rapid dissipation of metalaxyl residues in lettuce and cucumber and accordingly the accepted residue limit ($1.0 \mu\text{g g}^{-1}$) was recorded only after 17 and 29 days respectively. The dissipation of metalaxyl-mancozeb in/on grape berries sprayed at the concentration of 0.2 and 0.4 per cent a.i. corresponding to 720 and 1440 gram a.i. per hectare was studied by Narasimha Rao *et al.* (1993). Metalaxyl present in metalaxyl-mancozeb after third application at 0.2 and 0.4 per cent recorded initial deposits of 9.7 and $12.0 \mu\text{g}$ per gram which dissipated to below the tolerance limit of $2.0 \mu\text{g}$ per gram in 6.6 and 8.6 days and the half-life values were 2.7 and 3.3 days.

2.4.2 Mancozeb and ethylenethiourea : Foliar application of Dithane-M 45 at 2.0 lbs per acre to tomato under field conditions effected in slower rate of the residue

7A-5948



decay resulting in high fungicidal activity (Blazquez, 1973). Residues of Maneb and ETU were measured by Newsome *et al.* (1975) on crops of beans and tomatoes at 0 to 14 days after treatment with maneb (3.0 lbs/acre). The level of residues declined with time, beans were found containing more of each compound than tomatoes. After 14 days beans contained 13.0 and 0.11 $\mu\text{g g}^{-1}$ maneb and ETU, while tomato 10.0 and 0.07 $\mu\text{g g}^{-1}$ maneb and ETU respectively.

Newsome (1976) sprayed four EBDC fungicide formulation *viz.*, manzate D, 3.0 lbs (2.4 lbs a.i./acre), dithane -M 45, 3.0 lbs (2.4 lbs a.i./acre), polyram 2.0 lbs (1.6 lbs a.i./acre) and zineb 3.25 lbs (2.4 lbs a.i./acre) to tomatoes in field and the residues left by the manganese containing fungicides like mancozeb and manzate were significantly higher than polyram or zineb. With all the compounds, 24 to 37 per cent of the initial residues remained for 14 days after application. The highest level of these residues are associated with those treatments which resulted in the highest levels of parent EBDC. The presence of ETU on soybean leaves was also noticed by Nash (1976) after EBDC foliar spray (5 ml of 2500 $\mu\text{g g}^{-1}$). Aqueous solution of ^{14}C -ETU or ^{14}C -EBDC to 4 plants per pot treatments, decreased rapidly from 20 $\mu\text{g g}^{-1}$ after 1 day to less than 2 $\mu\text{g g}^{-1}$ after 15 days in the greenhouse.

Ripley and Simpson (1977) gave a spraying of zineb (5.04 kg a.i ha $^{-1}$) to Bartlett pears and immediately after spraying zineb was determined at about 350 and 5.2 $\mu\text{g g}^{-1}$ in foliage and pear fruits respectively. In foliage, zineb residues declined rapidly to about 100 $\mu\text{g g}^{-1}$ 12 to 15 days after second application and dissipated slowly thereafter. In the first 9 days zineb residues declined at about 27 $\mu\text{g g}^{-1} \text{ day}^{-1}$ and then leveled off to a rate of decline of about 4 $\mu\text{g g}^{-1} \text{ day}^{-1}$. While in fruits, the concentration of zineb declined to about 1.0 $\mu\text{g g}^{-1}$ within 18 to 21 days with the rate of dissipation being 0.7 $\mu\text{g g}^{-1} \text{ day}^{-1}$ for the first 7 days and 0.1 $\mu\text{g g}^{-1}$ over the rest of the trial period. The ETU was found at level of 0.02 $\mu\text{g g}^{-1}$ after first 3 to 7 days and declined thereafter to 0.01 $\mu\text{g g}^{-1}$ at harvest.

Analysis of tomatoes, potatoes, cucumbers, summer squash and cantaloupes taken from 17 different locations throughout the United States, where maneb was applied according to the label directions (2.24 to 3.36 kg ha $^{-1}$) showed no residual ETU (< 0.05 $\mu\text{g g}^{-1}$) on the raw agricultural commodities, even in the presence of up to 4.0 $\mu\text{g g}^{-1}$ of maneb (Pease and Holt, 1977). Rhodes (1977) observed the rapid rate of

degradation of ^{14}C -maneb or ^{14}C -ETU in tomato and beans and evidence that ETU is accumulating as the total ^{14}C -labelled residues on maneb treated crops disappears. The small amounts of ETU reported are attributed to problems with the analytical method for isolating ETU when used in the presence of maneb residue.

Residues of EBDC and ETU were monitored by Ripley and Cox (1978) in tomatoes after application of several EBDC formulations and after spraying at recommended rates, residues of EBDC on tomatoes were below the Canadian tolerance of $4.0 \mu\text{g g}^{-1}$ at recommended harvest interval. ETU was detected during the analysis period at levels of less than $0.05 \mu\text{g g}^{-1}$ on tomatoes. The mean values of $6.8 \mu\text{g g}^{-1}$ zineb, $0.38 \mu\text{g g}^{-1}$ dinocap and $0.03 \mu\text{g g}^{-1}$ ETU were found in grapes immediately after application of dicar and these residues dissipated by 50 per cent in the first 15 to 20 days, therefore slight dilution due to growth was observed (Ripley *et al.*, 1978). Throughout the sampling period, in two years, the ratio of dinocap : ETU : zineb concentration remained fairly constant at about 4.5 : 0.04 : 100 respectively.

Ross *et al.* (1978) noticed the residues of 1.70 and $0.5 \mu\text{g g}^{-1}$ mancozeb and metiram respectively in apple sprayed 9 times with EBDC fungicides after 42 days of application. The initial EBDC deposit of 12.8 and $12.5 \mu\text{g g}^{-1}$ for mancozeb and metiram were resulted from foliar sprays at 907 and 680 gram of dithane-M 45 (80 WP) and polyram (80 WP) per hectare. The ETU residues were $0.01 \mu\text{g g}^{-1}$ for both the plot with 3 cover sprays. The residue of the EBDC fungicides - maneb and mancozeb on tomato fruits ranged between 0.03 and $0.80 \mu\text{g g}^{-1}$ (mean $0.31 \mu\text{g g}^{-1}$) when field crops were sprayed (3.36 kg ha^{-1}) five or eight times at intervals of 3 to 4 days. On greenhouse tomatoes two or four sprays with mancozeb resulted in residues of up to 2.81 and $0.08 \mu\text{g g}^{-1}$ EBDC and ETU on the fruit (Von stryke and Jarvis, 1978).

Experiments were conducted (Newsome, 1978) in tomatoes to study the residues of mancozeb treated at rates of 1.5 and 3.0 lbs of mancozeb formulation (80 WP) per acre. The residues of mancozeb at one-half the maximum recommended rate of 3.0 lbs per acre ($4.8 - 0.53 \mu\text{g g}^{-1}$) resulted in approximately half the residue ($2.4 - 0.37 \mu\text{g g}^{-1}$) for 0 and 28 days after application. Nash and Beall (1980) reported that the maneb and zineb were present on the whole fruit at less than $1.0 \mu\text{g}$ per gram and both had half concentration times ($C_{1/2}$) of 14 days on leaves. Further it was also concluded that the dissipation rate had followed the first order equation

$\ln(C_t) = \ln(C_0) - Kt$, where C_0 - concentration $\mu\text{g g}^{-1}$ at t_0 , and K - dissipation rate (day^{-1}). The $C_{1/2}$ values of the EBDC fungicides and ETU in fruits and vegetable ranged from 3 to 35 days (7-11 days average) and 3 to 28 days (mean of 6 days).

The amount of ETU residues on leaves decreased rapidly to negligible levels within a week, unless EBDC fungicides had been applied to the soil as the latter were degraded simultaneously to give additional ETU which was then taken up by the plants (Smith *et al.*, 1988).

Udaykumar and Agarwall (1990) reported the half-life of 15 and 10 days for mancozeb and maneb respectively on brinjal plants. Both these compounds were most persistent on leaves and least on fruits. Besides the parent fungicides, three compounds namely ethylenethiuram disulfide (ETD), ethylenethiuram monosulfide (ETM) and ethylenethiourea (ETU) were detected in the brinjal fruits as metabolites or degradation products. The amount of ETU was $0.032 \mu\text{g g}^{-1}$ on zero day which increased to $0.51 \mu\text{g g}^{-1}$ 20 days after treatment following the metabolism, however a gradual decline was observed thereafter. This suggests the formation of ETU from EBDC fungicides and then its breakdown to certain other compounds.

Narasimha Rao *et al.* (1993) noticed the half-life of 3.1 to 3.3 days for mancozeb in grapes fruits and the time required to dissipate the chemical to below tolerance limit (3.0 mg/kg) was 5.3 and 7.7 days from four field sprays with 0.2 and 0.4 per cent a.i. metalaxyl-mancozeb (Ridomil-MZ 72 WP) per hectare. In a similar type of study conducted by Patil *et al.* (1993), the application of first three sprays of mancozeb (0.25 % a.i.) did not show any detectable levels of residues in fruits harvested 45 days after first spray. However, residue level on fruits harvested after fourth spray was little more ($3.02 - 3.27 \mu\text{g g}^{-1}$) than maximum residue limit (MRL) of $3.0 \mu\text{g g}^{-1}$. From the consumer safety point of view, a waiting period of one day could therefore be advocated if tomato crop was sprayed in fruit bearing stage with 0.25 per cent mancozeb.

Persistence of mancozeb (1.9 and $3.8 \text{ kg a.i ha}^{-1}$) was studied by Sharma *et al.* (1994) under field conditions in cabbage, knol-khol, tomato, okra and brinjal. The residues were detectable up to 30 days in cabbage, knol-khol, 10 days in tomato and brinjal and 7 days in okra. The waiting periods of 27, 22, 4, 3 and 1 day(s) were

worked out for cabbage, knol-khol, brinjal, okra and tomato respectively. The half-life values varied from 1.2 to 8.5 days for these vegetables reflecting the fast rate of dissipation of residues.

2.5 Decontamination of pesticide residues from tomato

The buildup of pesticide residues on vegetables and fruits following repeated applications at times persist in bioconcentration till harvest and therefore is potential health hazard. It is therefore desirable to decontaminate such of the quantities of pesticide residues before consumption. Tomato is one of such a vegetable that is consumed afresh, cooked and processed and therefore needs to ensure dislodging the residues, even when it is consumed afresh. While there is the possibility of deposits of pesticide residues on fruit pericarp, the systemics do move inside also. Various physical and chemical processes have been recommended for dislodging the residues from fruits and vegetables after harvest (Awasthi, 1986, 1993). Simple washing of fruits in water often dislodge deposits on the fruit surface, washing with chemical solutions tend to remove adsorbed residues while, cooking resort to reduce heat labile residues by chemical breakdown to nontoxic degradation products.

2.5.1 Washing : Marshal and Jarvis (1979) found that tomato juice from tomato washed in running water for 10 minutes contained 52 per cent less EBDC and 50 per cent less ETU that made from unwashed tomatoes. Trotter and Pardue (1982) agree about the effectiveness of washing on reducing EBDC residues from spinach, but found no indication that ETU residues are similarly affected, perhaps as a result of their being incorporated into spinach. The washing time (1 or 3 min.) did not seem to have any effect on the levels of ETU in the canned spinach (Gonzalez *et al.*, 1989). Sharma *et al.* (1994) suggested simple processing in freshly harvested vegetables was effective in decreasing the amount of mancozeb as a result of washing.

2.5.2 Cooking : Most of the studies indicated that the EBDC sprayed fungicides of crop samples (edible parts) upon cooking or boiling for 10 to 15 minutes reduced the active principle of EBDC fungicides [Ross *et al.* (1978) for apple, Ripley and Cox (1978) and Sharma *et al.* (1994) for tomatoes] whereas, the degradation product ETU levels raised [Newsome (1976) and Ripley and Cox (1978) for tomatoes, Ripley and Simpson (1979) for pears, Ripley *et al.* (1978) for grapes and Hajslova *et al.* (1986) for apples].

MATERIAL AND METHODS

III. MATERIAL AND METHODS

The present investigations were undertaken to study the kinetics and behaviour of applied fungicides in soils and tomato (*Lycopersicon esculentum*, Mill.) plant system. The details of the procedures used in the experiments are described below.

3.1 Experimental soils

For soil-fungicide interaction studies, three soils representing the major soil types of Karnataka were selected viz., sandy loam from Indian Institute of Horticultural Research (IIHR), Hessarghatta, Bangalore; sandy clay loam (organic matter rich) from Central Horticultural Experiment Station (CHES), Chettalli, Coorg district and clay soil from Agricultural Research Station (ARS), Hiriur, Chitradurga district. Surface soil samples from 0 to 15 cm depth were collected from spots which had no history of metalaxyl and mancozeb application. The samples were air dried, hand pounded and passed through 2 mm sieve. To bring uniformity and destroy unwanted weed seeds present in the soil, it was moistened to field capacity for a period of 8 to 10 days in plastic bucket. After incubation, the soil was air dried, powdered, passed through 2 mm sieve and stored in polythene bags. The physical and chemical properties of these soils (Table 2) were estimated as per the standard procedures given in Table 1.

3.2 Fungicides used

Analytical and formulation grade fungicides used in the present investigation were obtained from the manufactures or subsidiaries (Metalaxyl : Hindustan CIBA-GEIGY, Bombay, Mancozeb : Indofil Chemicals, Bombay and ETU : courtesy of Dr. S.K. Handa, Project Coordinator, AICRP (Pesticide residues) IARI, New Delhi). The fungicides were metalaxyl [*Methyl N-(2-methoxyacetyl)-N-(2,6-xylyl)-DL-alaninate*], mancozeb [{*1, 2-ethanediyl bis (carbamadithioate) (2-) manganese*} mixture with {*1,2-ethanediyl bis (carbamadithioate) (2-) zinc*}], Ethylenethiourea [*Imidazolidine 2-thione*], the major degradation product (metabolite) of mancozeb was also studied. The general properties (Windholz, 1983 and Worthing and Walker, 1983) of these compounds are illustrated in Table 3.

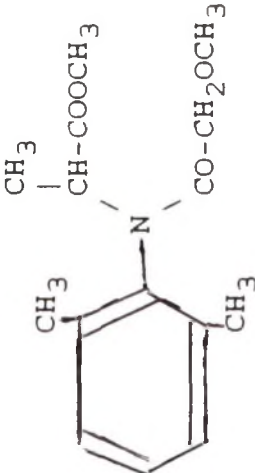
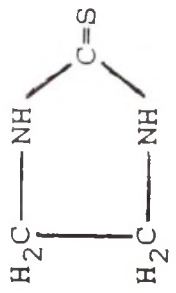
Table 1. Analytical methods followed in soil analysis.

Sl. No.	Determination	Methodology	Reference
1)	Mechanical analysis	Hydrometric method	Piper (1966)
2)	pH (Soil:water::1:2.5)	Potentiometric method	Jackson (1973)
3)	Electrical conductivity	Conductometric bridge	Jackson (1973)
4)	Bulk density	Keen's Raczkowski	Piper (1966)
5)	Field capacity	Keen's Raczkowski	Piper (1966)
6)	Organic carbon	Walkley and Black wet oxidation	Jackson (1973)
7)	Cation exchange capacity Non calcareous soil Calcareous	Leaching with 1 N ammonium acetate Leaching with 1 N sodium acetate	Black (1965)

Table 2. Physico-chemical properties of soils

Parameters	Bangalore	Chettalli	Hiriyur
1) Particle size distribution			
Sand (%)	49.10	65.15	41.45
Silt (%)	27.40	8.25	12.25
Clay (%)	23.50	26.50	46.30
Textural class	Sandy clay loam	Sandy clay	Clay
2) Taxonomical classification	Kandic haplustalfs	Ustic palehumults	Chromic haplusterts
3) pH (1:2.5)	7.09	6.24	8.76
4) Electrical conductivity (dS m ⁻¹)	0.19	0.20	0.21
5) Organic carbon (%)	0.50	1.80	0.32
6) Moisture content (%) at field capacity	28.99	38.75	42.02
7) Bulk density (g cm ⁻³)	1.38	1.28	1.20
8) Cation exchange capacity (cmol (p ⁺) kg ⁻¹)	13.88	17.63	41.38

Table 3. General properties of fungicides and metabolite under study.

Properties		Metalaxyl	Mancozeb	Ethylenethiourea
1) Common name		Metalaxyl	Mancozeb	Ethylenethiourea
2) Chemical name		Methyl N-(2-methoxyacetyl)-N-(,6-xylyl)-DL-alaninate	Polymeric manganese zinc (ethylene bis) dithiocarbamate	Imidazolidine 2-thione
3) Structural formula				
4) Trade name		Ridomil, Apron, Fubol	Dithane M-45, Manzate	--
5) Molecular weight		279.3	266.5	102.2
6) Melting point (°C)		71.8 - 72.0	192 - 204	203 - 204
7) Vapour pressure (mm Hg) at 20°C		5.62 x 10 ⁻⁶	Negligible	Not Available
8) Solubility (g/lit) in water		7.2 at 20°C	Negligible	20 at 30°C

* = [1,2 - ethanediyl bis (carbamadithioate) (2-) manganese mixture with [1,2 - ethanediyl bis (carbamadithioate) (2-) zinc

Ridomil MZ 72 WP= (8 % metalaxyl + 64 % mancozeb)

3.3 Experiment details

Following laboratory, glass house and field experiments constituted the present investigations:

- 3.3.1 Adsorption of metalaxyl and ethylenethiourea (ETU) in soils.
- 3.3.2 Degradation of metalaxyl, mancozeb and ETU in soils
- 3.3.3 Plant uptake of metalaxyl and ETU
- 3.3.4 Persistence of metalaxyl, mancozeb and ETU in/on tomato and in soil.
- 3.3.5 Decontamination of fungicide residues from tomato fruits.

3.3.1 Adsorption of metalaxyl and ethylenethiourea (ETU) in soils

The experimental methods for studies on soil adsorption for the various pesticides were essentially the same as described by Talbert and Fletchall (1965) and Adams and Paulina (1971) with some modifications. The adsorption of metalaxyl and ethylenethiourea to three soils at various shaking periods up to 12 hours indicated that shaking of soil with metalaxyl or ethylenethiourea for a period of 2 hours was found to be sufficient to attain equilibrium. As a consequence, the following procedure was adapted for studying their adsorption in soils.

Soil samples of 4 g of 2 mm sieved soils were shaken with 20 ml of aqueous solutions containing 0, 1.0, 2.5, 5.0, 10.0, 15.0 and 20.0 µg per ml metalaxyl or ethylenethiourea in 0.01M CaCl₂ in 40 ml screw capped glass centrifuge tubes. These concentrations were equivalent to 0, 5.0, 12.5, 25.0, 50.0, 75.0 and 100.0 µg of metalaxyl or ethylenethiourea per gram of soil respectively. The contents were equilibrated for 2 hours on a wrist action mechanical shaker. At the end of equilibration, the contents were centrifuged at 1500 rpm for 5 minutes. After centrifugation, a known quantity (10 ml) of the supernatant was quantitatively analysed for metalaxyl or ethylenethiourea in the solution. The quantity of metalaxyl or ethylenethiourea adsorbed was calculated by determining the difference between the initial concentration and concentration present in the equilibrium solution.

For each measurement, a fortified blank was included to measure the effect of adsorption of fungicide or metabolite to the wall of the centrifuge tube. The amount of fungicide adsorbed on soil was calculated by equation

$$A = (C_0 - C)V/M$$

where 'A' is the amount adsorbed (µg g⁻¹ of soil) C₀ and C are equilibrium

concentration ($\mu\text{g ml}^{-1}$) of fungicide after shaking without and with soil, V is the volume of solution (ml) and M is the weight of soil (g). Three replications were carried out for each measurement.

3.3.1.1 Adsorption isotherms : The adsorption isotherms were constructed using the best fit regression equation from the data on the quantity adsorbed and corresponding equilibrium concentration. They were fitted as per Freundlich adsorption equation, since this could be applied to such heterogeneous systems like soil-pesticide-water suspensions as

$$x/m = K C_e^{1/n} \dots\dots\dots (1)$$

where x/m - quantity of pesticide adsorbed per unit weight of soil ($\mu\text{g g}^{-1}$), C_e - equilibrium concentration ($\mu\text{g ml}^{-1}$), K and n are empirical constants. The Freundlich isotherm predicts that adsorption increases without limit (exponentially) as the bulk concentration increases. However, when this is expressed in logarithmic form it takes linear relationship which represented as under

$$\log (x/m) = \log (K) + 1/n \log (C_e) \dots\dots\dots (2).$$

The constants were obtained from the appropriate plots where intercept and slope of straight line represented K and $1/n$ respectively.

The adsorption varies with a number of soil properties, the soil organic carbon content being apparently the single best factor influencing the sorption coefficient. The sorption coefficient for a particular pesticide when normalised with respect to soil organic carbon (OC) content is essentially independent of the soil type and hence the normalised sorption coefficient (K_{oc}) is calculated as

$$K_{oc} = [(K) / (\% \text{ OC})] \times 100 \dots\dots\dots (4)$$

where K- Freundlich constant and % OC- per cent soil organic carbon.

To know the change in the Gibb's free energy for adsorption after knowing K and n from logarithmic plots, the following equation was used as followed by Osgerby (1970), Felsot & Dahm (1979) and Kumari & Singh (1992 and 1994).

$$dG^\circ = -R T \ln (K) \dots\dots\dots (3)$$

where dG° - Gibb's free energy change (K Cal mol^{-1}), R- Gas constant ($1.986 \text{ Cal mol}^{-1} \text{ K}^{-1}$), T- absolute temperature (Kelvin).

3.3.2 Degradation of metalaxyl, mancozeb and ETU in soils

Investigations on the influence of rate of application, moisture, temperature and autoclaving of soil on the degradation of metalaxyl, mancozeb and ethylenethiourea were studied in three soils. The following treatments were adopted for the degradation studies.

Treatment	Moisture	Temperature	Soil condition
T ₁ a or T ₁ b	Field capacity	25°C	Normal
T ₂	Air dry	25°C	Normal
T ₃	Field capacity	15°C	Normal
T ₄	Field capacity	25°C	Sterile

3.3.2.1 Persistence of metalaxyl and mancozeb in soils

The degradation of metalaxyl and mancozeb (to resemble Ridomil-MZ) was studied in three soil. Twenty five grams of each soil samples were weighed in to 150 ml conical flasks. After weighing one set of soils along with conical flask were sterilised in autoclave at 20 PSI for 15 minutes. The metalaxyl and mancozeb were added to each flask at 4.0 and 20.0 $\mu\text{g g}^{-1}$ respectively for all the treatments except in T₁a treatment which received 2.0 and 10.0 μg metalaxyl and mancozeb per gram soil in the form of solution. After mixing the soil with fungicides, the moisture and temperature were adjusted and incubated as mentioned in the treatment details. Each treatment was replicated three times. Periodically each flask was checked for the moisture contents and maintained by addition of required quantities of water. At regular intervals (0, 5, 10, 20, 30, 45, 60, 80 days after treatment), the conical flasks were drawn from each treatment and entire quantity of the soil contained in the flask (25 g) was used for estimation of metalaxyl and mancozeb remained and ethylenethiourea formed (in T₁ treatments) in the soil.

3.3.2.2 Persistence of ethylenethiourea in soils

The persistence of ethylenethiourea was also studied in three soils under the influence of rate of application, moisture, temperature and autoclaving of soil as explained in the section 3.3.2.1. The levels of ethylenethiourea used were 2.0 $\mu\text{g g}^{-1}$ for T₁a and 4.0 $\mu\text{g g}^{-1}$ for other treatments.

3.3.3 Plant uptake of metalaxyl and ETU

A pot culture experiment was conducted using IIHR, Hessarghatta, Bangalore soil to study the uptake and persistence of metalaxyl, mancozeb and ethylenethiourea in tomato and persistence of their residues in soil. The preliminary investigations revealed that application of metalaxyl-mancozeb (Ridomil-MZ 72 WP) at the rate of 1250 mg per 5 kg soil ($20 \mu\text{g g}^{-1}$ metalaxyl + $160 \mu\text{g g}^{-1}$ mancozeb) developed the toxicity symptoms in tomato plants after 10 days of application. Hence a lower dose of Metalaxyl-Mancozeb was selected for the study viz., 156.25 and 312.50 mg of formulation per 5 kg (pot), this corresponded to 2.5 and 5.0 $\mu\text{g metalaxyl g}^{-1}$ and 20.0 and 40.0 $\mu\text{g mancozeb g}^{-1}$ soil respectively.

Five kilogram soil was filled into earthen pots and then moistened to field capacity. One month old five tomato seedlings were transplanted into each pot and allowed 15 days for establishment. Afterwards, only three plants were allowed in each pot and metalaxyl-mancozeb (Ridomil-MZ 72 WP) was applied at rates of 156.25 and 312.50 mg per pot. The pots were moistened everyday. At regular intervals (0, 2, 5, 10, 15, 20, 25, 30 and 40 days after treatment), three pots were drawn from each treatment and plants were pulled out, washed with water. The adhered water was removed from the plant surface. The residues of fungicides were analysed in plant parts like foliage and fruits as well as in the soil. The metalaxyl and ethylenethiourea in the plants and fruits will correspond to uptake in tomato from soil applied fungicides. The persistence of metalaxyl, mancozeb and ethylenethiourea in soil was also monitored at different intervals.

3.3.4 Persistence of metalaxyl, mancozeb and ETU residues in/on tomato and in soil

A field experiment was conducted involving the tomato crop during summer 1995 (January to April) at IIHR, Hessarghatta, Bangalore to study the persistence of residues of metalaxyl, mancozeb and ethylenethiourea on/in tomato plants and in soil after the foliar spray application of metalaxyl-mancozeb (Ridomil-MZ 72 WP) at the effective rate of application (0.2 %) and also higher dose (0.4 % spray). The weather parameter of the experimental site are presented in Table 4.

Table 4. Weather parameter of the experimental site

Date/Month 1995	Evaporation (mm)	Temperature (°C)		Relative Humidity (%)
		Maximum	Minimum	
01-07th March	6.0 - 8.0	32 - 33	18 - 19	28 - 45
08-14th March	5.0 - 8.0	30 - 32	17 - 20	39 - 69
15-21th March	7.5 - 8.0	32 - 33	19 - 19	35 - 45
22-28th March	4.4 - 9.5	31 - 35	17 - 20	24 - 63
29-04th April	6.0 - 6.5	32 - 34	19 - 21	36 - 61
05-10th April	5.0 - 6.5	33 - 34	19 - 21	51 - 64

Note: No rainfall during the experiment, except 26-03-1995
(50.5 mm rainfall).

Field preparations were done according to agronomic requirements of tomato cultivation. Field experiment was laid out in different plots by constructing suitable bunds. A nursery of tomato CV Arka Saurab (S-IV) was raised and 25 days old seedlings were transplanted in test plot size of 4 m x 3 m consisting of 4 rows and 10 plants each row at a spacing of 45 cm. Furrows were opened 75 cm apart. A basal dose of 60, 100 and 60 kg N, P_2O_5 and K_2O per hectare was applied by placement. Irrigation was given before and after transplanting and as and when required. The crop was top dressed with 50 kg N per hectare after one month of transplanting and attended all recommended cultural practices. The fungicide Ridomil-MZ 72 WP (8 % metalaxyl and 64 % mancozeb) was applied in two sprayings, one at flowering stage (10-02-1995) and the second at 50 per cent fruiting (01-03-1995). An aqueous suspension of Ridomil-MZ 72 WP was applied by knapsack sprayer at the rate of 0.2 and 0.4 (30 and 60 g/15 lit. water) per cent sprayed to runoff stage at an interval of 20 days. Each treatment was replicated in 6 plots.

The sampling was commenced on the day of second spray and thereafter. From each plot nearly 250 gram samples of tomato foliage, fruits and soil were collected and pooled treatment wise. The plant and soil samples were immediately taken to the laboratory and processed for residue analysis. In each treatment, the samples of different plots were mixed, chopped into small pieces and sub samples (50 g or depending on residue concentration) were taken for residue analysis of metalaxyl, mancozeb and ethylenethiourea in foliage and fruits. The soil was air dried at room temperature, mixed and a representative sample of 25 g was taken for fungicide or metabolite residues analysis. In each treatment samples were replicated three times.

3.3.4.3 Decontamination of fungicide residues from tomato fruits :

Studies were carried out using 5 and 10 day old fungicide treated (sprayed) tomato fruits. The treatments involved washing alone and washing following cooking. The fruits were washed in running water for 2 minutes. In case of combination of washing and cooking, the washed fruits (100 g) were cooked in pressure cooker for 15 min. and processed for residue analysis.

3.4 Methods of residue analysis in tomato and soil

3.4.1 Metalaxyl residues : The method of residue determination of metalaxyl (Fig. 1) followed in the present investigation was essentially same as described by Ramsteiner (1976) with certain modifications.

3.4.1.1 Extraction : The representative chopped samples of tomato (foliage or fruits) were blended in Waring blender for 2 minutes at high speed. The sub samples of 50 gram was weighed into 250 ml conical flask and 75 ml of methanol was added and the contents were shaken for one hour on a mechanical shaker. The contents were filtered in a buckner funnel through Whatman No.1 filter paper using celite 545 as filtering aid. The latter was fitted to a filtering flask attached to an aspirator. Before filtration, the celite layer was washed with solvent (methanol) with light suction. The residue on the funnel was rinsed twice with 25 ml methanol each time. The filtrate was transferred to a 500 ml joint concentration flask.

3.4.1.2 Partitioning : The volume of the filtrate obtained after extraction and filtration was concentrated to approximately 10 ml by rotary vacuum evaporator at 40°C. The concentrate was transferred to a 250 ml separatory funnel and diluted with 20 ml distilled water. The aqueous mixture was partitioned with dichloromethane (3 x 40 ml) by vigorously shaking the separatory funnel for 5 minutes during each extraction. The dichloromethane layer was collected into 250 ml flask through funnel overlaid with anhydrous sodium sulphate on cotton plug and concentrated the extract to dryness using a rotary vacuum evaporator at 40°C.

3.4.1.3 Column chromatographic cleanup : Chromatographic glass columns (10 mm i.d x 300 mm length) were prepared for cleanup with a small plug of glass wool and 2.5 g anhydrous sodium sulphate overlaid by 20 g of activated acidic alumina (activity grade V). The columns were prewashed first with 50 ml n-hexane. The metalaxyl residues were then dissolved in n-hexane (2 x 10 ml) and transferred quantitatively to the column after repeated rinsing. The column was washed with 75 ml n-hexane and followed by 30 ml mixture of n-hexane and diethyl ether (1:1) at the rate of 2 to 3 ml per minute. Finally, the column was eluted with 100 ml mixture of n-hexane and diethyl ether (1:1) and elute was collected in 250 ml conical flask. The elute was concentrated to 40 ml. Activated charcoal (0.5 g) was added to

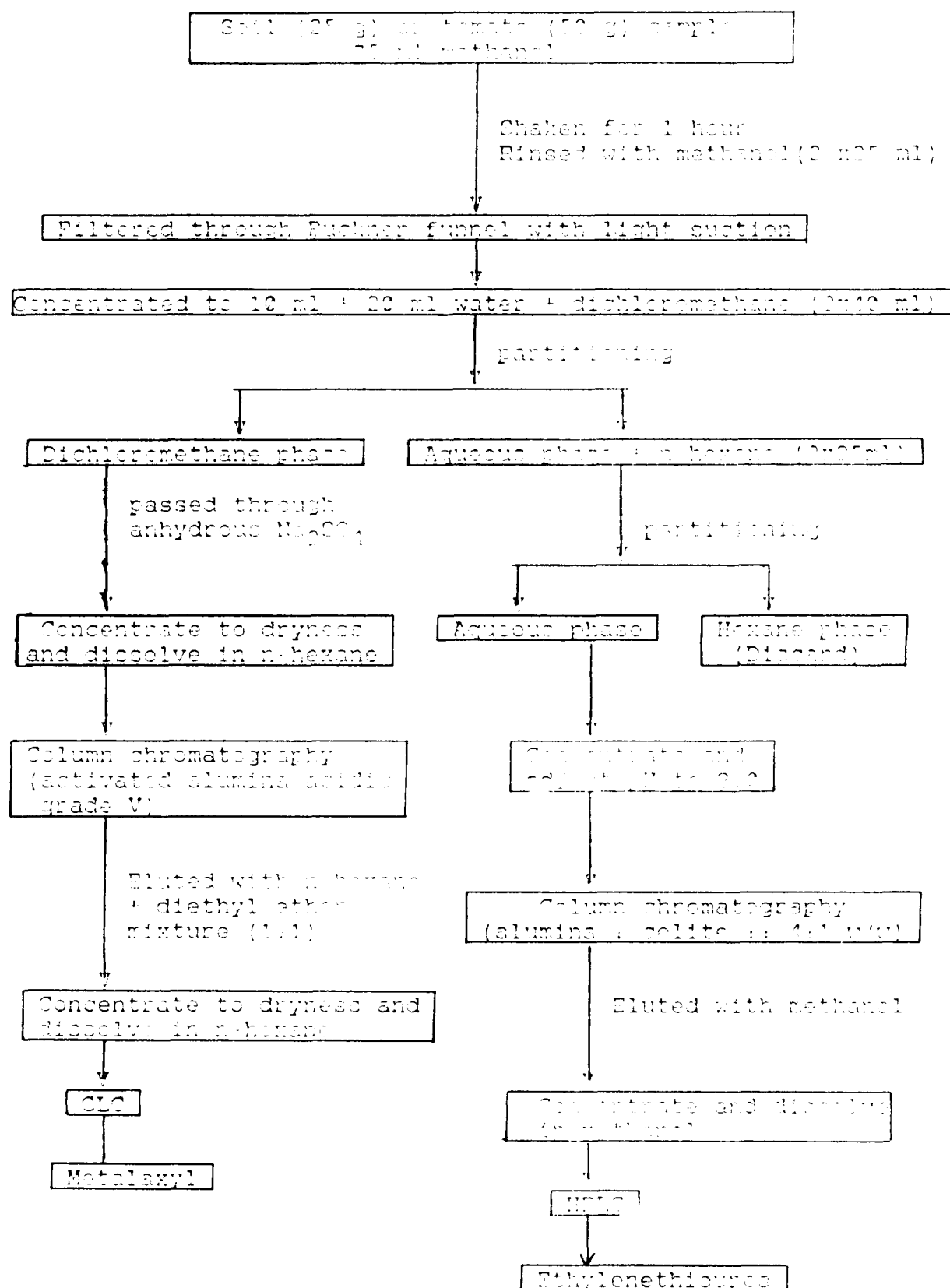


Figure 1. Flow chart showing the residue analysis of metalaxyl and ETU from tomato

the extract. The contents were shaken intermittently for 30 minutes, filtered through Whatman No.1 filter paper in 250 ml flask rinsing the flask and filter paper two times with n-hexane (2 x 10 ml). The filtrate was evaporated to dryness on a rotary vacuum evaporator at 40°C. The metalaxyl residues were dissolved in 5 ml of n-hexane and transferred to graduated test tube and made up to known volume by repeated washings. A suitable aliquot (1 µl) of the sample was injected into a Varian 3600 Gas Chromatograph (GC) with the help of microlitre syringe for quantitative analysis.

3.4.1.4 Gas Chromatography : The standardised Gas Chromatography parameters for metalaxyl were as follows :

Column	: 10 % OV-101 Chromosorb (80/100 mesh) 2m length x 1/8"
	inner diameter stainless steel.
Detector	: Thermoionic specific detector
Temperature	: Column oven : 225°C
	Injection Port : 230°C
	Detector : 280°C
Carrier gas and flow rate	: Nitrogen, 30 ml min ⁻¹
Air and flow rate	: Zero air, 180 ml min ⁻¹
Fuel gas and flow rate	: Hydrogen, 4.8 ml min ⁻¹
Bead current	: 3.075 amps
Voltage bias	: -4.5
Retention time	: 3.2 minute
Limit of detection	: 0.05 µg g ⁻¹

3.4.1.5 Calculation of the residue : Metalaxyl was separated on GLC and identified at the retention time of 3.2 minutes. The peak area of the chromatogram was measured for quantitative analysis of metalaxyl residues with respect to standard chromatogram (Fig. 2).

$$\text{Metalaxyl residue} = \frac{A_1 \times V_2 \times V_3 \times C \times f}{A_2 \times V_1 \times m}$$

(µg g⁻¹)

where A₁- Peak area of the sample (µV sec⁻¹), V₂- Volume of standard metalaxyl solution injected (µl), V₃- total volume of sample solution (ml), C- Concentration of

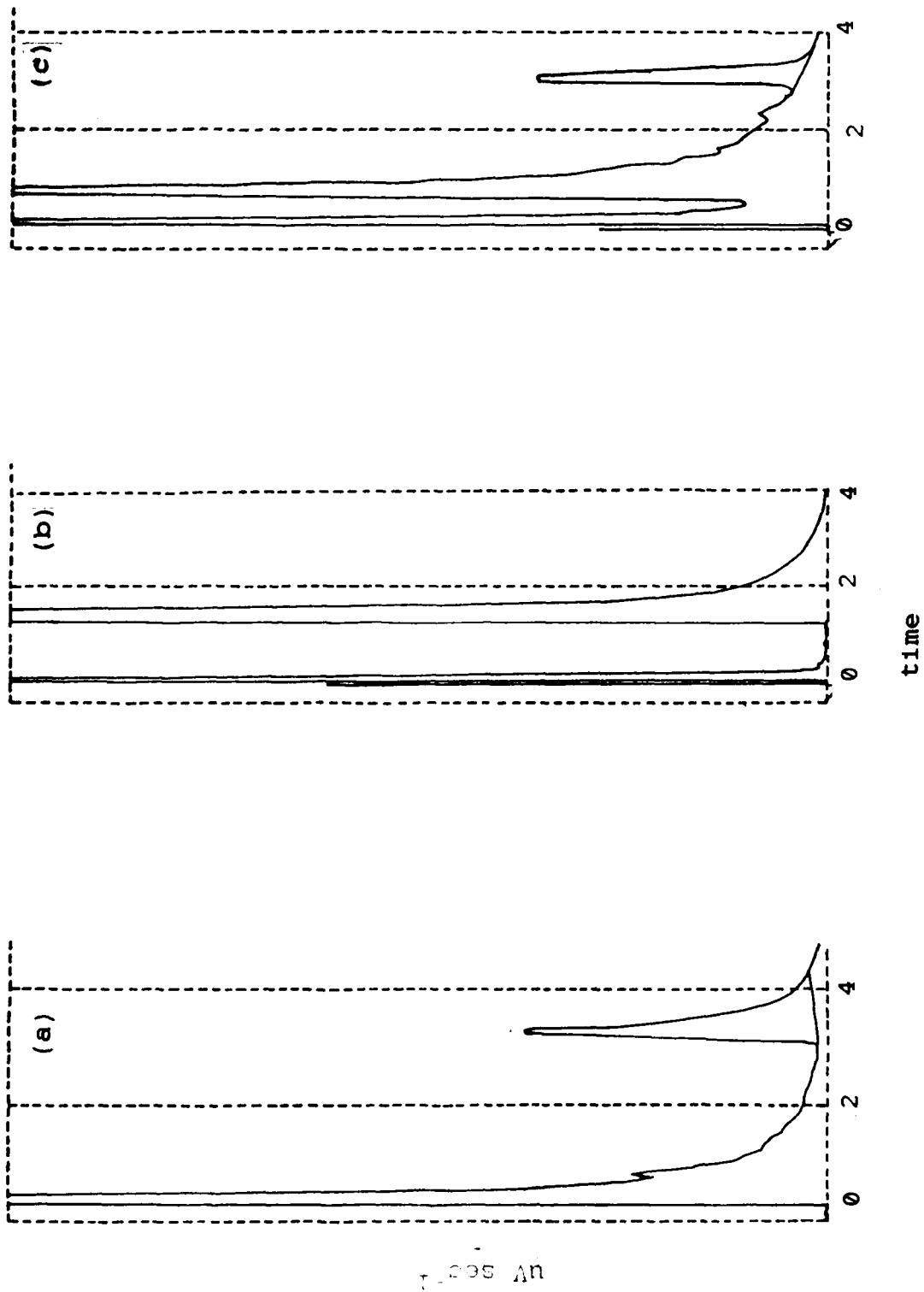


Figure 2. Chromatograms showing (a) metalaxyl (standard) (b) tomato (control) (c) tomato fortified with metalaxyl

standard metalaxyl solution ($\mu\text{g g}^{-1}$), A_2 - peak area of standard metalaxyl ($\mu\text{V Sec}^{-1}$), V_1 - Volume of sample solution injected (μl), m - mass of sample taken (g) and f - recovery factor= (100 / Per cent recovery).

The residues of metalaxyl in soil were analysed as explained under the section 3.4.1 excluding the step of column chromatography (3.5.1.3). The residues were expressed in μg per gram dry weight of soil, while tomato as μg per gram of fresh weight of plant samples. The recoveries of the fungicides or metabolite in soils and tomato are presented in Table 5.

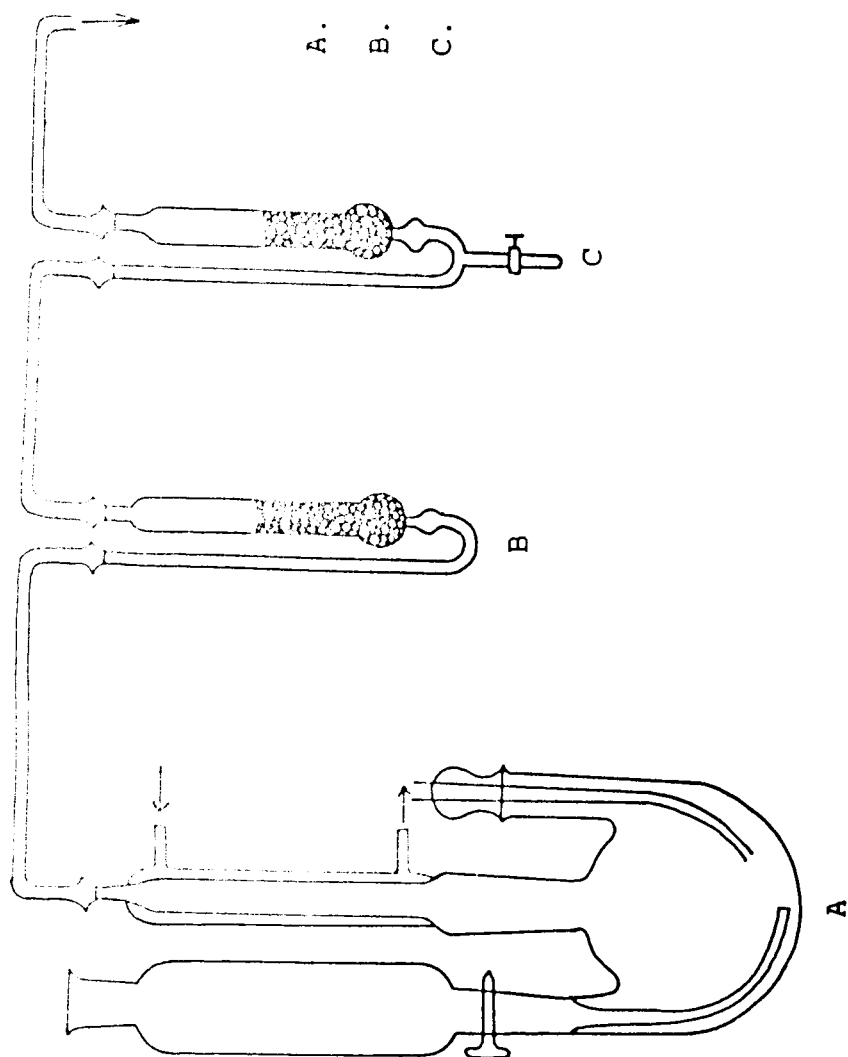
3.4.2 Mancozeb residues: The method of residue determination of mancozeb was followed as described by Keppel (1971), Vuik (1992) and Anon. (1993) (Bureau of Indian Standards) with little modifications.

3.4.2.1 Reagents :

- a) Viles reagent : Cupric acetate monohydrate (0.05 g) was dissolved in 25 ml water in a 1000 ml volumetric flask. To this 800 ml ethanol, 1 ml diethylamine and 20 ml triethanolamine were added and then made up the volume with ethanol.
- b) Sodium hydroxide (10 % W/V)
- c) Stannous chloride (40 % W/V)
- d) Carbon disulfide standard solution : To accurately weighed 25 ml volumetric flask containing 5 ml ethanol, about 0.1 ml CS_2 was added and closed the flask with stopper, reweighed and made up the volume to 25 ml with ethanol. From this 2 ml aliquot of solution was diluted with 100 ml ethanol. CS_2 was calculated in μg per ml.

3.4.2.2 Procedure : The apparatus used for the trapping of carbon disulfide (CS_2) from mancozeb was given in Fig. 3. For the first (H_2S) trap, 25 ml of 10 per cent NaOH solution and overlaid with 10 ml benzene were taken in a 250 ml joint conical flask. To the second (CS_2) trap 15 ml Viles (colour) reagent was taken in 40 ml joint graduated test tube. Connections were made as in Fig. 3, water was passed through reflux condenser and gentle vacuum was applied occasionally to the outlet of CS_2 trap.

A representative chopped 50 grams of fruits or foliage or air dried soil sample was taken in a 500 ml digestion flask. To this 100 ml distilled water, 20 ml



- A. Decomposition round bottom flask
- B. Hydrogen sulfide absorber
- C. Carbon disulfide absorber

Figure 3. Decomposition - distillation apparatus for determination of mancozeb residues

Table 5. Recoveries of metalaxyl, mancozeb and ethylenethiourea in soils and tomato.

Fungicide/ Metabolite	Quantities fortified ($\mu\text{g g}^{-1}$)	Quantities recovered ($\mu\text{g g}^{-1}$)		
		Soils		Tomato
		Bangalore	Chettalli	
Metalaxyl	2.0	1.81(90.5)	1.74(87.0)	1.80(90.0)
	4.0	3.77(94.3)	3.58(89.5)	3.67(91.8)
Mancozeb	10.0	8.16(81.6)	8.19(81.9)	7.81(78.1)
	20.0	17.46(87.3)	17.74(88.7)	16.92(84.7)
Ethylenethiourea	2.0	1.81(90.5)	1.76(88.0)	1.73(86.5)
	4.0	3.84(96.0)	3.71(92.8)	3.49(87.3)

Note: Figures in the parentheses indicate per cent recovery

concentrated hydrochloric acid (Conc. HCl) and 25 ml 40 per cent stannous chloride were added to the digestion flask and closed with stopper. The decomposition flask was heated and contents boiled for 45 minutes. After decomposition was complete vacuum line and inlet to CS₂ trap was disconnected. The inlet of CS₂ trap was washed with small quantities of ethanol and diluted to desired volume (25 ml) and mixed. Depending upon the concentration of CS₂ (mancozeb) dilution was carried out by taking known quantity of aliquot into required volume and made up with ethanol.

3.4.2.3 Spectrophotometry and calculation of mancozeb residues

The absorbance of sample solution in 1 cm quartz cell were measured at 380 nm using Viles reagent as blank. Mancozeb was quantitatively measured as equivalent carbon disulfide (CS₂) from the standard curve prepared earlier. The limit of detection of mancozeb was 1 µg per gram (0.5 µg CS₂). Mancozeb residues were calculated as follows:

$$\text{Mancozeb residue } (\mu\text{g g}^{-1}) = [(\mu\text{g CS}_2 \times 1.75 \times V \times f) / m]$$

where V- Volume or dilution (ml), f- recovery factor, m- mass of the sample (g) and (1.75)- Conversion factor from CS₂ to mancozeb.

3.4.3 Ethylenethiourea residues : The HPLC method for the residue determination of ethylenethiourea in the sample was followed as explained by Onley and Yip (1971); Onley *et al* . (1977); Smith *et al* . (1982) and Handa (1993) and as described in flow chart (Fig. 1).

3.4.3.1 Extraction : The extraction of ethylenethiourea from tomato or soil was carried out as outlined in section 3.4.1.1.

3.4.3.2. Partitioning : After partitioning with dichloromethane as explained under 3.4.1.2 for metalaxyl residue analysis, the aqueous phase containing ETU was partitioned in n-hexane (3 x 75 ml) through separatory funnel after vigorous shaking for 5 minutes. After separation of the layers, the lower aqueous phase was collected and concentrated to 10 ml in a rotary vacuum evaporator at 60°C. The pH of the concentrated extract was adjusted to 8.0 with the addition of required amounts of liquid ammonia.

3.4.3.3 Column chromatographic cleanup : Chromatographic columns (1.8 cm i.d x 30 cm length) were packed with a small plug of cotton overlaid with 1 cm of anhydrous sodium sulphate and mixture of 20 g activated alumina : celite 545 (4:1 w/w). The concentrated alkaline extract was poured slowly into the column and the column was eluted with 100 ml of methanol at the rate of 2 to 3 ml per minute. The elute was evaporated to dryness on a rotary vacuum evaporator at 50°C. The residues were dissolved in 10 ml of HPLC grade solvent mixture (water : methanol : 95:5 v/v) and filtered through millipore membrane filter using hypodermic syringe for ultra cleaning. An aliquot (20 µl) of the sample was injected into a Shimatzu LC 6A model HPLC with the help of microlitre syringe.

3.4.3.4 High performance liquid chromatography and calculations

The operational parameters of HPLC for ETU were:

Column : Shim-Pak CLC-ODS(M) 15 cm length x 46 mm i.d stainless steel
(5 micron particle diameter 100Å, pore diameter stationary phase)

Mobile phase and flow rate:-	Water : methanol (95:5 V/V) and 2 ml min ⁻¹
Detector	: UV spectrophotometer at 240 nm
Sensitivity	: 0.08 AUFS
Limit of detection	: 0.05 µg g ⁻¹
Retention time	: 0.75 minute

The peak of ETU was identified with the help of retention time and peak area was measured. The ETU residues were calculated by comparing with known standard (5 µg g⁻¹) Chromatogram (Fig 4) and similar formulae as represented in the section 3.4.1.5.

The estimation of ETU residues was carried out without any cleanup of soil extracts and other steps were similar as under 3.4.3.

3.5 Degradation kinetics

In all the persistence studies, the amount of pesticide residues that remained at different intervals were fitted to first order exponential decay equation in order to interpret the rate of degradation.

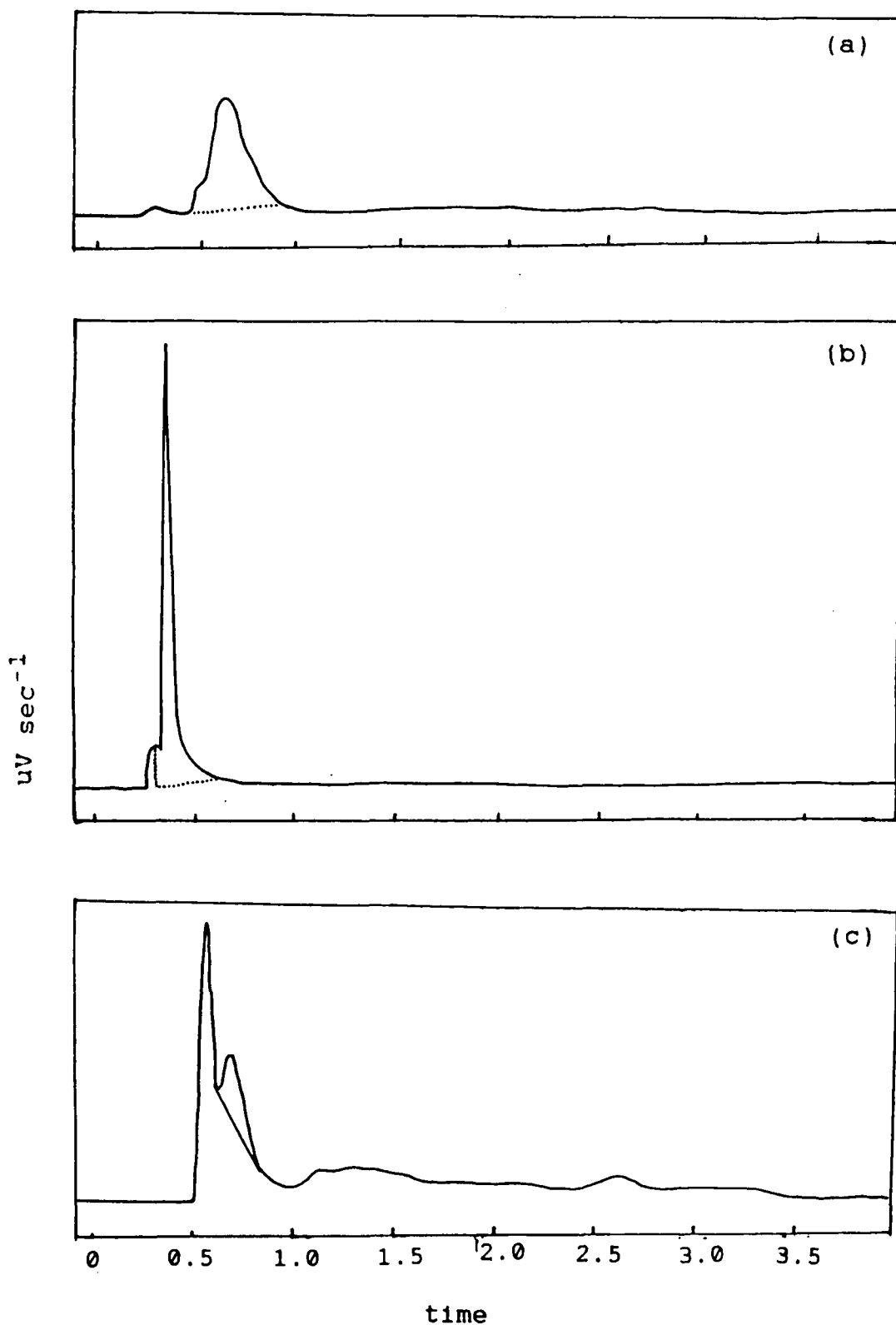


Figure 4. Chromatograms showing (a) ETU (standard)
(b) tomato (control) (c) tomato fortified
with metalaxyl-mancozeb (Ridomil - MZ)

The exponential decay equation as suggested by Nash and Beall (1980) is as follows:

$$C_t = C_o e^{-Kt} \quad \text{..... (5)}$$

where C_t - the concentration ($\mu\text{g g}^{-1}$) after time, t (day), C_o - initial concentration ($\mu\text{g g}^{-1}$) and K - rate constant (day^{-1}).

The constants K , C_o and determination coefficient (R^2) were calculated as given by Snedecor and Cochran (1967)

$$K = \frac{[n\sum\{\ln(C).t\} - \{\sum\ln(C)\}(\sum t)]}{[n\sum t^2 - (\sum t)^2]} \quad \text{..... (6)}$$

$$C_o = e^{[\sum\ln(C) - K(\sum t)] / n} \quad \text{.....(7)}$$

$$R^2 = \frac{[n\sum\ln(C).t - \{\sum\ln(C)\}(\sum t)]^2}{[n\sum\{\ln(C)^2\} - \{\sum\ln(C)\}^2][n\sum t^2 - (\sum t)^2]} \quad \text{.....(8)}$$

The time required to get 50 per cent (half-life) reduction in pesticide concentration was computed using equation (2) described for first order reaction kinetics.

$$\begin{aligned} C_t &= C_o e^{-Kt} \\ \ln(C_t) &= \ln(C_o) - Kt \\ Kt &= \ln(C_o) - \ln(C_t) = \ln(C_o/C_t) \\ t &= [\ln(C_o/C_t) / K] \\ \text{When } t &= t_{1/2}, C_t = C_o/2, \text{ then} \\ t_{1/2} &= [\ln\{(C_o)/C_{o/2}\} / K] = [\ln(2) / K] \\ t_{1/2} &= [0.693 / K] \quad \text{.....(9)} \end{aligned}$$

The assumption made to the equation (9) may holds good only in certain cases that the theoretical zero day (C_o) calculated should not deviate from the actual rate (*i.e.*, it should coincide to zero day value), otherwise the predicted half-life may not be true representative value.

If theoretical zero day (C_0) as calculated by the formula (7) is considered as it is, the half-life ($t_{1/2}$ modified), will be as follows:

$$t_{1/2} \text{ (modi.)} = \{ \ln [(C_0)/(C_{t_{1/2}})] / K \}$$

where $t_{1/2}$ (modi.)- half-life modified (days), C_0 - initial concentration ($\mu\text{g g}^{-1}$) derived from the formulae, $C_{t_{1/2}}$ - half of the actual concentration ($\mu\text{g g}^{-1}$) and K- degradation rate constant (day^{-1}). Half-life ($t_{1/2}$) (modified) was found to predict the most nearest situation.

3.5.1 Waiting periods : One of the most important uses of residue data is the determination of time required between application of pesticides and harvest of the crops. The waiting period is the period in days required to reduce the amount of pesticide residue below the prescribed maximum residue level (as established by regulatory agencies viz., FAO/WHO/EPA) for safe consumption of agricultural produce (human-being and animals).

This can be calculated (Hoskins, 1961) from the equation (5) as

$$\text{i.e. } C_t = C_0 e^{-Kt}$$

$$\ln(C_t) = \ln(C_0) - Kt$$

let $T_{(tol)}$ - time to reach the tolerance and again rearranging the equation

$$T_{(tol)} = \{ \ln [(C_0)/\text{MRL}] / K \}$$

where $T_{(tol)}$ - waiting period (days), MRL- maximum residue limit ($\mu\text{g g}^{-1}$) and K- degradation rate constant (day^{-1}).

RESULTS

IV. RESULTS

The studies on the fate of fungicides- metalaxyl (systemic) and mancozeb (contact) along with its major and significant metabolite ethylenethiourea, were carried out in relation to adsorption and degradation in soils, their plant uptake and persistence of residues on/in tomato fruits from the point of safety to the consumer. The results of these studies are presented in this chapter.

4.1 Adsorption of metalaxyl and ETU by soils

4.1.1 Metalaxyl : The adsorption of metalaxyl in soils are presented in Table 6 and Figure 5. Adsorption of metalaxyl was lower at low equilibrating solution and increased with increase in metalaxyl concentration. The adsorption of metalaxyl ranged from 3.20 to 36.02, 3.35 to 38.77 and 3.48 to 38.21 $\mu\text{g g}^{-1}$ in Bangalore, Chettalli and Hiriur soils respectively. With the increase in the quantity of metalaxyl added, the metalaxyl adsorption increased gradually but the percentage of adsorption showed a gradual decrease. The percentage adsorption decreased from 64.00 to 36.02; 66.90 to 38.77 and 69.56 to 38.21 in Bangalore, Chettalli and Hiriur soils respectively. Among the three soils studied, Hiriur soil adsorbed maximum amount of metalaxyl. The extent of adsorption was highest in these soils when the metalaxyl was added at the rate of 5.0 $\mu\text{g g}^{-1}$. Based on their adsorption capacity to metalaxyl, the three soils could be arranged in the decreasing order as Hiriur > Chettalli > Bangalore.

4.1.1.1 Adsorption isotherm constants of metalaxyl : The Freundlich constants (K and $1/n$) and the Gibb's free energy of metalaxyl are given in Table 7. The Freundlich constant "K", as an index of affinity of the adsorbent surface for the adsorbate was 5.870 $\mu\text{g ml}^{-1}$ in Bangalore soil, 6.511 $\mu\text{g ml}^{-1}$ in Chettalli soil and 7.281 $\mu\text{g ml}^{-1}$ in Hiriur soil, depicting the same order of adsorption mentioned earlier. The linearity of slopes " $1/n$ " were 0.696, 0.698 and 0.679 for Bangalore, Chettalli and Hiriur soils respectively. The coefficients of determination (R^2) were very high (>0.978) in all the soils studied. The sorption coefficients for organic carbon (K_{oc}) values were maximum in Hiriur soil (2275 $\mu\text{g ml}^{-1}$) and minimum in Chettalli (362 $\mu\text{g ml}^{-1}$) soil. The results relating to the change in free energy (dG°) of adsorption of metalaxyl were -4.367, -4.623 and -4.829 K J mol^{-1} in Bangalore, Chettalli and Hiriur soils respectively.

Table 6. Adsorption of metalaxyl in soils.

Soil	Parameters	Metalaxyl added ($\mu\text{g g}^{-1}$)					
		5.0	12.5	25.0	50.0	75.0	100.0
Bangalore	Equi. conc. ($\mu\text{g ml}^{-1}$)	0.36	1.29	2.67	6.09	9.16	12.79
	Amount adsorbed ($\mu\text{g g}^{-1}$)	3.20	6.01	11.63	19.52	29.19	36.02
	Per cent adsorption	64.00	48.52	46.52	39.04	38.91	36.02
Chettalli	Equi. conc. ($\mu\text{g ml}^{-1}$)	0.31	1.27	2.74	5.18	8.82	12.24
	Amount adsorbed ($\mu\text{g g}^{-1}$)	3.35	6.13	11.95	22.65	30.82	38.77
	Per cent adsorption	66.90	49.04	47.80	45.31	41.23	38.77
Hiriyur	Equi. conc. ($\mu\text{g ml}^{-1}$)	0.30	1.17	2.37	4.85	8.78	12.35
	Amount adsorbed ($\mu\text{g g}^{-1}$)	3.48	6.63	13.15	25.71	31.07	38.21
	Per cent adsorption	69.56	53.00	52.58	51.42	41.44	38.21

Equi. conc., = Equilibrium concentration

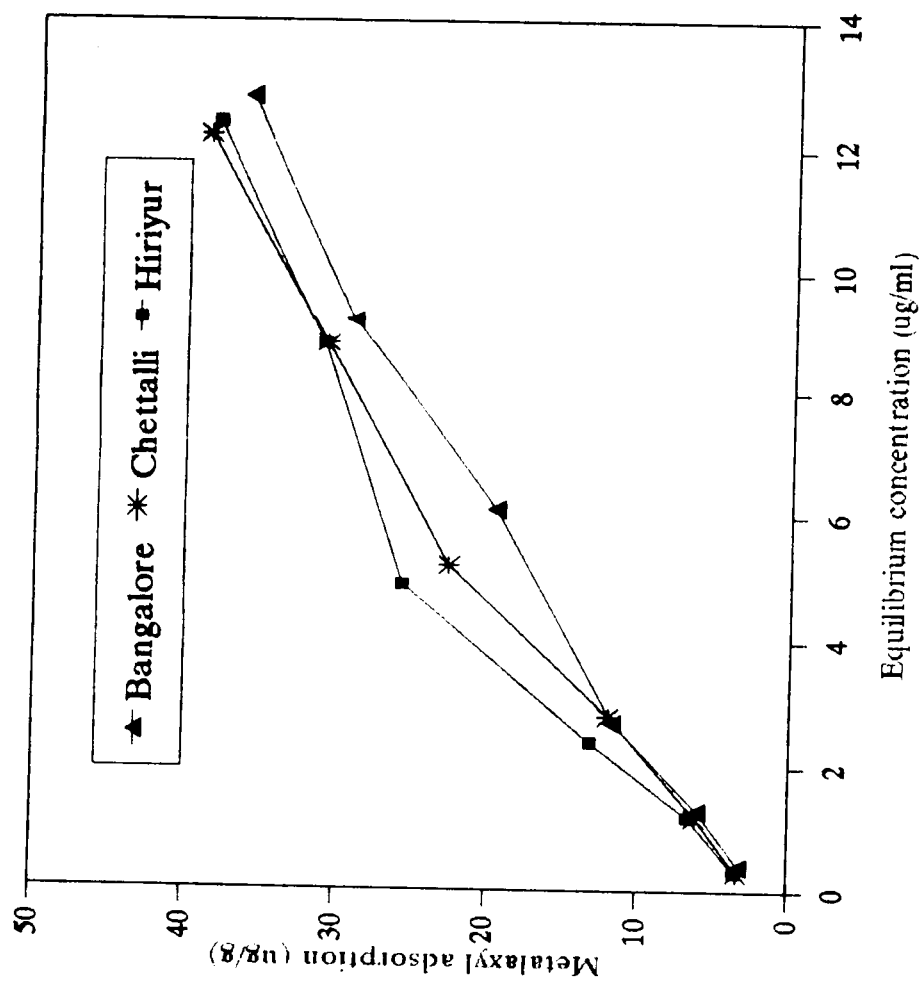


Figure 5. Adsorption of metalaxyl in soils

Table 7. Adsorption isotherm constants and free energy of metalaxyl in soils.

Soil	$\frac{\text{Freundlich constant}}{K}$	R^2	K_{OC}	Gibb's free energy dG (K J mol ⁻¹)
Bangalore	5.870	0.996	1174	-4.367
Chettalli	6.511	0.698	362	-4.623
Hiriyur	7.281	0.679	2275	-4.899

R^2 = coefficient of determination

K_{OC} = sorption coefficient for organic carbon

4.1.2 Ethylenethiourea : The quantities of ETU adsorbed by soils are presented in Table 8 and Figure 6. Amount adsorbed progressively increased with increase in amounts of ETU added. The per cent adsorption was maximum only from the application of $5.0 \mu\text{g g}^{-1}$ in all the soils. The adsorption of ETU was highest in Chettalli soil followed by Bangalore and Hiriur soils. Accordingly the per cent adsorption was about 40.60 in Bangalore, 41.00 in Chettalli and 31.00 in Hiriur soil. The quantity of ETU adsorption ranged from 2.03 to 21.10; 2.05 to 25.70 and 1.55 to 16.78 $\mu\text{g g}^{-1}$ for Bangalore, Chettalli and Hiriur soils respectively.

4.1.2.1 Adsorption isotherm constants of ETU : The adsorption isotherms of ETU for different soils are presented in Figure 6 and their parameters are given in Table 9. The Freundlich constant values were higher for Chettalli ($3.023 \mu\text{g ml}^{-1}$) than Bangalore ($2.306 \mu\text{g ml}^{-1}$) and Hiriur ($1.860 \mu\text{g ml}^{-1}$) soils. The $1/n$ values were 0.748, 0.778 and 0.755 for Bangalore, Chettalli and Hiriur soils and the coefficient of determination (R^2) values were greater than 0.947 indicating the best fit for Freundlich equation. The sorption coefficients for organic carbon (K_{oc}) values were lower ($<581 \mu\text{g ml}^{-1}$) in almost all the soils. The free energy (dG°) values for various soils ranged from -1.532 to -2.729 K J mol^{-1} for ETU adsorption.

4.2 Persistence of fungicides residues in soils

The results of the persistence study of the added fungicides and/or metabolite in the three soils and the influence of rate of applications, moisture regimes, temperatures and soil sterilization are detailed in the following paragraphs.

4.2.1 Metalaxyl : The persistence of metalaxyl in soils was studied by incubating soils at 2.0 and $4.0 \mu\text{g g}^{-1}$ treatment levels under field capacity (0.033 M Pa) conditions of the soil moistures maintained in a laboratory experiment. The data on the persistence of metalaxyl in soils as influenced by different factors are given in Tables 10 and 11. It could be seen that with advance of time, there was an incremental decrease in the concentration of metalaxyl following the applications at 2.0 and $4.0 \mu\text{g g}^{-1}$ (Fig. 7). Persistence of metalaxyl residues was for a longer period in Chettalli soil followed by Bangalore and Hiriur soils. When the application rate was $2.0 \mu\text{g g}^{-1}$, the residues of metalaxyl recovered were 1.55, 1.00 and $0.56 \mu\text{g g}^{-1}$ in Bangalore soil, 1.48, 0.86 and $0.61 \mu\text{g g}^{-1}$ in Chettalli soil and 1.47, 0.80 and

Table 8. Adsorption of ethylenethiourea in soils.

Soils	Parameters	Ethylenethiourea added ($\mu\text{g g}^{-1}$)				
		5.0	12.5	25.0	50.0	100.0
Bangalore	Equi. conc. ($\mu\text{g ml}^{-1}$)	0.59	1.93	3.94	7.99	11.68
	Amount adsorbed ($\mu\text{g g}^{-1}$)	2.03	2.84	5.30	10.05	16.65
	Per cent adsorption	40.60	22.72	21.20	20.10	22.20
Chettalli	Equi. conc. ($\mu\text{g ml}^{-1}$)	0.59	1.63	3.45	7.18	11.18
	Amount adsorbed ($\mu\text{g g}^{-1}$)	2.05	4.35	7.75	14.10	19.15
	Per cent adsorption	41.00	34.80	31.00	28.20	25.50
Hiriyur	Equi. conc. ($\mu\text{g ml}^{-1}$)	0.69	2.03	4.04	8.18	12.48
	Amount adsorbed ($\mu\text{g g}^{-1}$)	1.55	2.94	4.79	9.11	12.61
	Per cent adsorption	31.00	23.50	19.16	18.22	16.82

Equi. conc., = Equilibrium concentration

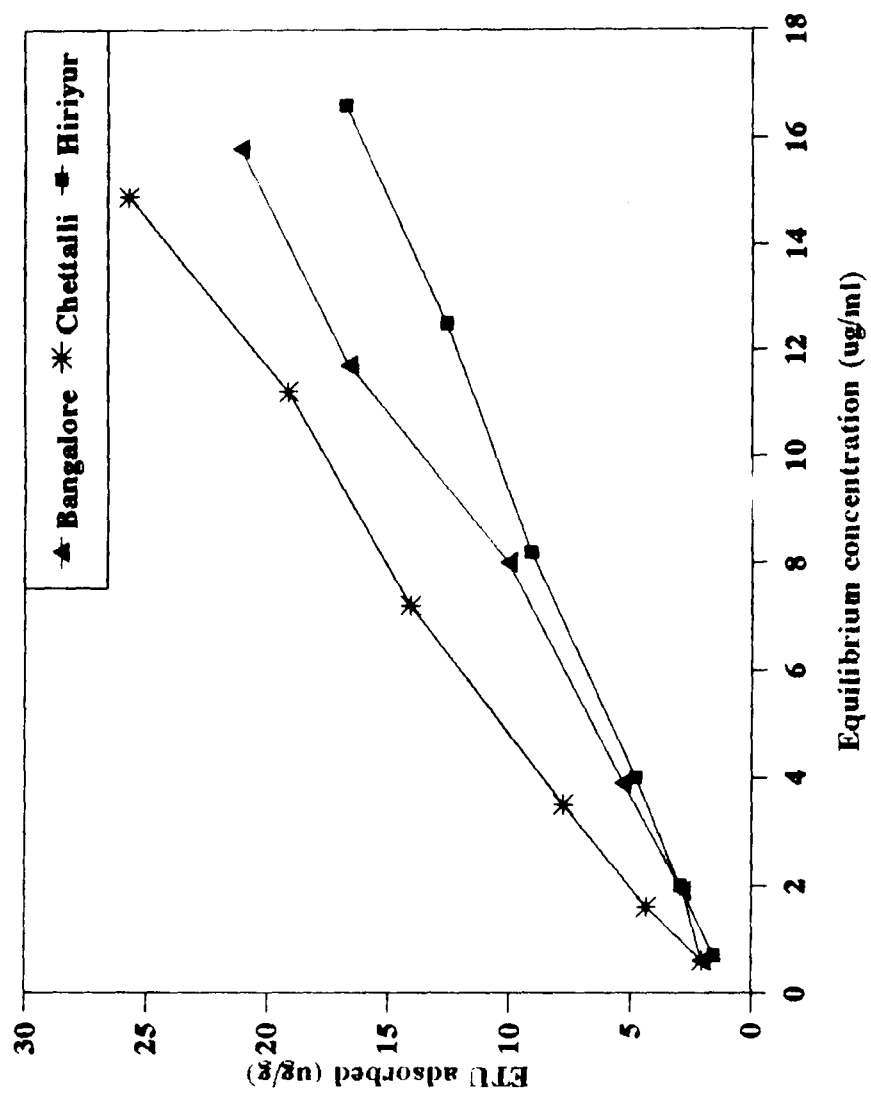


Figure 6 Adsorption of ETU in soils

Table 9. Adsorption isotherm constants and free energy values of ethylenethiourea in soils.

Soil	$\frac{\text{Freundlich constant}}{K}$	R^2	K_{OC}	Gibb's free energy $dG \text{ (K J mol}^{-1}\text{)}$
Bangalore	2.306	0.947	461	-2.062
Chettalli	3.023	0.778	168	-2.729
Hiriyur	1.860	0.755	581	-1.532

R^2 = coefficient of determination

K_{OC} = sorption coefficient for organic carbon

0.45 $\mu\text{g g}^{-1}$ in Hiriur soil on 5th, 45th and 80th day of incubation at field capacity respectively. While incubation of soil with 4.0 $\mu\text{g g}^{-1}$ metalaxyl registered a residue of 3.68 to 1.59, 3.34 to 1.61 and 3.51 to 1.19 $\mu\text{g g}^{-1}$ in Bangalore, Chettalli and Hiriur soils from 5th day to 80th day. With decrease in moisture content of soils from field capacity to air dry conditions, the rate of metalaxyl degradation decreased. In air dry conditions, the quantities of metalaxyl remaining at the end of the 80th day were 2.79, 2.15 and 2.17 $\mu\text{g g}^{-1}$ in Bangalore, Chettalli and Hiriur soils respectively at the dose of 4.0 $\mu\text{g g}^{-1}$ level. With increase in moisture level from air-dry to field capacity, the persistence of the metalaxyl residues was decreased and the degradation of the metalaxyl increased from 5 to 30 per cent. It is also observed that the difference in metalaxyl degradation between two moisture regimes were found to be more pronounced after 30 days of incubation period in all the three soils (Fig. 8).

The persistence of metalaxyl was comparatively more in soils incubated at 15°C temperature. However, the rate of degradation of metalaxyl was far less in Bangalore soil as compared to Chettalli and Hiriur soils incubated at 15°C temperature (Fig. 9). The recovery of metalaxyl after 5 and 80 days was in the order of 3.60 to 2.43, 3.47 to 1.97 and 3.40 to 2.81 $\mu\text{g g}^{-1}$ in Bangalore, Chettalli and Hiriur soils. Among the soils, the Hiriur soil recorded less persistence of metalaxyl as compared to Chettalli and Bangalore soils.

The degradation pattern of metalaxyl in three soils as influenced by autoclaving is presented in Table 10 and Figure 10. The persistence of metalaxyl in all the three soils was high in sterile soils than non-sterile soils throughout the incubation period. It could be seen that the metalaxyl concentration in the sterilized soil decreased with time, getting reduced to 70.0, 65.3 and 57.8 per cent in Bangalore, Chettalli and Hiriur soils respectively by 80th day of incubation while the persistence values of the metalaxyl in unsterilised soil were 39.8, 40.3 and 29.8 per cent for Bangalore, Chettalli and Hiriur soils.

4.2.1.2 Degradation kinetics of metalaxyl in soils

The data on the persistence and degradation of metalaxyl were fitted to first order exponential equation in order to interpret the rate of degradation and kinetics in different soils at different factors like application rates, moistures, temperatures and

Table 10. Persistence of metalaxyl residues in different soils as affected by rate of application and moisture regime.

Soils	Metalaxyl added ($\mu\text{g g}^{-1}$)	Moisture regimes	Metalaxyl recovered ($\mu\text{g g}^{-1}$) at different intervals (days)						
			5	10	20	30	45	60	80
Bangalore	2.0	Field capacity	1.55	1.32	1.28	1.06	1.00	0.92	0.56
	4.0	Field capacity	3.68	3.24	3.05	2.84	2.75	1.92	1.59
	4.0	Air dry	3.87	3.51	3.46	3.36	3.23	2.96	2.79
Chettalli	2.0	Field capacity	1.48	1.39	1.26	1.10	0.86	0.75	0.61
	4.0	Field capacity	3.34	3.15	3.01	2.61	2.29	2.05	1.61
	4.0	Air dry	3.89	3.53	3.44	3.01	2.77	2.37	2.15
Hiriyur	2.0	Field capacity	1.47	1.22	1.12	0.92	0.80	0.61	0.45
	4.0	Field capacity	3.51	3.09	2.97	2.54	2.29	1.53	1.19
	4.0	Air dry	3.87	3.49	3.38	2.88	2.64	2.40	2.17

Table 11. Persistence of metalaxyl in soils as affected by different temperatures and sterilization.

Soils	Temperature (°C)	Soil condition	Metalaxyl recovered ($\mu\text{g g}^{-1}$) at different intervals (days)						
			5	10	20	30	45	60	80
Bangalore	15	Non-sterile	3.60	3.38	3.24	3.10	2.97	2.50	2.43
	25	Non-sterile	3.68	3.24	3.05	2.84	2.75	1.92	1.59
	25	Sterile	3.90	3.66	3.54	3.43	3.33	2.93	2.80
Chettalli	15	Non-sterile	3.47	3.39	3.21	2.96	2.73	2.16	1.97
	25	Non-sterile	3.34	3.15	3.01	2.61	2.29	2.05	1.61
	25	Sterile	3.75	3.67	3.35	3.19	3.11	2.98	2.61
Hiriyur	15	Non-sterile	3.40	3.26	3.17	2.69	2.35	2.03	1.81
	25	Non-sterile	3.51	3.09	2.97	2.54	2.29	1.53	1.19
	25	Sterile	3.87	3.63	3.54	3.31	3.09	2.59	2.31

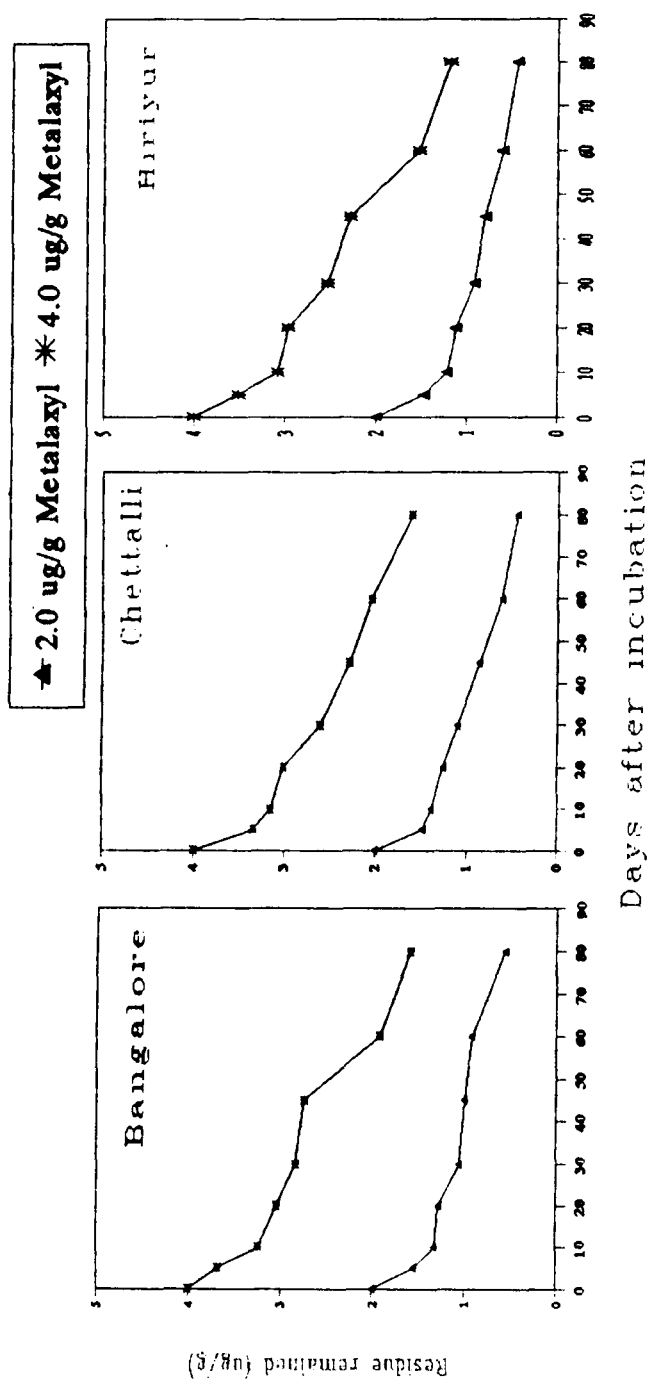


Figure 7 Persistence of metalaxyl residues in soils under the influence of application rates

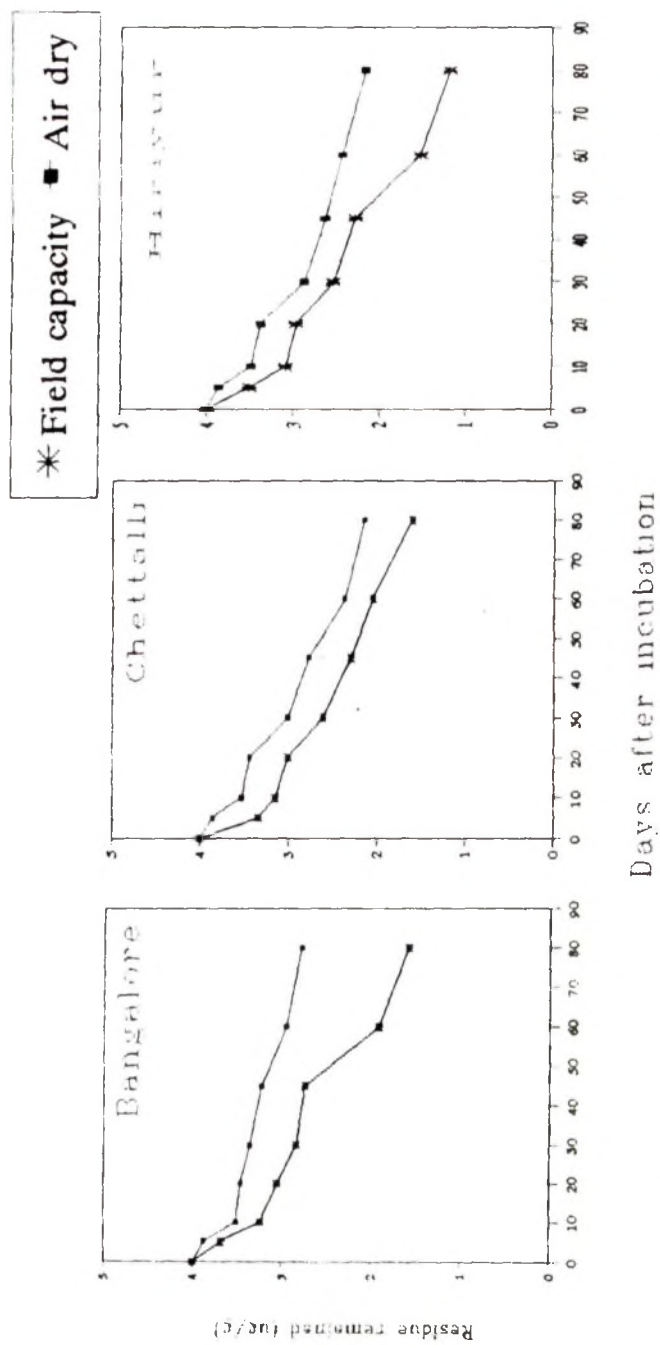


Figure 8. Persistence of metalaxyl residues in soils under the influence of moisture regimes

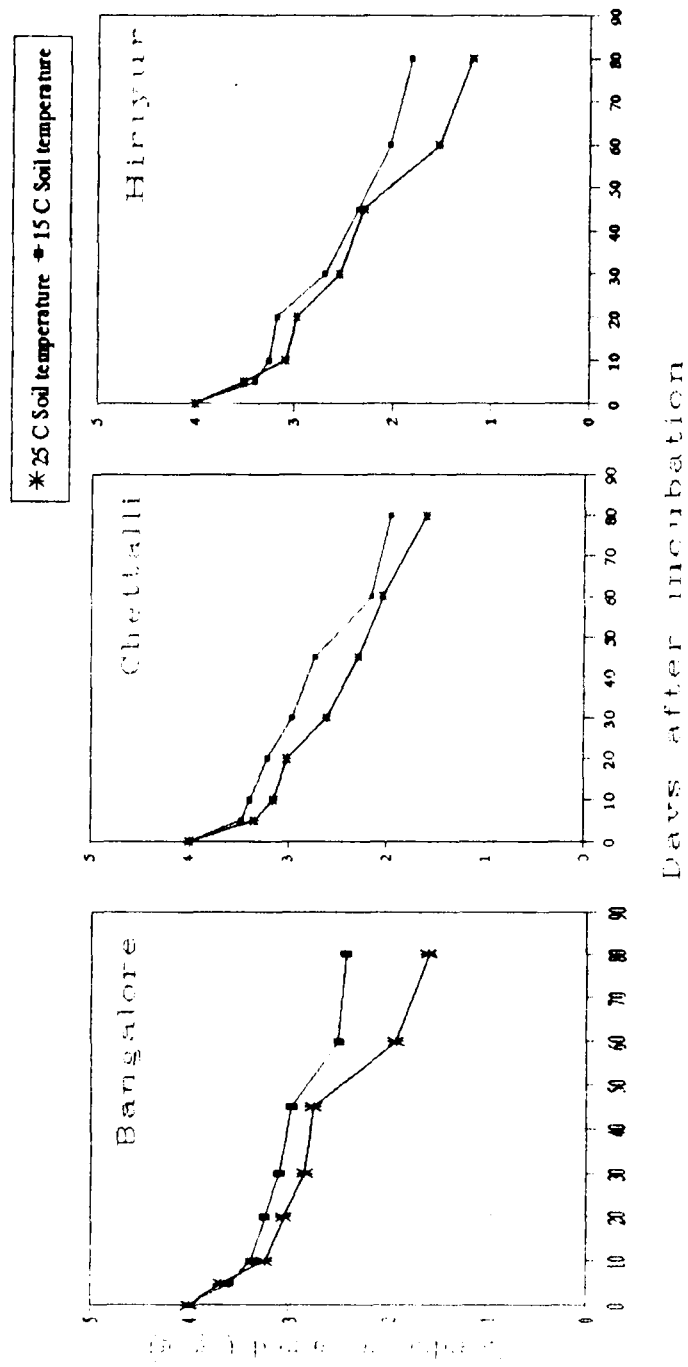


Figure 9 Persistence of metolaxyl residues in soils under the influence of soil temperatures

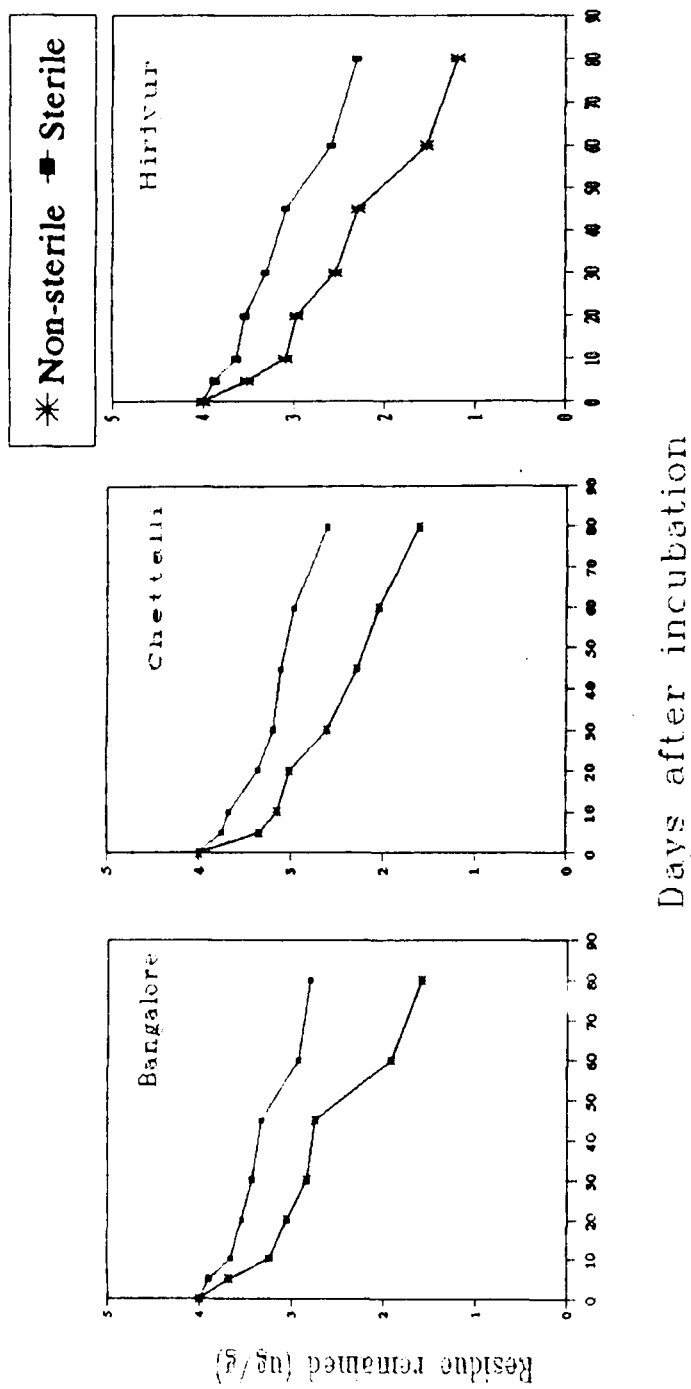


Figure 10. Persistence of metalaxyl residues in soils under the influence of micro-organisms

sterile soil conditions are presented in Table 12 and 13. The coefficient of determination (R^2) values were greater than 0.918.

The rate of degradation of metalaxyl followed an exponential pattern indicating that the process obeyed first order kinetics. The data were plotted as a straight line on natural logarithmic scale as $\ln(C_t) = \ln(C_0) - Kt$, where C_t - concentration of the fungicide ($\mu\text{g g}^{-1}$) at time t (days), C_0 - initial concentration added ($\mu\text{g g}^{-1}$) and K - degradation rate constant (day^{-1}).

The values of K were obtained either by the slope of the graph plotted $\ln(C_t)$ versus time or from the formulae. The values of C_0 either can be assumed or may be estimated by the formulae. Although ' K ' has units of " ug day^{-1} ", it is customarily expressed as " day^{-1} ". By utilising the above first order equation, the time required to get 50 per cent decrease in concentration was also noted considering two methods explained earlier. In almost all the soils, the regression equations were a better predictor of residues ($\mu\text{g g}^{-1}$) at any time (day) for the fungicides and metabolite studied.

The persistence of fungicide studied in soils or tomatoes are expressed as $C_t = C_0 e^{-Kt}$. The first order rate degradation constants (K_{deg}) were higher at $2.0 \mu\text{g g}^{-1}$ application rate (12.87×10^{-3} to $16.56 \times 10^{-3} \text{ day}^{-1}$) as compared to $4.0 \mu\text{g g}^{-1}$ (10.24×10^{-3} to $14.20 \times 10^{-3} \text{ day}^{-1}$). Higher rate constants for Hiriyrur soil ($16.56 \times 10^{-3} \text{ day}^{-1}$) reveal faster degradation of metalaxyl as compared to Chettalli ($13.50 \times 10^{-3} \text{ day}^{-1}$) and Bangalore ($12.87 \times 10^{-3} \text{ day}^{-1}$) soils.

The degradation rate constants (K_{deg}) decreased with decreasing moisture content from field capacity to air dry conditions in all the soils. The degradation rate constants were 4.20×10^{-3} , 7.83×10^{-3} and $7.97 \times 10^{-3} \text{ day}^{-1}$ under air dry moisture and 10.84×10^{-3} , 14.20×10^{-3} and $10.24 \times 10^{-3} \text{ day}^{-1}$ under field capacity in Bangalore, Hiriyrur and Chettalli soils respectively.

With the increase in soil temperature from 15°C to 25°C , the degradation rate constants were increased from 5.83×10^{-3} to 10.84×10^{-3} , 8.41×10^{-3} to 10.24×10^{-3} and 9.54×10^{-3} to $14.19 \times 10^{-3} \text{ day}^{-1}$ for Bangalore, Chettalli and Hiriyrur soils

Table 12. Equations explaining the kinetics of metalaxyl residues degradation in different soils as affected by application rates and moisture regimes.

Soils	Metalaxyl added ($\mu\text{g g}^{-1}$)	Moisture regimes	Exponential equation	$K_{(\text{deg})}$ (10^{-3} day^{-1})	Half-life (days)		R^2
					$t_{1/2}$	$t_{1/2}(\text{mod})$	
Bangalore	2.0	Field capacity	$C = 1.705 e^{(-0.0129)t}$	12.87	53.83	41.46	0.918
	4.0	Field capacity	$C = 3.886 e^{(-0.0108)t}$	10.84	63.96	61.28	0.955
	4.0	Air dry	$C = 3.848 e^{(-0.0042)t}$	4.20	165.11	155.81	0.945
Chettalli	2.0	Field capacity	$C = 1.687 e^{(-0.0135)t}$	13.50	51.34	38.74	0.956
	4.0	Field capacity	$C = 3.661 e^{(-0.0102)t}$	10.24	67.70	59.04	0.976
	4.0	Air dry	$C = 3.946 e^{(-0.0079)t}$	7.97	86.97	85.26	0.986
Hiriyur	2.0	Field capacity	$C = 1.635 e^{(-0.0166)t}$	16.56	41.84	29.69	0.958
	4.0	Field capacity	$C = 3.813 e^{(-0.0142)t}$	14.20	48.82	45.44	0.970
	4.0	Air dry	$C = 3.878 e^{(-0.0078)t}$	7.83	88.52	84.57	0.970

$K_{(\text{deg})}$ = Rate of residue degradation

$t_{1/2}(\text{mod})$ = Half-life (modified)

R^2 = Determination coefficient

Table 13. Equations explaining the kinetics of metalaxyl residues degradation in different soils as affected by temperatures and sterilization.

Soils	Temperature (°C)	Soil condition	Exponential equation	$K_{(deg)}$ (10^{-3} day^{-1})	Half-life (days) $t_{1/2}$	R^2
Bangalore	15	Non-sterile	$C = 3.735 e^{(-0.0058)t}$	5.83	118.87	0.939
	25	Non-sterile	$C = 3.886 e^{(-0.0108)t}$	10.84	63.96	0.955
	25	Sterile	$C = 3.928 e^{(-0.0044)t}$	4.39	157.93	0.968
Chettalli	15	Non-sterile	$C = 3.789 e^{(-0.0084)t}$	8.41	82.44	0.970
	25	Non-sterile	$C = 3.661 e^{(-0.0102)t}$	10.24	67.70	0.976
	25	Sterile	$C = 3.879 e^{(-0.0049)t}$	4.89	141.63	0.968
Hiriyur	15	Non-sterile	$C = 3.703 e^{(-0.0095)t}$	9.54	72.70	0.972
	25	Non-sterile	$C = 3.813 e^{(-0.0142)t}$	14.19	48.82	0.970
	25	Sterile	$C = 4.006 e^{(-0.0068)t}$	6.79	102.09	0.984

$K_{(deg)}$ = Rate of residue degradation

$t_{1/2(mod)}$ = Half-life (modified)

R^2 = Determination coefficient

respectively. The degradation rate constants were lowest in sterilized soils compared to non-sterilized soil. The values decreased from 10.84×10^{-3} to $4.39 \times 10^{-3} \text{ day}^{-1}$ in Bangalore, 10.24×10^{-3} to $4.89 \times 10^{-3} \text{ day}^{-1}$ in Chettalli and 14.19×10^{-3} to 6.79×10^{-3} in Hiriur soils.

The half-life of metalaxyl residues in soil varied from 41.84 to 165.11 days from different treatments. The half-life periods were increased with increasing level of application of metalaxyl from 2.0 to $4.0 \mu\text{g g}^{-1}$. In almost all the cases, the half-life calculated based on $t_{1/2} = [0.693 / K] = [\ln(2)/K]$ values were maximum compared to $t_{1/2}(\text{modified}) = [\ln(C_0/C_{t_{1/2}})/K]$. The differences in the half-life values of metalaxyl depended on the value of C_0 calculated based on the formulae explained earlier. The calculated C_0 values were less, irrespective of application rates except in sterilized soil of Hiriur. The calculated half-life ($t_{1/2}$) values were always greater than observed values (Tables 10 and 11). The persistence of metalaxyl in Chettalli soils noticed the highest half-life (67.70 days) at $4.0 \mu\text{g g}^{-1}$ application rate. The half-life values were increased from 61.28 to 155.81 days for Bangalore, 59.04 to 85.26 days for Chettalli and 45.44 to 85.57 days for Hiriur soils when the moisture content decreased to air dry from field capacity.

The half-life values of 107.14, 75.98 and 64.57 days were recorded for 15°C , temperature treatment while 61.28, 59.04 and 45.47 days for 25°C temperature conditions in Bangalore, Chettalli and Hiriur soils. The metalaxyl residues were degraded very slowly in sterile soils conditions as evidenced by its half-life ranging from 102.30 to 153.75 days compared to 45.47 to 61.28 days for non-sterile soil conditions in all the three soils.

4.2.2 Mancozeb : Quantities of mancozeb remained in soils as a function of various factors are presented in Tables (14 and 15) and Figures (11 to 14). It was observed that with increase in time of incubation, the quantities of mancozeb remaining in the soils decreased and the decrease in residues followed an exponential pattern. Persistence of mancozeb was slightly increased with increase in the quantities of mancozeb applied. The application rate of $10.0 \mu\text{g g}^{-1}$, resulted in persistence of $6.45 \mu\text{g g}^{-1}$ residues on the 5th day which decreased further with incubation to $0.87 \mu\text{g g}^{-1}$ on 60th day. While the residues from increased application rate

Table 14. Persistence of mancozeb residues in different soils as affected by rate of application and moisture regimes.

Soils	Mancozeb added ($\mu\text{g g}^{-1}$)	Moisture regimes	Mancozeb recovered ($\mu\text{g g}^{-1}$) at different intervals (days)					
			5	10	20	30	45	60
Bangalore	10.0	Field capacity	6.45 (0.52)	5.21 (0.45)	3.40 (0.47)	1.92 (0.58)	1.24 (0.47)	0.87 (0.31)
	20.0	Field capacity	12.83 (0.87)	10.35 (0.72)	6.63 (0.75)	5.67 (0.99)	3.93 (0.68)	2.46 (0.49)
	20.0	Air dry	15.74	12.04	9.18	7.62	5.87	3.82
Chettalli	10.0	Field capacity	5.90 (0.45)	4.91 (0.47)	3.35 (0.58)	2.41 (0.53)	1.69 (0.52)	1.20 (0.37)
	20.0	Field capacity	14.15 (0.75)	10.60 (0.76)	8.37 (0.89)	6.18 (0.94)	4.29 (0.85)	2.43 (0.64)
	20.0	Air dry	17.78	12.46	9.89	7.54	6.27	4.97
Hiriyur	10.0	Field capacity	5.67 (0.44)	4.98 (0.53)	3.53 (0.55)	1.87 (0.57)	1.60 (0.38)	1.18 (0.33)
	20.0	Field capacity	14.46 (0.70)	10.03 (0.88)	8.63 (0.96)	5.59 (0.95)	4.11 (0.79)	3.44 (0.61)
	20.0	Air dry	17.90	11.33	9.59	7.38	5.66	4.97

Note: Figures in parentheses indicate the ethylenethiourea

Table 15. Persistence of mancozeb in soils as affected by different temperatures sterilization

Soils	Temperature (°C)	Soil condition	Mancozeb recovered ($\mu\text{g g}^{-1}$) at different intervals(days)					
			5	10	20	30	45	60
Bangalore	15	Non-sterile	14.25	11.77	8.61	6.38	4.61	3.23
	25	Non-sterile	12.83	10.35	6.63	5.67	3.93	2.46
	25	Sterile	16.14	13.40	11.15	9.11	6.87	5.69
Chettalli	15	Non-sterile	15.37	11.86	9.87	7.19	5.29	3.84
	25	Non-sterile	14.15	10.60	8.37	6.18	4.29	2.43
	25	Sterile	16.04	12.65	10.54	8.49	6.87	5.76
Hiriyur	15	Non-sterile	15.10	11.29	9.37	6.75	5.52	3.79
	25	Non-sterile	14.46	10.03	8.63	5.59	4.11	3.44
	25	Sterile	17.30	14.04	11.93	8.54	7.05	6.27

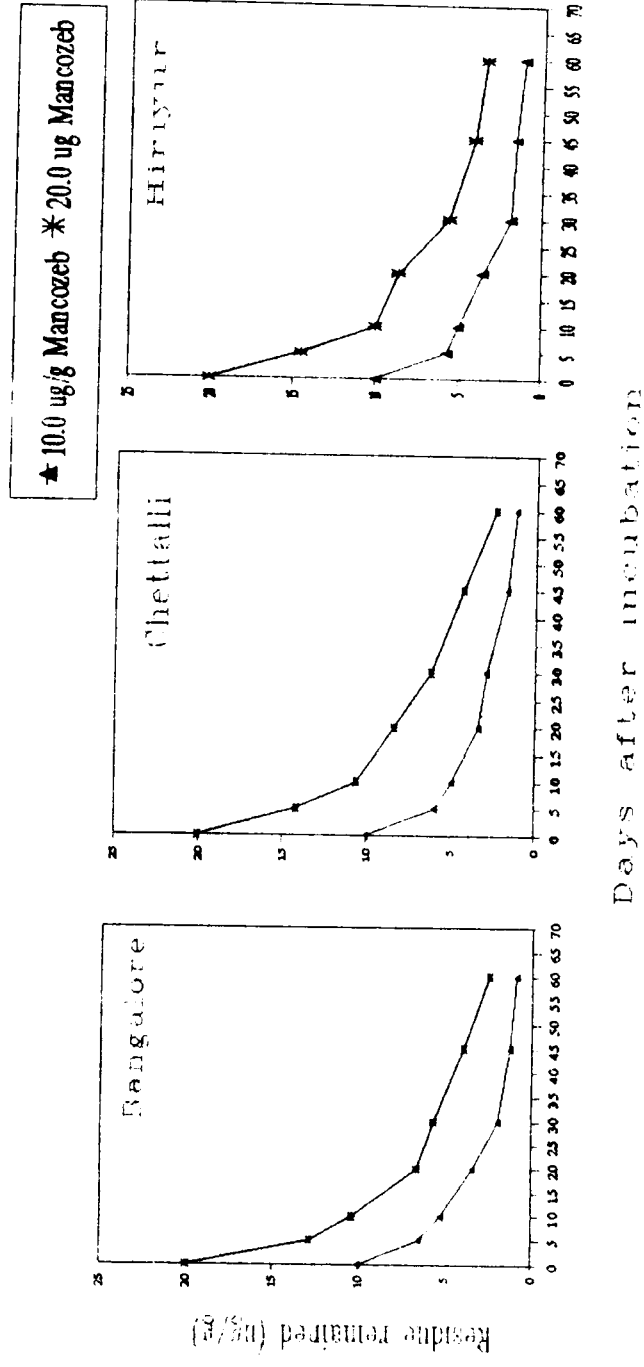


Figure 11. Persistence of mancozeb residues in soils under the influence of application rates

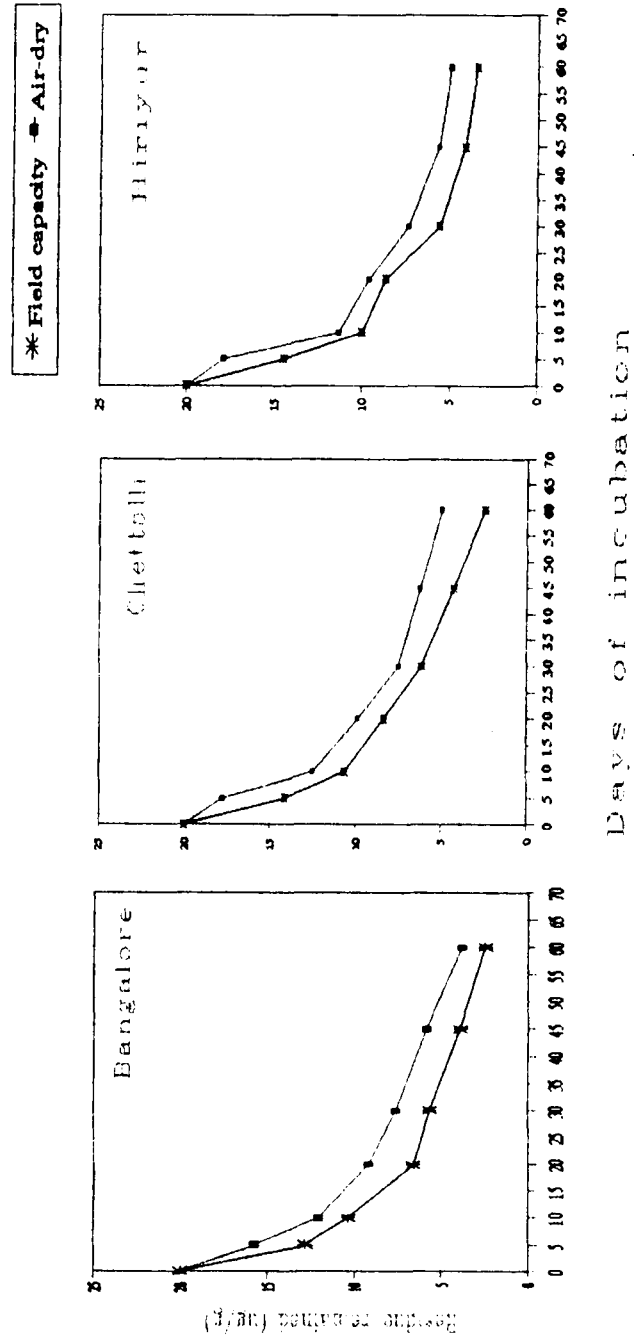


Figure 12 Persistence of mancozeb residues in soils under the influence of soil moisture-regimes

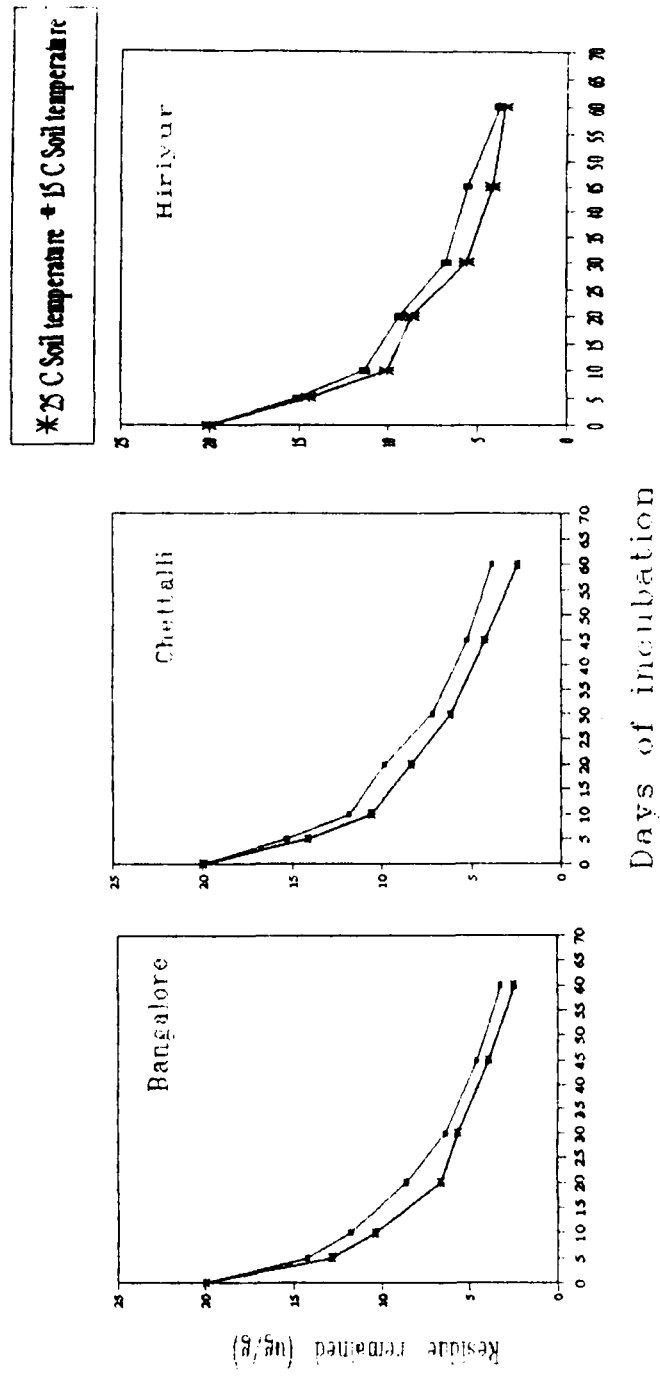


Figure 13. Persistence of mancozeb residues in soils under the influence of soil temperatures

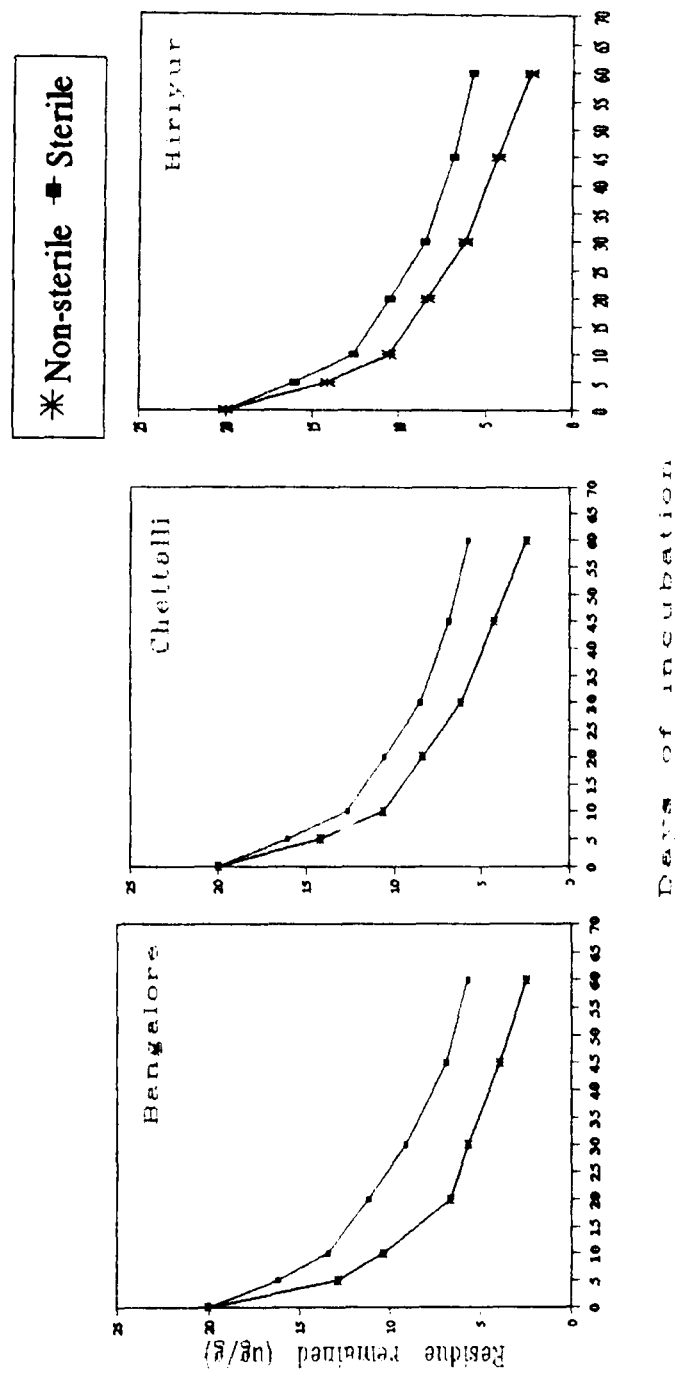


Figure 14. Persistence of mancozeb residues in soils under the influence of micro-organisms

of $20.0 \mu\text{g g}^{-1}$ resulted in the persistence of $12.83 \mu\text{g g}^{-1}$ on 5th day and only $2.46 \mu\text{g g}^{-1}$ on 60th day in Bangalore soil, whereas the persistence of $5.90 \mu\text{g g}^{-1}$ in Chettalli soils after 5 days was found to reduce to $1.20 \mu\text{g g}^{-1}$ by 60th day and $5.67 \mu\text{g g}^{-1}$ in Hiriur soil after 5 days reduced to $1.18 \mu\text{g g}^{-1}$ by 60th day respectively from the application rate of $10.0 \mu\text{g g}^{-1}$. Similarly the higher application rate of $20.0 \mu\text{g g}^{-1}$ resulted in the persistence of 14.15 and $14.46 \mu\text{g g}^{-1}$ after 5 days in Chettalli and Hiriur soils were reduced to 2.43 and $3.44 \mu\text{g g}^{-1}$ respectively by 60 days of incubation (Fig.11). The residues of 12.83 , 14.15 and $14.46 \mu\text{g g}^{-1}$ were recorded on 5th day from $20.0 \mu\text{g g}^{-1}$ mancozeb treatment when soil was maintained at field capacity level of moisture in Bangalore, Chettalli and Hiriur soils.

However, under air-dry condition, higher residues of 15.74 , 17.70 and $17.90 \mu\text{g g}^{-1}$ were detected on 5th day in Bangalore, Chettalli and Hiriur soils respectively (Fig.12). Similar persistence pattern of mancozeb residues was found in all the three soils. Soil moisture was found to influence greatly the rate of degradation of mancozeb residues in soils as there was one and half fold higher recovery of residues in air dry soils to that of field capacity level of moisture. Incubation of mancozeb treated soil at 15°C temperature resulted in the recovery of 14.25 , 15.37 and $15.10 \mu\text{g g}^{-1}$ residues on 5th day that were reduced to 3.23 , 3.84 and $3.79 \mu\text{g g}^{-1}$ by 60 days of incubation respectively for Bangalore, Chettalli and Hiriur soils. Whereas incubation at 25°C temperature the residues left in Bangalore, Chettalli and Hiriur soils were 12.83 , 14.15 and $14.46 \mu\text{g g}^{-1}$ on 5th day and 2.46 , 2.43 , $3.44 \mu\text{g g}^{-1}$ on 60th day respectively. Finally, the temperatures had only slight effect on the degradation of mancozeb in all the soils (Fig. 13).

Sterile conditions increased the persistence of mancozeb as compared to non-sterile soils. In all the incubation periods, nearly one and half to two times higher residues were recovered in sterile as compared to non-sterile soils. The different soil types did not showing marked effect on the degradation of mancozeb (Fig.14). The residues were decreased to 10.35 , 10.60 and $10.03 \mu\text{g g}^{-1}$ in non-sterile condition, while 13.40 , 12.65 and $14.04 \mu\text{g g}^{-1}$ in sterile conditions of Bangalore, Chettalli and Hiriur soils by 10th day following application of $20.0 \mu\text{g g}^{-1}$ mancozeb.

The ETU converted from mancozeb in the soil was also monitored from different application rates in all the soils (Table 14). It was found that even within 5 days of application of mancozeb ($20.0 \mu\text{g g}^{-1}$) to soils appreciable quantities of ETU were recovered to the extent of 0.87, 0.75 and $0.70 \mu\text{g g}^{-1}$ in Bangalore, Chettalli and Hiriur soils respectively. Similarly, the residues of ETU on 5th day in Bangalore, Chettalli and Hiriur soils were 0.52, 0.45 and $0.44 \mu\text{g g}^{-1}$ level from $10.0 \mu\text{g g}^{-1}$ level of application rate. The residues of ETU were increased up to 30 days of incubation and declined thereafter. The maximum ETU content in mancozeb treated soil were 0.99, 0.94 and $0.95 \mu\text{g g}^{-1}$ at $20.0 \mu\text{g g}^{-1}$ level, while 0.58, 0.53 and $0.57 \mu\text{g g}^{-1}$ at $10.0 \mu\text{g g}^{-1}$ levels on 30th day in Bangalore, Chettalli and Hiriur soils respectively. In this study, the rate at which ETU formed and degraded will not be very clear since both the processes may be occurring simultaneously in soils.

4.2.2.1 Degradation kinetics of mancozeb in soils : The results of the persistence and degradation of mancozeb were fitted to first order exponential equation, to predict the rate of degradation. The results presented in the Tables 16 and 17 depict the equations explaining mancozeb residues degradation kinetics under the influence of different factors. The determination coefficients (R^2) were greater than 0.916 in all the soils from treatments indicating degradation followed first order exponential equation.

It was found that the equations presented were better predictor of the residues at time (t). The rate constants were higher at lower dose of application ($10.0 \mu\text{g g}^{-1}$) when compared to higher dose of rate of application ($20.0 \mu\text{g g}^{-1}$) in soils. In Bangalore soil, the rate constants were higher (31.84×10^{-3} to $40.22 \times 10^{-3} \text{ day}^{-1}$) compared to Chettalli (32.31×10^{-3} to $32.74 \times 10^{-3} \text{ day}^{-1}$) and Hiriur (28.45×10^{-3} to $33.72 \times 10^{-3} \text{ day}^{-1}$) soils.

The rate constants were found to be lower in air-dry moisture conditions compared to field capacity in all the soils. The K_{deg} values were highest in Bangalore ($25.61 \times 10^{-3} \text{ day}^{-1}$), followed by Hiriur ($23.33 \times 10^{-3} \text{ day}^{-1}$) and Chettalli ($22.96 \times 10^{-3} \text{ day}^{-1}$) soils under air dry moisture regimes. The similar values for field capacity moisture regimes were 31.84×10^{-3} , 32.31×10^{-3} and $28.45 \times 10^{-3} \text{ day}^{-1}$ for Bangalore, Chettalli and Hiriur soils respectively.

Table 16. Equations explaining the kinetics of mancozeb residues degradation in different soils as affected by application rate and moisture regime.

Soils	Mancozeb added ($\mu\text{g g}^{-1}$)	Moisture regimes	Exponential equation	$K_{(\text{deg})}$ (10^{-3} day^{-1})	Half-life (days)		R^2
					$t_{1/2}$	$t_{1/2}(\text{mod})$	
Bangalore	10.0	Field capacity	$C = 8.057 e^{(-0.0402)t}$	40.22	17.23	11.86	0.970
	20.0	Field capacity	$C = 15.519 e^{(-0.0318)t}$	31.84	21.76	13.80	0.958
	20.0	Air dry	$C = 17.293 e^{(-0.0256)t}$	25.61	27.06	21.39	0.972
Chettalli	10.0	Field capacity	$C = 7.422 e^{(-0.0327)t}$	32.74	21.16	12.07	0.951
	20.0	Field capacity	$C = 16.897 e^{(-0.0323)t}$	32.31	21.45	16.23	0.980
	20.0	Air dry	$C = 17.507 e^{(-0.0229)t}$	22.96	30.19	24.39	0.942
Hiriyur	10.0	Field capacity	$C = 7.282 e^{(-0.0337)t}$	33.72	20.55	11.15	0.924
	20.0	Field capacity	$C = 15.845 e^{(-0.0285)t}$	28.45	24.33	16.18	0.935
	20.0	Air dry	$C = 17.093 e^{(-0.0233)t}$	23.33	29.71	22.98	0.916

$K_{(\text{deg})}$ = Rate of residue degradation

$t_{1/2}(\text{mod})$ = Half-life (modified)

R^2 = Determination co-efficient

Table 17. Equations explaining the kinetics of mancozeb residues degradation in different soils as affected by temperatures and sterilization.

Soils	Temperature (°C)	Soil condition	Exponential equation	K(deg) (10 ⁻³ day ⁻¹)	Half-life (days)		R ²
					t _{1/2}	t _{1/2} (mod)	
Bangalore	15	Non-sterile	C = 16.757 e ^{(-0.0289)t}	28.86	24.01	17.89	0.976
	25	Non-sterile	C = 15.519 e ^{(-0.0318)t}	31.84	21.76	13.80	0.958
	25	Sterile	C = 17.666 e ^{(-0.0203)t}	20.25	34.23	28.10	0.972
Chettalli	15	Non-sterile	C = 17.267 e ^{(-0.0263)t}	26.28	26.38	20.78	0.976
	25	Non-sterile	C = 16.897 e ^{(-0.0323)t}	32.31	21.45	16.23	0.980
	25	Sterile	C = 17.048 e ^{(-0.0198)t}	19.83	34.95	26.90	0.947
Hiriyur	15	Non-sterile	C = 16.710 e ^{(-0.0258)t}	25.82	26.84	19.88	0.960
	25	Non-sterile	C = 15.845 e ^{(-0.0285)t}	28.48	24.33	16.16	0.935
	25	Sterile	C = 18.085 e ^{(-0.0198)t}	19.79	35.02	29.94	0.951

K(deg) = Rate of residue degradation

t_{1/2}(mod) = Half-life (modified)

R² = Determination co-efficient

The values of rate constant for Bangalore, Chettalli and Hiriur soils were 28.86×10^{-3} and $25.82 \times 10^{-3} \text{ day}^{-1}$ when soil was incubated at 15°C and 31.84×10^{-3} , 32.3×10^{-3} and $28.48 \times 10^{-3} \text{ day}^{-1}$ when maintained at 25°C temperature. Therefore, the degradation rate constants (K_{deg}) decreased with increasing temperature from 15°C to 25°C , indicating that no significant change in degradation of mancozeb was observed due to change in temperature from any of the soils.

It was observed that the persistence of mancozeb was higher in sterile soil condition as compared to non-sterile. The K_{deg} values were reduced from 31.84×10^{-3} to 20.25×10^{-3} , 32.21×10^{-3} to 19.83×10^{-3} and 28.48×10^{-3} to $19.79 \times 10^{-3} \text{ day}^{-1}$ for Bangalore, Chettalli and Hiriur soils.

Since the rate of degradation of mancozeb in soils is high, the half-life ($t_{1/2}$) period was found to be very low compared to metalaxyl under different factors like application rates, soil moistures, soil temperatures, sterile and non-sterile soil conditions. However, the half-life were slightly higher from higher rate of applications. The half-life for 10.0 and $20.0 \mu\text{g g}^{-1}$ mancozeb were 11.86 and 13.80 , 12.07 and 16.23 and 11.15 and 16.18 days in Bangalore, Chettalli and Hiriur soils. The time required for 50 per cent degradation of mancozeb in Bangalore soil were low as compared to Hiriur and Chettalli soils and the corresponding values were 13.80 , 16.18 and 16.23 days for field capacity moisture and 21.39 , 22.98 and 24.39 days for air dry moisture regimes. The half-life values ranged from 17.89 to 28.78 , 13.80 to 16.23 days for 15°C and 25°C temperatures maintained in different soils. The notable values of half-life were recorded in sterile soil condition (26.90 to 29.94 days) compared to non-sterile soil (13.80 to 16.23 days).

4.2.3 Ethylenethiourea : Quantities of ETU that persisted in soils at different periods following the effect of different factors are presented in Tables 18 and 19 (Fig. 15 to 18). The persistence of ETU in soils was observed to be largely dependent on the changes in moisture, temperature and microbial activities. In all the soils, its concentrations decreased relatively at faster rates of loss in lower doses as compared to higher doses (Figure 15). The amount of ETU remaining at the end of 60th day of incubation were not detected from $2.0 \mu\text{g g}^{-1}$ treatment, while 0.22 , 0.14 and $0.12 \mu\text{g g}^{-1}$ residues were detected from $4.0 \mu\text{g g}^{-1}$

Table 18. Persistence of ethylenethiourea residues in different soils as affected by rate of application and moisture regimes.

Soils	ETU added ($\mu\text{g g}^{-1}$)	Moisture regimes	ETU recovered ($\mu\text{g g}^{-1}$) at different intervals (days)					
			5	10	20	30	45	60
Bangalore	2.0	Field capacity	1.58	0.97	0.60	0.35	0.16	ND
	4.0	Field capacity	3.38	2.57	1.77	1.15	0.59	0.22
	4.0	Air dry	3.64	3.30	2.47	2.13	1.59	0.38
Chettalli	2.0	Field capacity	1.46	0.84	0.61	0.33	0.16	ND
	4.0	Field capacity	3.20	2.31	1.73	1.05	0.45	0.14
	4.0	Air dry	3.55	3.16	2.46	1.90	1.36	0.66
Hiriyur	2.0	Field capacity	1.43	0.91	0.68	0.29	0.09	ND
	4.0	Field capacity	3.34	2.53	1.78	0.99	0.42	0.12
	4.0	Air dry	3.57	3.29	2.53	1.87	1.03	0.52

ND = Not detected

Table 19. Persistence of ethylenethiourea in soils as affected by different temperatures and sterilization.

Soils	Temperature (°C)	Soil condition	ETU recovered ($\mu\text{g g}^{-1}$) at different intervals (days)					
			5	10	20	30	45	60
Bangalore	15	Non-sterile	3.56	3.08	2.10	1.50	0.81	0.43
	25	Non-sterile	3.38	2.57	1.77	1.15	0.59	0.22
	25	Sterile	3.65	3.28	2.69	2.20	1.90	1.08
Chettalli	15	Non-sterile	3.36	2.73	2.11	1.34	0.66	0.42
	25	Non-sterile	3.20	2.31	1.73	1.05	0.45	0.14
	25	Sterile	3.66	3.21	2.81	1.97	1.54	0.83
Hiriyur	15	Non-sterile	3.45	2.91	2.10	1.33	0.52	0.30
	25	Non-sterile	3.34	2.53	1.78	0.99	0.42	0.12
	25	Sterile	3.77	3.24	2.92	2.11	1.36	0.73

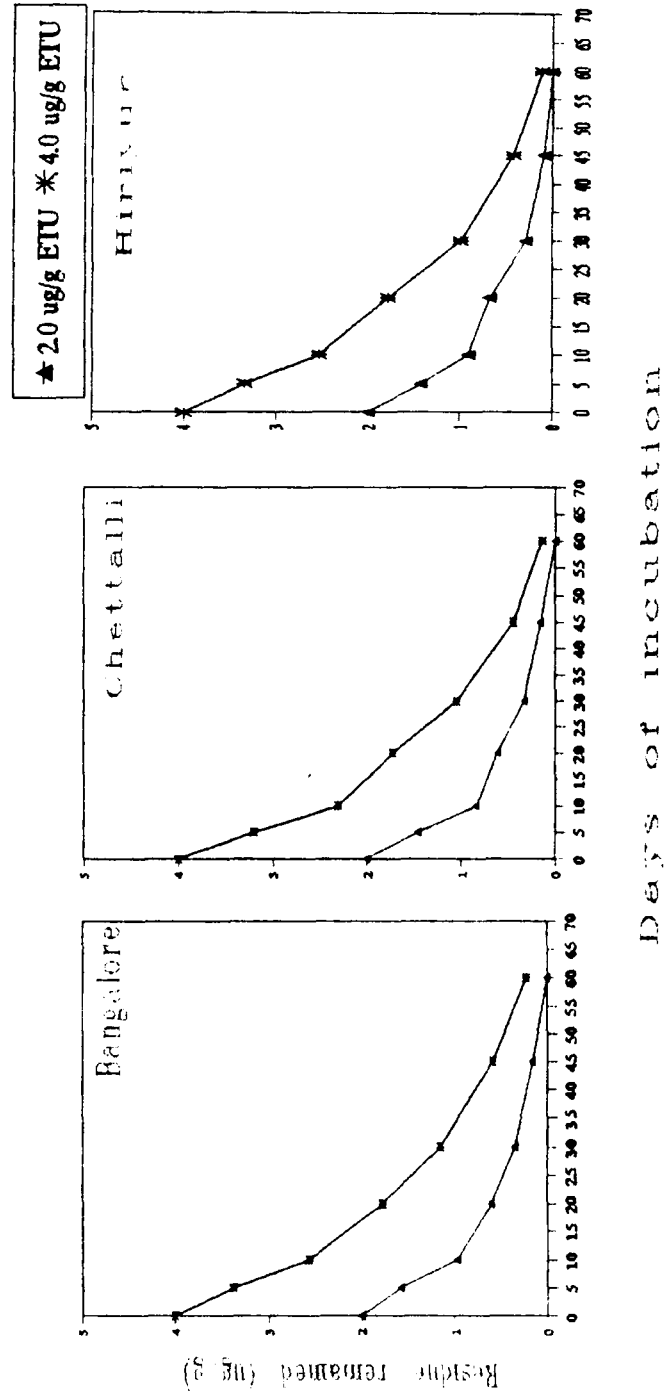


Figure 15 Persistence of ETU residues in soils under the influence of application rates

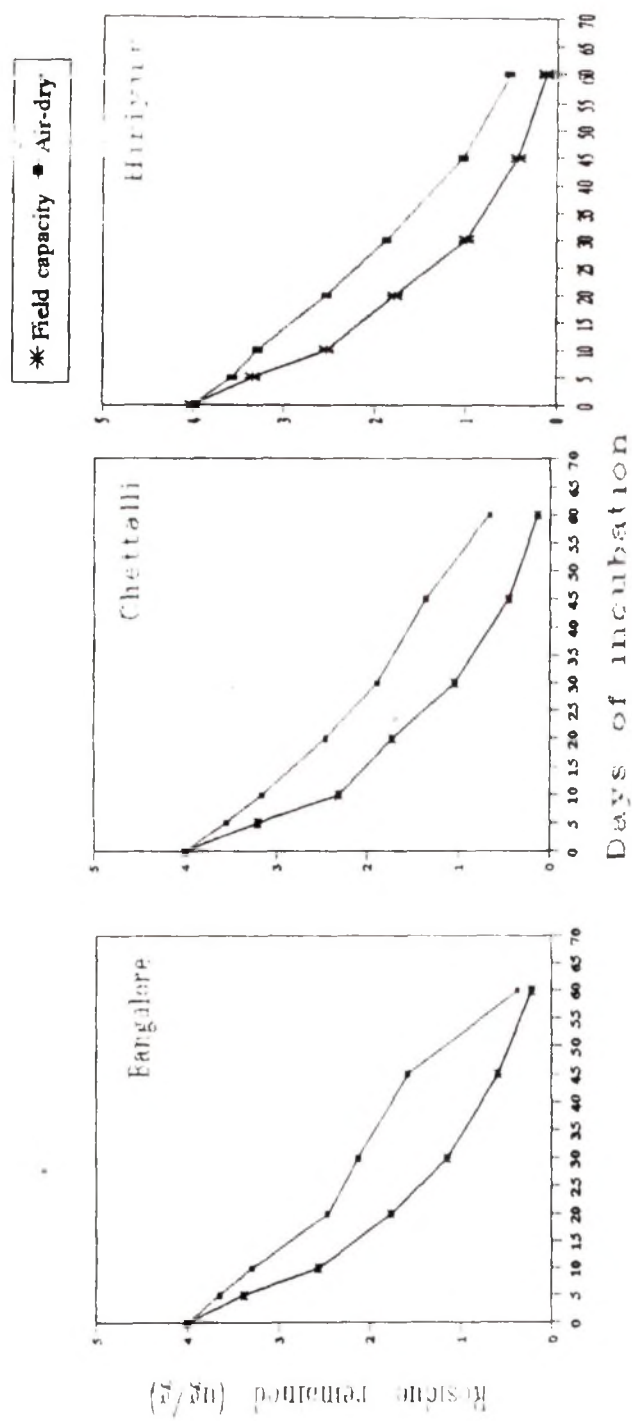


Figure 16. Persistence of DDT residues in soils under the influence of soil moisture

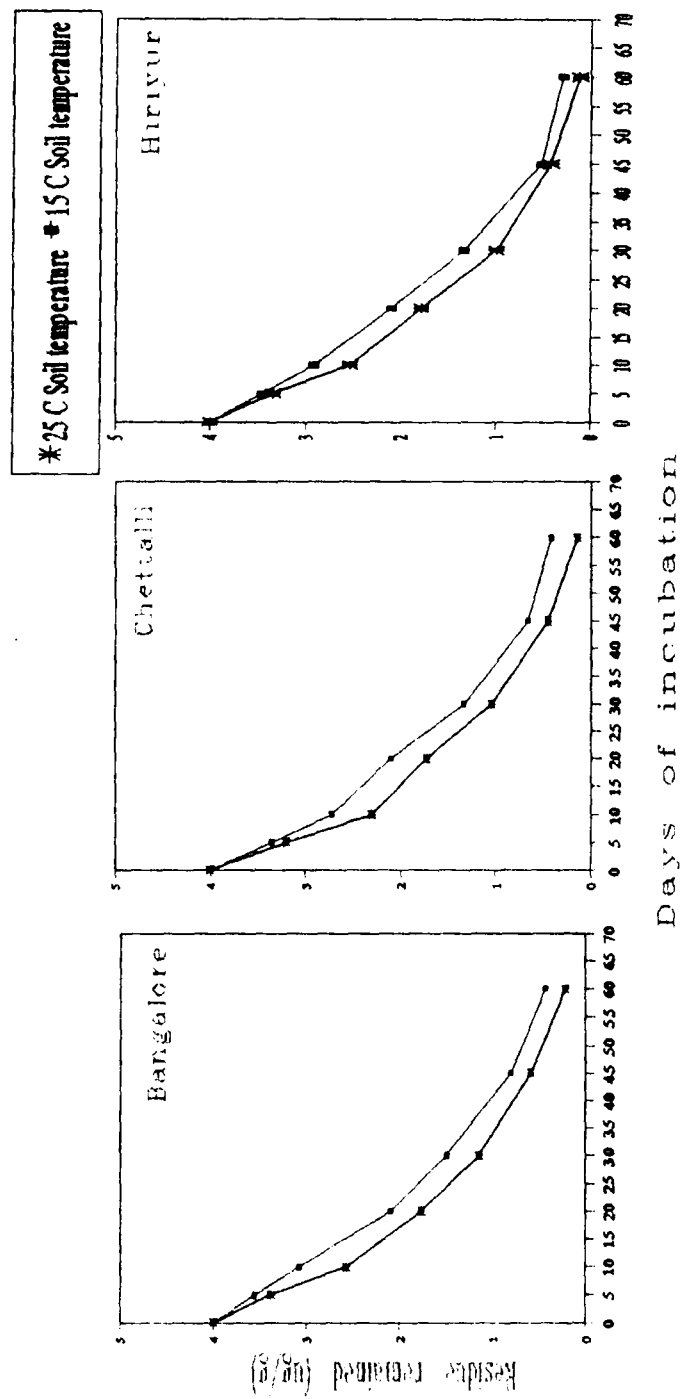


Figure 17. Persistence of ETU residues in soils under the influence of soil temperatures

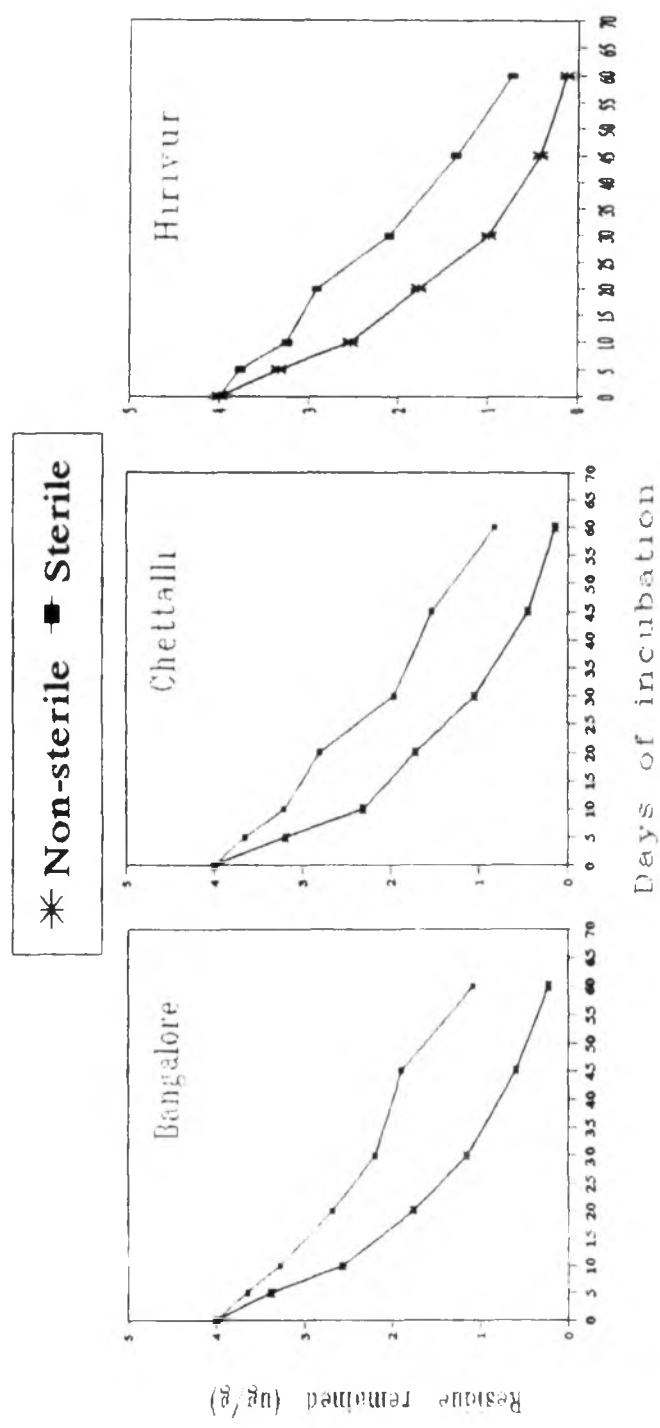


Figure 18 Persistence of ETU residues in soils under the influence of micro-organisms

level in Bangalore, Chettalli and Hiriur soils. The quantity of ETU retained by the soils maintained at field capacity in an unaltered form was in the decreasing order of Hiriur > Chettalli > Bangalore soil while as in case of soils under air dry conditions, the Chettalli soil was found to retain higher quantity of ETU than other two soils (Fig. 16).

The persistence of ETU was comparatively more at 15°C temperature than 25°C (Fig.17). The quantities of ETU persisted at the end of 10th and 60th day were 3.08 and 0.43 $\mu\text{g g}^{-1}$ in Bangalore soil, 2.73 and 0.42 $\mu\text{g g}^{-1}$ in Chettalli soil and 2.91 and 0.30 $\mu\text{g g}^{-1}$ in Hiriur soil under 15°C soil temperature. Whereas if 25°C temperature, the corresponding values were 2.57, 2.31 and 2.53 and 0.22, 0.14 and 0.12 $\mu\text{g g}^{-1}$ on 10th and 60th day in Bangalore, Chettalli and Hiriur soils. The persistence of ETU was comparatively more in sterile soils as compared to non-sterile soils (Fig. 18). In the sterile soils the residue levels of 3.65, 3.66 and 3.77 $\mu\text{g g}^{-1}$ on 5th day further reduced to 1.08, 0.83 and 0.73 $\mu\text{g g}^{-1}$ by 60th day in Bangalore, Chettalli and Hiriur soils. Similar values for non-sterile soils were 3.38, 3.20 and 3.34 $\mu\text{g g}^{-1}$ on 5th day and 0.22, 0.14 and 0.12 $\mu\text{g g}^{-1}$ on 60th day in the three soils respectively.

4.2.3.1 Degradation kinetics of ethylenethiourea in soils :

The degradation of ETU in different soil types studied followed the first-order reaction kinetics. The quantities of ETU that persisted at any time was dependent upon the initial concentration added and the specific reaction rate constant (K_{deg}) (Tables 20 and 21). The determination coefficient (R^2) values were greater than 0.972 indicated that the persistence data fitted to first order exponential equation.

The values of the initial concentration (C_0) estimated were higher than the applied rate except at the lower rate of application (2.0 $\mu\text{g g}^{-1}$) in Bangalore (1.97 $\mu\text{g g}^{-1}$) and Chettalli (1.797 $\mu\text{g g}^{-1}$) soils. The first order rate constants (K_{deg}) ranged from 55.07 $\times 10^{-3}$ to 66.97 $\times 10^{-3} \text{ day}^{-1}$ for 2.0 $\mu\text{g g}^{-1}$ level and 46.99 $\times 10^{-3}$ to 56.96 $\times 10^{-3} \text{ day}^{-1}$ for 4.0 $\mu\text{g g}^{-1}$ level of ETU in soils. The degradation rate constants were lower at higher rate of application (4.0 $\mu\text{g g}^{-1}$), air dry moisture, soil temperature maintained at 15°C and sterile soil conditions similar to metalaxyl and mancozeb in soils. The K_{deg} values for air dry moisture were almost 50 per cent less than field capacity soil moisture. Similar trend was also noticed in sterile soil

Table 20. Equations explaining the kinetics of ethylenethiourea residues degradation in different soils as affected by application rates and moisture regimes.

Soils	ETU added ($\mu\text{g g}^{-1}$)	Moisture regimes	Exponential equation	$K_{(\text{deg})}$ (10^{-3} day^{-1})	Half-life (days)		R^2
					$t_{1/2}$	$t_{1/2}(\text{mod})$	
Bangalore	2.0	Field capacity	$C = 1.917 e^{(-0.0562)t}$	56.18	12.34	11.58	0.994
	4.0	Field capacity	$C = 4.297 e^{(-0.0469)t}$	46.99	14.75	16.28	0.992
	4.0	Air dry	$C = 4.129 e^{(-0.0237)t}$	23.74	29.19	30.53	0.984
Chettalli	2.0	Field capacity	$C = 1.797 e^{(-0.0551)t}$	55.07	12.58	10.64	0.986
	4.0	Field capacity	$C = 4.386 e^{(-0.0537)t}$	53.70	12.90	14.62	0.984
	4.0	Air dry	$C = 4.220 e^{(-0.0285)t}$	28.48	24.34	26.22	0.978
Hiriyur	2.0	Field capacity	$C = 2.045 e^{(-0.0669)t}$	66.97	10.35	10.68	0.986
	4.0	Field capacity	$C = 4.666 e^{(-0.0569)t}$	56.96	12.17	14.87	0.982
	4.0	Air dry	$C = 4.484 e^{(-0.0337)t}$	33.67	20.58	23.98	0.980

$K_{(\text{deg})}$ = Rate of residue degradation

$t_{1/2}(\text{mod})$ = Half-life (modified)

R^2 = Determination co-efficient

Table 21. Equations explaining the kinetics of ethylenethiourea residues degradation in different soils as affected by temperatures and sterilization.

Soils	Temperature (°C)	Soil condition	Exponential equation	K(deg) (10 ⁻³ day ⁻¹)	Half-life (days)		R ²
					t _{1/2}	t _{1/2(mod)}	
Bangalore	15	Non-sterile	C = 4.325 e ^{(-0.0375)t}	37.51	18.48	20.56	0.996
	25	Non-sterile	C = 4.297 e ^{(-0.0469)t}	46.99	14.75	16.28	0.992
	25	Sterile	C = 4.064 e ^{(-0.0203)t}	20.29	34.15	34.94	0.972
Chettalli	15	Non-sterile	C = 4.117 e ^{(-0.0385)t}	38.51	17.99	18.75	0.994
	25	Non-sterile	C = 4.386 e ^{(-0.0537)t}	53.70	12.90	14.62	0.984
	25	Sterile	C = 4.227 e ^{(-0.0252)t}	25.19	27.51	29.71	0.968
Hiriyur	15	Non-sterile	C = 4.469 e ^{(-0.0448)t}	44.81	15.47	17.94	0.988
	25	Non-sterile	C = 4.666 e ^{(-0.0569)t}	56.96	12.17	14.87	0.982
	25	Sterile	C = 4.439 e ^{(-0.0278)t}	27.81	24.92	28.67	0.972

K(deg) = Rate of residue degradation

t_{1/2 (mod)} = Half-life (modified)

R² = Determination co-efficient

($20.29 \times 10^{-3} \text{ day}^{-1}$) condition and non-sterile soil condition ($37.51 \times 10^{-3} \text{ day}^{-1}$) for Bangalore soil. At lower temperature (15°C), the degradation rate constants were 37.51×10^{-3} , 38.51×10^{-3} and $44.81 \times 10^{-3} \text{ day}^{-1}$ for Bangalore, Chettalli and Hiriur soils respectively.

The conventional method of half-life calculation invariably results in under estimation of half-life values as compared to modified method (Tables 20 & 21). Accordingly the half-life of ETU in Bangalore soil were 11.58 and 16.28 days for 2.0 and $4.0 \mu\text{g g}^{-1}$ ETU application rates. Almost similar half-life were recorded for Chettalli and Hiriur soils.

The half-life ($t_{1/2}$) values were 30.53, 26.22 and 23.98 days at air-dry moisture and 16.28, 14.62 and 14.87 days at field capacity moisture in Bangalore, Chettalli and Hiriur soils respectively. In soil maintained at 25°C temperature, the half-life were shorter (16.28, 14.62 and 14.87 days) compared to lower temperature of 15°C (20.56, 18.75 and 17.94 days) in all the three soils. The higher half-life values were recorded in sterile soils as compared to non-sterile soil conditions on all the three soils.

4.3 Plant uptake and persistence of fungicide and/or metabolite by tomato and their persistence in soil

4.3.1 Plant uptake of fungicides

4.3.1.1 Metalaxyl : Data on the uptake and persistence of metalaxyl by tomato plants grown in Bangalore soil are presented in Table 22 and Figure 19. The soil applied metalaxyl-mancozeb (Ridomil MZ 72 WP) in pots at 112.5 and 225.0 $\mu\text{g a.i. per pot}$ was found to be absorbed continuously by tomato plants up to 15 days irrespective of the dose. The buildup of plant absorbed residues were found declining after attaining the maximum concentration. The progressive absorption of metalaxyl residues buildup in tomato plants from $3.03 \mu\text{g g}^{-1}$ initial concentration after 2nd day to the maximum concentration of $6.11 \mu\text{g g}^{-1}$ after 15 days following soil application of 112.5 mg per pot. The absorption of metalaxyl residues in tomato were also higher from the higher level of metalaxyl-mancozeb ($225.0 \text{ mg a.i. pot}^{-1}$) application.

Table 22. Plant uptake and persistence of metalaxyl and ethylenethiourea (ETU) in tomato.

Treatment conc. (mg Ridomil-MZ a.i pot ⁻¹)	Fungicide/ metabolite	Residues (ug g ⁻¹) recovered at different intervals (days)							
		2	5	10	15	20	25	30	40
112.5 (12.5 + 100)	Metalaxyl	3.03	4.40	5.84	6.11	4.69	2.70	1.06	0.39
	ETU	0.12	0.26	1.11	1.09	1.45	1.09	(0.26) 0.44 (ND)	(0.19) ND (ND)
225.0 (25.0 + 200)	Metalaxyl	5.23	8.35	9.31	10.38	6.21	5.05	1.85	0.62
	ETU	0.15	0.42	1.79	1.78	2.47	2.04	(0.48) 0.81 (0.06)	(0.36) ND (ND)

Note: Figures in the parenthesis denote residues in fruits

ND = not detected

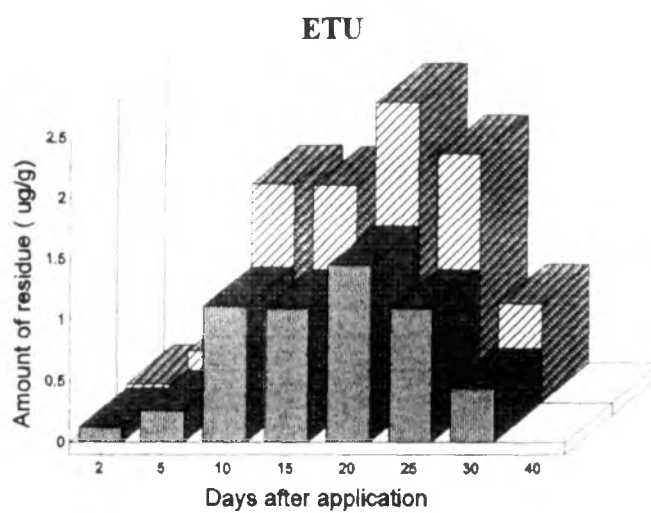
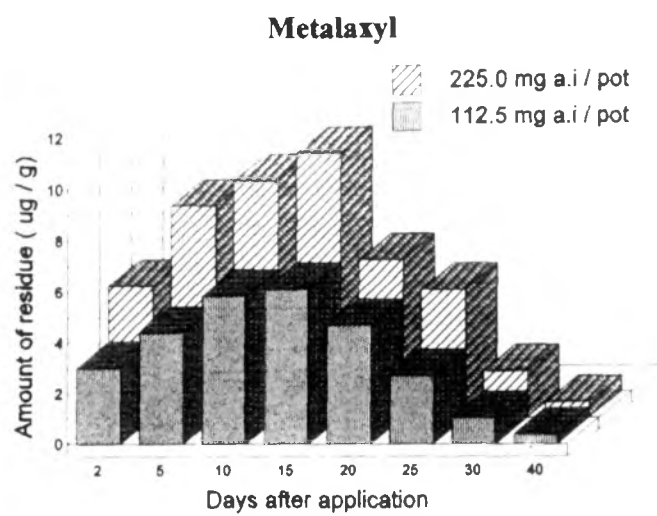


Figure 19. Uptake of fungicide residues by tomato after the application of Metalaxyl- MZ to Soil

The plant absorbed metalaxyl from the two application rates finally moved to the final sink in fruits. Accordingly, the tomato fruits harvested after 30 and 40 days of metalaxyl-mancozeb application were found to contain 0.26 and 0.19 $\mu\text{g g}^{-1}$ residues from lower dose and 0.48 and 0.36 $\mu\text{g g}^{-1}$ from higher dose respectively.

4.3.1.2 Ethylenethiourea : The uptake of ETU resulting from metabolism of mancozeb in soil, by tomato crop was also noticed from the metalaxyl-mancozeb treated pots. The mancozeb metabolises to ETU in soil which is easily absorbed by tomato plants. It was observed that absorption of ETU increased as the time lapsed (up to 20 days) and thereafter the concentration of ETU was reduced. Even within 10th days of application of metalaxyl-mancozeb, the appreciable quantities of ETU were 1.11 and 1.79 $\mu\text{g g}^{-1}$ fresh weight of tomato from 112.5 and 225.0 mg pot^{-1} rates respectively. In fruits trace of ETU residues (0.06 $\mu\text{g g}^{-1}$) were found till 30 days and they could not be detected after 40 days of application.

4.3.2 Retention of fungicides residues in soil under tomato crop cover : The results of the persistence of fungicides residues in soils under tomato crop cover are given in Tables 23 and 24 (Figure 20).

4.3.2.1 Metalaxyl : It was observed that as the time progressed, the persistence of metalaxyl decreased drastically. The initial concentration of metalaxyl added to soil were 12.5 and 25.0 mg pot^{-1} corresponded to 2.5 and 5.0 $\mu\text{g g}^{-1}$. Within 20 days of application of metalaxyl-mancozeb, the residues of metalaxyl reduced to 0.26 and 0.83 $\mu\text{g g}^{-1}$ from the initial concentrations of 2.5 and 5.0 $\mu\text{g g}^{-1}$ respectively. The residues were below detectable limit/from the lower application rates however 0.33 $\mu\text{g g}^{-1}$ metalaxyl residues were detected from the higher dose.

The data on the first order exponential equation and their kinetics of persistence in soils are presented in Table 24. The K_{deg} values were 106.30×10^{-3} and $72.45 \times 10^{-3} \text{ day}^{-1}$ from the initial residue levels of 2.5 and 5.0 $\mu\text{g g}^{-1}$ metalaxyl. The time required for 50 per cent dissipation was only 6.69 days in lower dose and 8.73 days for higher dose.

4.3.2.2 Mancozeb : Application of 112.5 $\text{mg metalaxyl -mancozeb pot}^{-1}$ resulted in the build up of 20.0 $\mu\text{g g}^{-1}$ on zero day which on degradation reduced to 13.01 $\mu\text{g g}^{-1}$

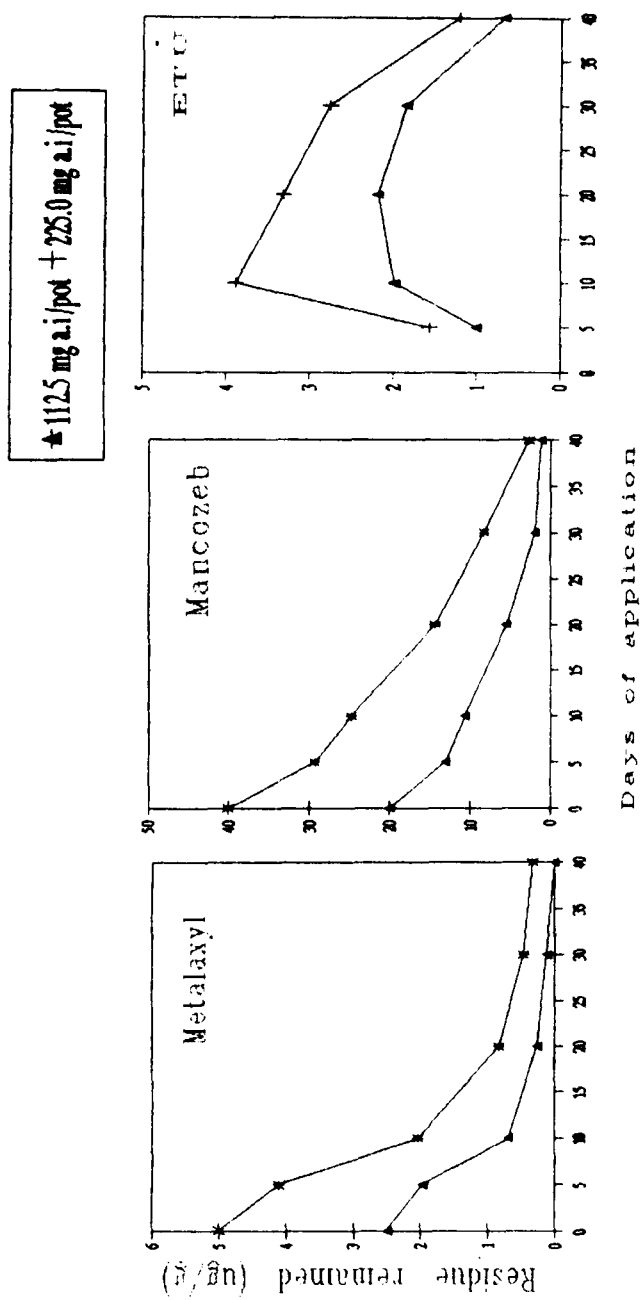


Figure 20. Persistence of fungicide residues in Bangalore soil under tomato crop cover

Table 23. Persistence of fungicides and its metabolite residues in Bangalore soil under tomato crop cover.

Treatment conc. (mg Ridomil-MZ a.i pot ⁻¹)	Fungicide/ metabolite	Residues (ug g ⁻¹) recovered at different intervals (days)						
		0	5	10	20	30	40	
112.5 (12.5 + 100)	Metalaxyl	2.50	1.97	0.69	0.26	0.12	ND	
	Mancozeb	20.00	13.01	10.65	5.47	1.92	1.15	
	ETU	ND	1.00	1.99	2.18	1.84	0.68	
112.5 (12.5 + 100)	Metalaxyl	5.00	4.11	2.04	0.83	0.47	0.33	
	Mancozeb	40.00	29.24	24.77	14.32	8.23	2.67	
	ETU	ND	1.56	3.89	3.32	2.76	1.22	

ND = Not detected

Table 24. Equations explaining the kinetics of fungicide residues degradation in Bangalore soil under tomato crop cover.

Treatment conc. (mg Ridomil-MZ a.i pot ⁻¹)	Fungicides	Exponential equation	K _(deg) (10 ⁻³ day ⁻¹)	Half-life (days) ----- t _{1/2} t _{1/2} (mod)	R ²
112.5(12.5 + 100)	Metalaxyl	C = 2.545 e ^{(-0.1063)t}	106.30	6.52 6.69	0.972
	Mancozeb	C = 20.368 e ^{(-0.0730)t}	73.00	9.49 9.74	0.990
225.0(25.0 + 200)	Metalaxyl	C = 4.706 e ^{(-0.0724)t}	72.45	9.57 8.73	0.964
	Mancozeb	C = 44.118 e ^{(-0.0638)t}	63.77	10.87 12.41	0.966

K_(deg) = Rate of residue degradation

t_{1/2} (mod) = Half-life (modified)

R² = Determination co-efficient

on 5th day and $1.15 \mu\text{g g}^{-1}$ on 40th day while at the application rate of $225.0 \text{ mg metalaxyl-mancozeb pot}^{-1}$ the residues were 40.0, 29.24 and $2.67 \mu\text{g g}^{-1}$ on 0, 5 and 40th day respectively. The determination coefficient (R^2) values were greater than 0.966 reflecting that the persistence data fitted to first order exponential equation.

The first order rate constants (K_{deg}) were higher compared to laboratory incubation studies. The rate constant values were higher from lower level of application rate, however only slight variation of half-life (9.74 to 12.41 days) were observed in the residue degradation from the two treatment concentration.

4.3.2.3 Ethylenethiourea : The residues of ETU resulting from metabolism of mancozeb were 1.00 and $0.68 \mu\text{g g}^{-1}$ from $112.5 \text{ mg pot}^{-1}$ and 1.56 and $1.22 \mu\text{g g}^{-1}$ from $225.0 \text{ mg pot}^{-1}$ after 5th and 40th day respectively. The maximum concentrations of $2.18 \mu\text{g g}^{-1}$ ETU was recovered after 20 days from $112.5 \text{ mg pot}^{-1}$ treatment whereas $3.89 \mu\text{g g}^{-1}$ ETU residues were recovered after 10 days only from $225.0 \text{ mg pot}^{-1}$ treatment.

4.4 Persistence of fungicide residues in/on tomato and soil

4.4.1 Persistence of fungicide residues in/on tomato: Data on the persistence pattern of fungicide residues in tomato foliage and fruits following foliar application of metalaxyl-mancozeb (Ridomil MZ 72 WP) at 0.2 and 0.4 per cent spray concentrations in two spray at flowering stage and at 50 per cent fruiting are presented in Tables 25 and 26 and Figures 21 and 22.

4.4.1.1 Metalaxyl : The initial deposits of the metalaxyl from the second spray were 5.83 and $12.88 \mu\text{g g}^{-1}$ in tomato foliage and 1.27 and $1.88 \mu\text{g g}^{-1}$ in tomato fruits from the two applications respectively. The residues of the metalaxyl over the period decreased exponentially and the concentrations were reduced to 0.18 and $0.26 \mu\text{g g}^{-1}$ in foliage and 0.10 and $0.22 \mu\text{g g}^{-1}$ in fruits after 30th day of foliar spray.

The persistence values were fitted to first order exponential equation (Table 27) and the coefficient of determination (R^2) was more than 0.935 that indicated the best fit between $\ln$ (residues) verses time in days. The K_{deg} values were 150.85×10^{-3} and $186.49 \times 10^{-3} \text{ day}^{-1}$ for tomato foliage and 85.07×10^{-3} and $73.04 \times 10^{-3} \text{ day}^{-1}$

Table 25. Persistence of the residues of fungicides and metabolite in/on tomato foliage
under field conditions.

Foliar spray conc. (% Ridomil-MZ)	Fungicide/ metabolite	Residues ($\mu\text{g g}^{-1}$) recovered at different intervals (days)						
		0	5	10	15	20	30	40
0.2	Metalaxyl	5.83	2.52	1.01	0.64	0.39	0.18	ND
	Mancozeb	40.40	33.37	22.84	20.60	16.18	9.76	2.51
	ETU	2.24	1.36	0.93	1.43	0.92	0.47	0.25
0.4	Metalaxyl	12.88	4.78	1.17	0.92	0.66	0.26	0.10
	Mancozeb	99.85	89.52	63.12	57.67	48.06	16.65	4.43
	ETU	3.06	2.42	1.85	2.12	1.46	1.18	0.34

ND = Not detected

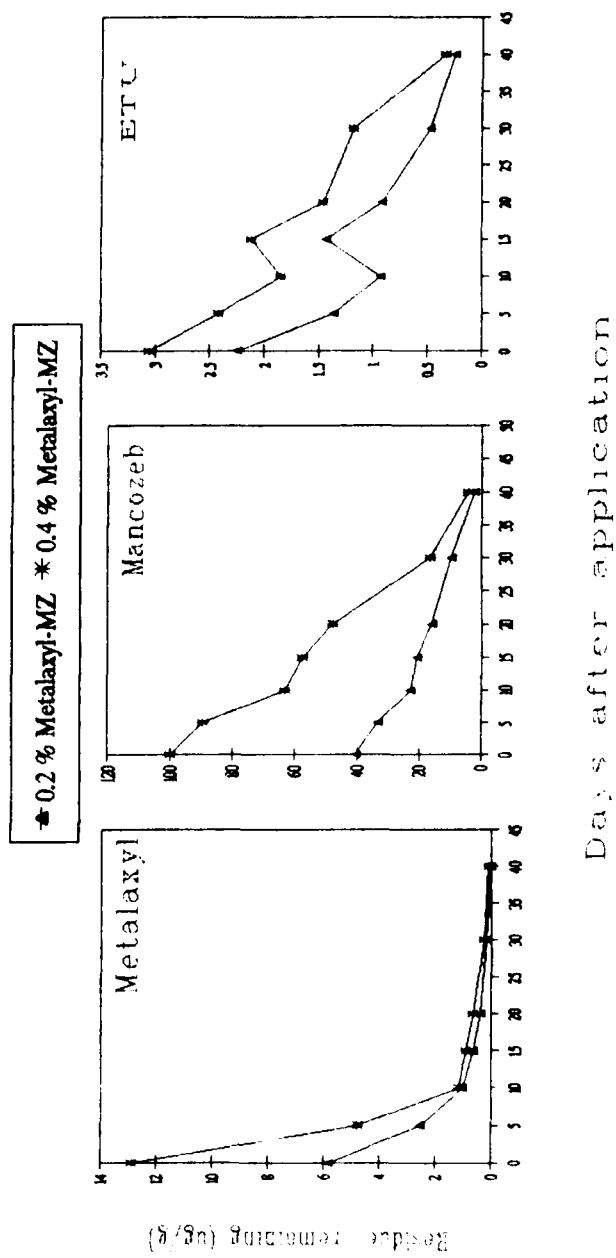


Figure 21. Persistence of fungicide residues on tomato foliage under field conditions

Table 26. Persistence of the residues of fungicides and metabolite in/on tomato fruits
under field conditions.

Foliar spray conc. (% Ridomil-MZ)	Fungicide/ metabolite	Residues ($\mu\text{g g}^{-1}$) recovered at different intervals (days)						
		0	5	10	15	20	30	40
0.2	Metalaxyl	1.27	0.83	0.48	0.37	0.21	0.10	ND
	Mancozeb	10.28	5.29	2.66	1.42	1.21	ND	ND
	ETU	2.15	1.10	0.64	0.63	0.28	0.12	ND
0.4	Metalaxyl	1.88	1.45	0.71	0.53	0.41	0.22	ND
	Mancozeb	15.49	9.98	5.96	3.03	2.11	0.65	ND
	ETU	2.85	2.03	1.28	0.91	0.53	0.22	ND

ND - Not detected

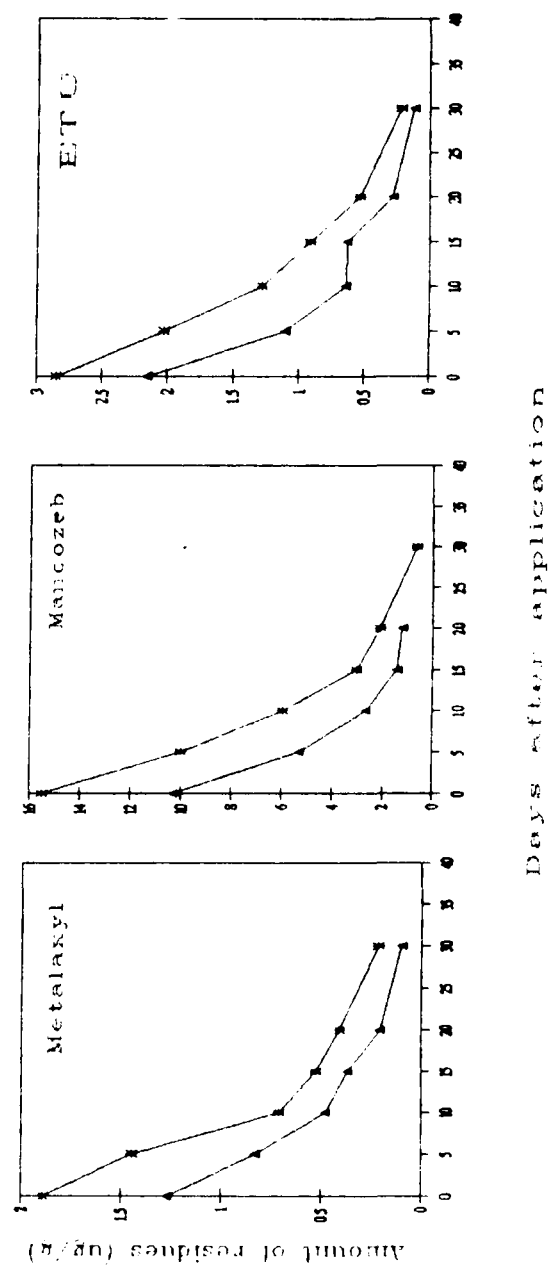


Figure 22 Dissipation of fungicide residues in tomato fruits under field conditions

Table 27. Equations explaining the kinetics of fungicide residues dissipation and safety constants
in/or tomato foliage and fruits.

Parts of tomato	Foliar spary (% Ridomil-MZ)	Fungicides	Exponential equation	$K_{(deg)}$ (10^{-3} day $^{-1}$)	Half-life (days)		R^2	Waiting Period (days)
				$t_{1/2}$	$t_{1/2}$	$t_{1/2(mod)}$		
Foliage	0.2	Metalaxyl	$C = 5.441 e^{(-0.1508)t}$	150.85	4.59	4.14	0.982	15.83
		Mancozeb	$C = 47.626 e^{(-0.0642)t}$	64.17	10.80	13.37	0.933	43.08
	0.4	Metalaxyl	$C = 11.555 e^{(-0.1865)t}$	186.49	3.72	3.13	0.943	16.83
		Mancozeb	$C = 139.777 e^{(-0.0759)t}$	75.91	9.13	13.56	0.922	50.61
Fruits	0.2	Metalaxyl	$C = 1.235 e^{(-0.0851)t}$	85.07	8.15	7.82	0.994	10.63
		Mancozeb	$C = 9.226 e^{(-0.1119)t}$	111.89	6.19	5.23	0.966	10.04
	0.4	Metalaxyl	$C = 1.781 e^{(-0.0730)t}$	73.04	9.49	8.75	0.972	17.39
		Mancozeb	$C = 16.322 e^{(-0.1063)t}$	106.31	6.52	6.95	0.996	15.93

$K_{(deg)}$ = Rate of residue degradation

$t_{1/2(mod)}$ = Half-life (modified)

R^2 = Determination co-efficient

Maximum residue limit (MRL): Metalaxyl = 0.5 ug g $^{-1}$, Mancozeb = 3.0 ug g $^{-1}$

for fruits at 0.2 and 0.4 per cent treatment concentrations respectively. The half-life values of residues degradation ranged from 3.13 to 4.14 days for foliage and 7.82 to 8.75 days for tomato fruits.

The waiting periods were calculated using first order equation and the re-arrangement made as

$$T_{(tol)} = [\{\ln(C_0) / MRL\} / K],$$

where $T_{(tol)}$ - waiting period (days), $\ln$ - natural logarithm, C_0 - initial concentration ($\mu\text{g g}^{-1}$) and MRL - maximum residue limit ($\mu\text{g g}^{-1}$) established by regulatory agencies like FAO, WHO, EPA and others. The maximum residue limit of $0.5 \mu\text{g g}^{-1}$ has been assigned for metalaxyl on tomato. The waiting periods worked out on the basis of persistence pattern of residues and the prescribed MRL ranged from 10.63 and 17.39 days for fruits and 15.83 and 16.23 days for foliage respectively when tomato crop was sprayed at 0.2 and 0.4 per cent metalaxyl-mancozeb spray.

4.4.1.2 Mancozeb : The residues of mancozeb resulting from the foliar applications of metalaxyl-mancozeb (Ridomil MZ 72 WP) were found to persist in higher quantities and for longer period on tomato foliage as compared to tomato fruits. Accordingly, the initial deposits of 40.40 and 99.85 $\mu\text{g g}^{-1}$ on foliage from 0.2 and 0.4 per cent spray concentrations persisted for 40 days, while initial deposits of 10.28 and 15.49 $\mu\text{g g}^{-1}$ on tomato fruits persisted for 20 to 30 days from the two application rates respectively. Mancozeb residues on foliage dissipated at the half-life of 13.37 and 13.56 days reflecting no variation in residue dissipation due to treatment concentrations. Similarly no variation in dissipation pattern of residues was noticed in fruits from the two application rates where half-life values ranged between 5.23 and 6.95 days. The dissipation pattern based on half-life values for foliage and fruits reflect the prolonged persistence of residues on foliage as compared to fruits. Accordingly, the K_{deg} values were higher in fruits (106.31×10^{-3} to $111.89 \times 10^{-3} \text{ day}^{-1}$) than foliage (64.17×10^{-3} to $75.91 \times 10^{-3} \text{ day}^{-1}$).

The dissipation pattern of mancozeb residues with respect to the prescribed MRL of $3.0 \mu\text{g g}^{-1}$ on tomato helped in working out the waiting periods of 43.08 to 50.61 days for foliage and 10.04 to 15.93 days for fruits, that may be observed to escape toxicity hazards.

4.4.1.3 Ethylenethiourea : The metabolism of mancozeb to ETU in plant system from the first application made at 50 per cent flowering stage and repeat application at fruiting of tomato resulted in the maximum recovery of 2.24 and 3.06 $\mu\text{g g}^{-1}$ ETU in foliage immediately after the second spray from the two respective spray concentrations (Table 25 and Figure 21). These residue concentrations of ETU were found to persist in foliage for over 40 days to the extent of 0.25 and 0.34 $\mu\text{g g}^{-1}$. The persistence pattern of ETU residues in fruits is presented in Table 26. Like foliage, fruits also recorded highest buildup of ETU residues immediately after second spray to the extent of 2.15 and 2.85 $\mu\text{g g}^{-1}$ from the lower and higher spray application concentration. The persistence of ETU residues in fruits was recorded up to 30 days to the extent of 0.12 and 0.22 $\mu\text{g g}^{-1}$ respectively. The first harvest of mature fruits coincided with the last detectable residue after 30 days of spraying. It was interesting to note that till the first harvest ETU residues were at the bioconcentration levels with respect to the prescribed MRL of 0.05 $\mu\text{g g}^{-1}$ (FAO/WHO, 1993). However, the later harvests did not contain any detectable ETU residues.

4.4.2 Persistence of fungicides residues in soil: Persistence pattern of fungicide residues in soils resulting from the foliar spray applications to tomato crop at 0.2 and 0.4 per cent metalaxyl-mancozeb spray concentrations in a field experiment are presented in Table 28 and Figure 23.

4.4.2.1 Metalaxyl : The foliar spray applications of metalaxyl-mancozeb also made their way to soil and resulted in surface deposits of 0.95 and 1.58 $\mu\text{g g}^{-1}$ following the second spray application at 0.2 and 0.4 per cent concentrations. The residues were found to persist for over 30 days following the progressive dissipation of residues at the half-life of 8.77 and 9.68 days (Table 29). The exponential equation was predicted better and the coefficient of determination (R^2) was more than 0.972 in all the treatment combinations. The initial concentration (C_0) derived from the equation almost coincides with values observed on zero day. So there was no difference in the half-life values calculated by both the methods. The rate of degradation constants were 75.64×10^{-3} and $75.67 \times 10^{-3} \text{ day}^{-1}$ for 0.2 and 0.4 per cent spray. The K_{deg} values were higher in field experiment compared to laboratory incubation studies.

4.4.2.2 Mancozeb and ETU : The initial buildup of 2.76 and 4.31 $\mu\text{g g}^{-1}$ mancozeb deposits on soil surface under the crop cover of tomato were found to persist for

Table 28. Persistence of the residues of fungicides and metabolite in soil following
foliar spray of Ridomil-MZ on tomato crop.

Foliar spray conc. (% Ridomil-MZ)	Fungicide/ metabolite	Residues ($\mu\text{g g}^{-1}$) recovered at different intervals (days)				
		0	5	10	20	30
0.2	Metalaxyl	0.95	0.66	0.36	0.24	0.09
	Mancozeb	2.76	1.68	0.92	0.22	ND
	ETU	0.45	0.88	0.58	0.23	ND
0.4	Metalaxyl	1.58	1.20	0.67	0.46	0.15
	Mancozeb	4.31	2.53	1.38	0.60	ND
	ETU	0.75	1.46	1.10	0.41	ND

ND = Not detected

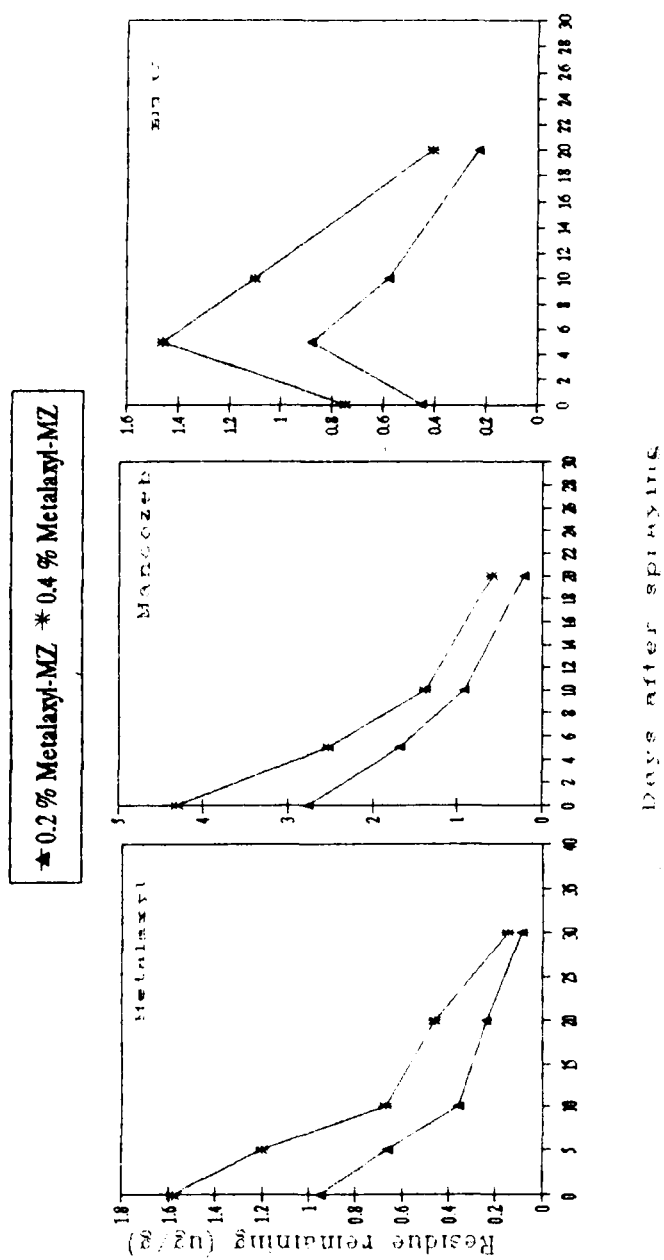


Figure 23 Persistence of fungicide residues in soil after spraying of Metalaxyl-MZ on tomato crop

Table 29. Equations explaining the kinetics of fungicide residues degradation in soil following foliar spray of Ridomil-MZ on tomato crop.

Foliar spray conc. Fungicides (%Ridomil-MZ)	Exponential equation	$K_{(deg)}$ (10^{-3} day $^{-1}$)	Half-life (days)		R^2
			$t_{1/2}$	$t_{1/2(mod)}$	
0.2	Metalaxyl $C = 0.922 e^{(-0.0756)t}$	75.64	9.16	8.77	0.978
	Mancozeb $C = 3.012 e^{(-0.1279)t}$	127.85	5.42	6.10	0.994
0.4	Metalaxyl $C = 1.644 e^{(-0.0757)t}$	75.67	9.16	9.68	0.972
	Mancozeb $C = 4.114 e^{(-0.0988)t}$	98.78	7.02	6.55	0.992

$K_{(deg)}$ = Rate of residue degradation

$t_{1/2(mod)}$ = Half-life (modified)

R^2 = Determination co-efficient

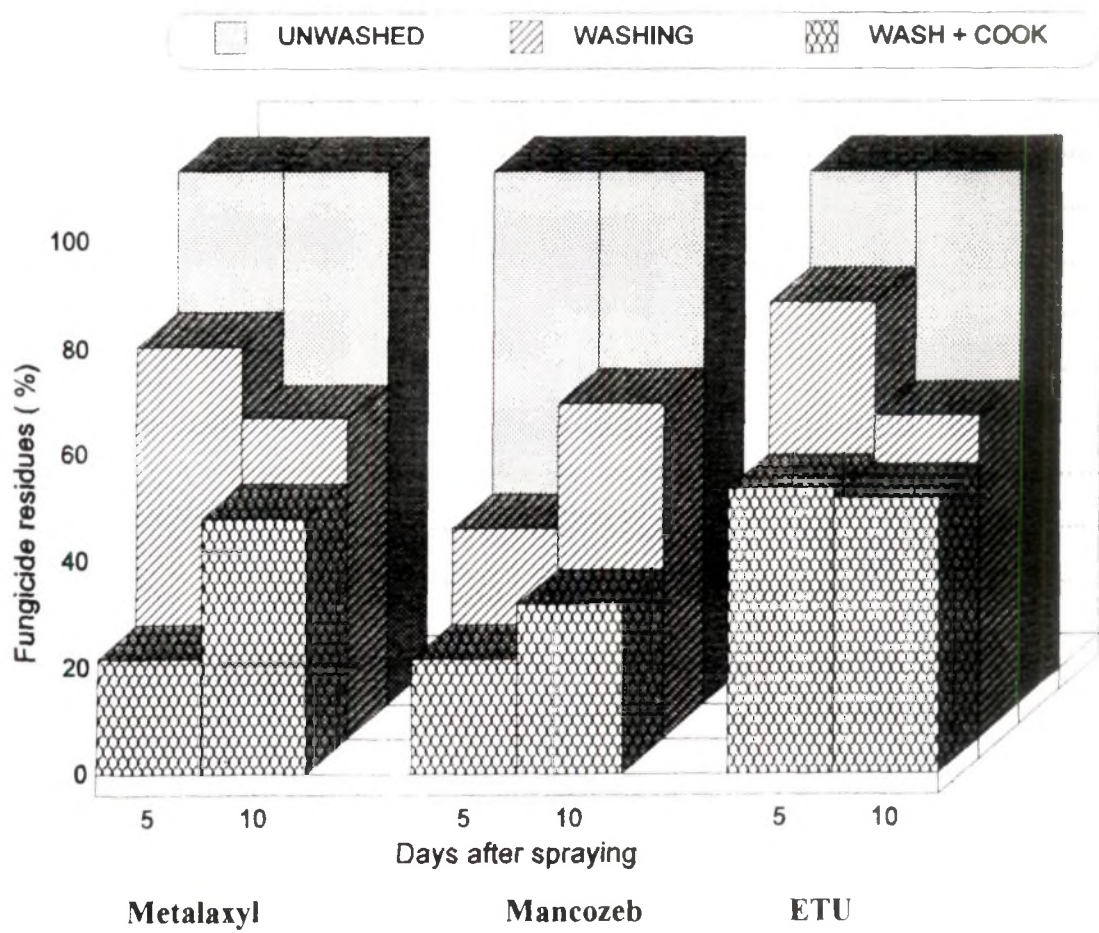


Figure 24. Decontamination of fungicide residues from tomato fruits

20 days only (Table 28). The residues degraded at the half-life of 6.10 and 6.55 days from the two application concentrations. The calculation of residue half-life by conventional method was found to be higher for higher application rate and lower for lower application rate. The rate of residue degradation and the persistence pattern of mancozeb residues in soil reflect rapid loss from soil under crop cover in field conditions. The rate constants for mancozeb degradation in soil were 127.85×10^{-3} and $98.70 \times 10^{-3} \text{ day}^{-1}$ for the two application rates.

The metabolism of mancozeb from the two treatment concentrations effected the buildup of ETU at the maximum concentrations of 0.88 and $1.46 \mu\text{g g}^{-1}$ after 5 days of second spraying. The residues of ETU prolonged in soil for 20 days only to the extent of 0.23 and $0.41 \mu\text{g g}^{-1}$.

4.5 Decontamination of fungicide residues from tomato fruits

The buildup of fungicide residues on tomato fruits from repeated foliar application may affect the persistence of residues in bioconcentration levels on surface as well as inside the tissues and therefore, the fruits from such treatment were subjected to decontamination through washing followed by cooking to find their effect on residue loss. The results are detailed in Table 30 and Figure 24.

4.5.1 Metalaxyl : Residue decontamination processes like washing of residue loaded tomato fruits in water and washing followed by cooking under pressure were found to dislodge residues from fruit surface as well as residues moved inside fruit at varying levels of decontamination. A residue load of $0.83 \mu\text{g g}^{-1}$ in tomato from 0.2 per cent spray on 5th day was found to be reduced to 0.62 and $0.18 \mu\text{g g}^{-1}$ from the two processes whereas after 10 days the residues dislodged to the extent of 0.29 and $0.23 \mu\text{g g}^{-1}$ from the buildup of $0.48 \mu\text{g g}^{-1}$. A similar decontamination effect could be observed on tomato fruits from the higher dose of spray application.

4.5.2 Mancozeb : The effect of washing and washing followed by cooking of tomato fruits contaminated with mancozeb residues was predominantly far superior as per the extent of reduction of residues from $5.29 \mu\text{g g}^{-1}$ on 5th day resulting from 0.2 per cent spray application to 2.09 and $1.14 \mu\text{g g}^{-1}$ levels as well as after 10 days old residues of $2.26 \mu\text{g g}^{-1}$ to 1.43 and $0.72 \mu\text{g g}^{-1}$ levels. The extent of decontamination was slightly higher on the residues resulting from higher application concentration.

Table 30. Decontamination of fungicide residues from tomato fruits.

Foliar spray (% Ridomil-MZ)	Decontamination Processes	Residues recovered ($\mu\text{g g}^{-1}$) after treatment in intervals days)				
		5		10		
		Metalaxyl	Mancozeb	ETU	Metalaxyl	Mancozeb ETU
0.2	Unwashed	0.83	5.29	1.10	0.48	2.26 0.64
	Washing	0.62	2.09	0.92	0.29	1.43 0.39
	Washing+cooking	0.18	1.14	0.61	0.23	0.72 0.33
0.4	Unwashed	1.45	9.98	2.03	0.71	5.96 1.28
	Washing	1.03	5.96	1.23	0.36	2.27 0.68
	Washing+cooking	0.31	2.53	1.05	0.28	1.38 0.52

4.5.3 Ethylenethiourea : Washing of the ethylenethiourea loaded residues could remove only insignificant levels of residues persisting on fruit skin from 1.10 to 0.92 $\mu\text{g g}^{-1}$ whereas the washing followed by cooking processes dislodged the residues to 0.61 $\mu\text{g g}^{-1}$ level for the residues resulting after 5 days from application rate of 0.2 per cent spray concentration. The 10 day-old residues of 0.64 $\mu\text{g g}^{-1}$ were accordingly reduced to 0.39 and 0.33 $\mu\text{g g}^{-1}$ level from the two decontamination processes respectively. A little higher reduction was noticed on the residues resulting from higher application rate of 0.4 per cent spray application.

DISCUSSION

V. DISCUSSION

The salient features of the results on "Behaviour of metalaxyl and mancozeb residues in soils and tomato" presented in the previous chapter are discussed in this chapter in the light of literature available.

5.1 Adsorption of metalaxyl and ethylenethiourea in soils

The biological efficacy of pesticides applied to soil is largely governed by the process of adsorption. Adsorption not only affects the movement of added compounds, but also regulates their availability to pests as well as their decomposition.

5.1.1 Metalaxyl : The data on adsorption of metalaxyl on soils are presented in Table 6 and Figure 5. The metalaxyl adsorption increased with increasing quantity of metalaxyl added, but the percentage adsorption decreased at higher levels of application due to progressive saturation of adsorption sites with increasing concentration. Among the soils, the Hiriya soil adsorbed larger quantities of the metalaxyl than Chettalli and Bangalore soils.

The Freundlich equation provided a good fit ($R^2 = >0.978$) to adsorption of metalaxyl in all the soils (Table 7). The Freundlich constant K values ranged from 5.870 to 7.281 ml g^{-1} in different soils. The K values followed the order Hiriya (7.281 ml g^{-1}) > Chettalli (6.511 ml g^{-1}) > Bangalore (5.870 ml g^{-1}) soils which is also the decreasing order of clay content in the soils. The sorption coefficient K_{oc} is commonly a measure of pesticide sorption to soil organic carbon. The absolute quantity of metalaxyl adsorbed by unit weight of organic carbon content of soils ranged from 362 to 2275. The absolute quantity of metalaxyl adsorbed and K_{oc} values are maximum in Hiriya soil. In Chettalli soil the adsorption coefficient (K) is maximum compared to Bangalore soil, but the K_{oc} value is lower (362) than Bangalore (1174) suggesting strong organic matter-clay interaction in the soils.

The findings in present investigation are in agreement with Sharom and Edgington (1982) where K values from Freundlich's adsorption isotherms of Ontario soils were 0.45, 0.89, 1.25 and 31.62 nanomoles per gram for Fox sandy loam, Honeywood loam, Guelph loam and Bradford muck respectively. Similar observations

were also made by Bailey and Coffey (1984) for metalaxyl adsorption and K values ranging from 0.20 to 0.60 nanomoles per gram of soil.

The values of $1/n$ (energy of adsorption) were less than 1.0 and indicated a non-linear relationship between the fungicide concentration and adsorption and can be characterised as L-type of adsorption isotherms. The L-shaped isotherm signify that soil has a high affinity for the pesticides. The adsorption occurs in a planar configuration and bonding is multi functional. The Gibb's free energy (dG^0) values are also in line with the order of adsorption of metalaxyl in the soils. The free energies for adsorption of pesticides are always negative which indicated that sorption is exothermic in soil-pesticide-water system.

The adsorption of metalaxyl and their kinetics of measurements (K and dG^0) in different soils decreased in the order of Hiriur > Chettalli > Bangalore, which is also the order of decreasing clay content in the soils. Hence, the higher adsorption of metalaxyl in Hiriur soil is mainly attributed to higher clay content (46.30 %), cation exchange capacity [$41.38 \text{ c mol (p}^+) \text{ kg}^{-1}$] and surface area than other soils. In Chettalli soil, adsorption of metalaxyl was found to be higher than Bangalore soil due to its higher clay content, organic carbon and cation exchange capacity (Table 2).

5.1.2 Ethylenethiourea : The data presented in Table 8 indicates that adsorption of ETU increased with increasing initial concentration and the per cent adsorption decreases with increasing concentration. All the three soils showed low adsorption of ETU when compared to metalaxyl. This may be due to higher solubility of ETU in water (20 g/lit). The ETU adsorption in soil was found to be in the order of Chettalli > Bangalore > Hiriur. The sorption data of ETU can be better explained by Freundlich equation as in case of metalaxyl. The kinetics of adsorption of ETU (Table 9), indicated the order of decreasing values of K in Chettalli > Bangalore > Hiriur soils. The Freundlich constant K values for ETU are less than observed for metalaxyl in these soils and may be attributed to very low adsorption of ETU than metalaxyl. The K_{oc} values were low in Chettalli (168) than Bangalore (461) and Hiriur (581) soils. The Gibb's free energy (dG^0) values also followed as in case of Freundlich constant (K). The adsorption isotherms of ETU can be classified as L-type, since the energy of adsorption ($1/n$) values were less than unity in all the soils. The $1/n$ values of ETU were higher than metalaxyl adsorption in soils.

The Freundlich equations for ETU are explained better ($R^2 = > 0.94$) in all the soils. The adsorption of ETU in the soils, Freundlich constant (K), free energy (dG°) are higher and K_{oc} is lower in Chettalli soil followed by Bangalore and Hiriya soils. Perhaps, this trend could be mainly attributed to organic carbon content and not related to texture of the soils. In Chettalli soil, the adsorption of ETU and its parameters are high because of the high organic carbon (1.8 %) in the soils. The results of this study confirms the finding of Miles and Doerge (1991) where sorption coefficient (K_d) ranged from 0.03 to 0.12 for different soils and clay minerals and concluded that the adsorption of ETU was mainly due to clay minerals.

5.2 Degradation of metalaxyl, mancozeb and ETU in soils

Degradation of fungicides added to the soil is mediated purely as a result of volatilization from soil surface, chemical hydrolysis and microbiological actions. The rate of degradation is dependent upon the soil type, adsorption-desorption equilibrium, moisture content, temperature, rate of application and nature of the compound (Bailey and White, 1970).

5.2.1 Metalaxyl : The persistence and degradation of metalaxyl was studied in Bangalore, Chettalli and Hiriya soils as influenced by application rates, moisture, temperature and soil biological activity. Data on the persistence (Tables 10 and 11) and kinetics of degradation of metalaxyl (Tables 12 and 13) in soils indicate that metalaxyl had prolonged persistence in all the soils. The persistence data indicated a close correspondence to first order kinetics ($R^2 = > 0.918$). The rate constants and half-life values were derived by linear regression analysis of residual concentration on a logarithmic scale against time of incubation for each soil. Increased application rate increased persistence of metalaxyl in all the soils (Fig. 7). With first order kinetics, rate constants should be independent of initial concentration, but for many herbicides, rate of loss decreased as initial concentration increased (Hurle and Walker, 1980). The reduced degradation rates at higher initial concentrations might result from a limitation in the number of reaction sites in soil (Hance and Mckene, 1971) and toxic effects on microorganisms or enzyme inhibition (Hurle, 1981 and Zheng *et al.*, 1989). So the half-life values ($t_{1/2}$) are slightly higher in higher rate of application of metalaxyl, because the half-life values are inversely proportional to the rate constant (K_{deg}) values. The 50 per cent dissipation of metalaxyl calculated by the conventional method is slightly higher than the modified method. This may be due to ignoring of the initial

concentration calculated based on the observed dissipation of the pesticide. In majority of the cases the initial concentration calculated based on the equation may not coincide with the actual initial dose applied. The C_0 (calculated) will therefore be varied depending upon the magnitude of the rate of the dissipation of the fungicide.

Degradation of metalaxyl was faster under field capacity moisture regime than under air dry moisture in all the soils (Fig. 8). The rate constants for air dry is lesser than field capacity moisture in all the soils. The rate constants for Bangalore, Chettalli and Hiriur soils are 4.20×10^{-3} , 7.97×10^{-3} and $7.83 \times 10^{-3} \text{ day}^{-1}$ under air conditions and 10.84×10^{-3} , 10.24×10^{-3} and $14.20 \times 10^{-3} \text{ day}^{-1}$ under field capacity moisture regime. The half-life value for air dry soils (84.57 - 155.81 days) are higher than field capacity (45.44 - 61.28 days). As temperature increases from 15°C to 25°C, the persistence of metalaxyl decreased, consequently rate constants and half-life were also decreased at 25°C temperature. The half-life of metalaxyl were 107.14, 75.98 and 64.57 days for 15°C, while 61.28, 59.04 and 45.47 days for 25°C temperature conditions in Bangalore, Chettalli and Hiriur soils. The moisture at field capacity level and temperature of 25°C increased the degradation of metalaxyl in soils.

It may be presumed that soil moisture at field capacity and temperature of 25°C enhances the biological degradation of metalaxyl since these conditions are favourable to microbiological activity that can affect the degradation of metalaxyl in soils.

To monitor the relative importance of chemical and microbial degradation, the persistence of metalaxyl was studied under sterile (autoclaved) and non-sterile (non-autoclaved) soil conditions. The persistence of metalaxyl was higher in sterile condition of all the three soils suggesting the major pathway of degradation as microbiological (Table 11 and Figure 10). The per cent degradation of metalaxyl was lower in sterile condition (30.0 - 42.3 %) compared to non-sterile condition (59.8 - 70.3 %). In the present study it is assumed that, the lesser degradation in sterile condition is due to the suppression of microbial activity though it is known that autoclaving can also alter the soil organic and mineral matter sufficiently to inhibit several important classes of abiotic soil reactions (Wolf *et al.*, 1989).

Sharom and Edgington (1982) reported that the half-life of metalaxyl in soils were 3 to 8 weeks. The higher value of half-life (8 weeks) of metalaxyl in muck

soil was attributed to the higher adsorptive capacity of the soil due to its very high organic matter content (62.8 %) that may have made metalaxyl less available to the soil microorganisms. Bailey and Coffey (1984) also explained that metalaxyl was very stable in chemically sterilised soils with more than 90 per cent of the initial concentration remaining after 70 days. However, metalaxyl was degraded in unsterilised soil which indicates that biological degradation was responsible for its loss in soils.

In contrast to the results obtained in this study, the metalaxyl half-life in soils were 14 days (Bailey and Coffey, 1984) and 6 days (Drobey and Coffey, 1991) indicating a high potential for specific soils to degrade metalaxyl.

The major transformation involved in the metabolism of metalaxyl is hydroxylation on the aromatic ring or on either methyl side chain of the ring by the microorganisms (*Syncophastrum racemosum*) as explained by Zheng *et al.* (1989). Hydroxylation on the methyl side chain of the ring can result in the formation of metalaxyl benzyl alcohol.

Among the soils, irrespective of the factors, the degradation of metalaxyl was more in Hiriur followed by Chettalli and Bangalore soils. The lower persistence of metalaxyl in Hiriur soil may be due to unstability of fungicide in alkaline pH that may favour degradation of metalaxyl. However, the organic matter, clay and cation exchange capacity of the soils did not affect the persistence of metalaxyl, since the metalaxyl is highly soluble in water (7.1 g / Lit). Though the adsorption of metalaxyl in soils ranged from 36.02 to 38.77 $\mu\text{g g}^{-1}$ from 100 $\mu\text{g g}^{-1}$ level, the metalaxyl will come back to the solution when advanced to degradation in order to meet the equilibrium. The rate of decomposition of metalaxyl was also reported to be influenced by soil types (Musumeci and Ruegg, 1986).

5.2.2 Mancozeb : The persistence and degradation of mancozeb in soils are influenced by various factors like type of soil, application rate, soil moisture, temperature and soil sterilization. The data are presented in Tables 14 and 15 and Figures 11 to 14 and their kinetics of degradation (Tables 16 and 17).

The data suggests a close correspondence to first order degradation kinetics ($R^2 > 0.916$). The persistence of mancozeb was almost the same in all the three

soils and the degree of degradation varied due to different factors studied. More than 40 per cent degradation of residues was noticed with in 10 days reflected the rapid rate of degradation. Further degradation of remaining 50.0 per cent residues has taken more than 50 days after incubation reflecting slow rate of residue degradation in the second phase. The results of the present study are in conformity with the finding of Doneche (1983).

It appears that the application rates of mancozeb did not affect the degree of degradation. However, a slightly higher values of half-life and lower degradation rate constants were noticed for higher rate of application in soil. This may be attributed to possible toxic effects on microorganisms or enzyme inhibition as reported by Hurle (1981). Increase in moisture content from air dry to field capacity and temperature from 15°C to 25°C also increased the degradation of mancozeb in all the soils.

The per cent degradation of mancozeb were 75.2 to 80.9 in air dry moisture while 82.8 to 87.9 in field capacity moisture levels over 60 days after application of fungicide. The similar trend was also noticed for temperature treatment. Compared to soil moisture regimes, the temperature treatments affected narrow range of degradation of mancozeb in all the soils. Correspondingly the higher value of degradation rate constants in field capacity and temperature of 25°C (28.48×10^{-3} to $32.31 \times 10^{-3} \text{ day}^{-1}$) compared to air dry moisture (22.96×10^{-3} to $25.61 \times 10^{-3} \text{ day}^{-1}$) and at 15°C temperature (25.82×10^{-3} to $28.86 \times 10^{-3} \text{ day}^{-1}$). The half-life values are maximum in air dry moisture and temperature of 15°C compared to field capacity moisture and temperature of 25°C. These results concur very close to Doneche (1983) who has reported complete elimination of mancozeb residues within 3 months and concluded major role of chemical and biological pathway of residue degradation in soils. Similar degradation rates for maneb and zineb were reported by Nash and Beall (1980) as also by Zhang *et al.* (1991). Whereas Calumpang *et al.* (1993) observed the rapid loss of mancozeb in soils with an initial half-life of 2.9 days, after 15 days mancozeb was virtually undetectable when mancozeb was applied at the rate of $5.5 \text{ kg a.i ha}^{-1}$ in field conditions. The difference in the values of the degradation of the mancozeb in the present investigation and those reported by Calumpang *et al.* (1993) may be due to the differences in the properties of the soils.

The data presented in Tables 15 and 17 indicated that sterile soil condition had a higher persistence of mancozeb than that of non-sterile soil at all the periods of incubation. The residues left after 60 days of mancozeb application were two times higher in sterile soil condition as compared to non-sterile soil conditions. This suggested that soil microbes play an important role in degradation of mancozeb in soils. The longer persistence in sterile soils may be due to the fact that autoclaving the soil result in destruction of physical and chemical structure and thereby affect the reaction of soils with organic chemicals (Wolf *et al.*, 1989). Doneche (1983) also emphasised and explained the importance of biological degradation of mancozeb in soils.

The difference in the persistence of mancozeb in the three soils in present study was less irrespective of the differences in the soil properties (Table 2), while Gupta and Bhattacharjee (1987) reported faster degradation of mancozeb in acid soils particularly at pH below 4 than alkaline soils. It may be seen that none of the soil in present study has such low pH values.

The conversion of ETU from mancozeb was noticed in all the soils throughout incubation period. The amount of ETU conversion was maximum up to 30 days of incubation. The amount of ETU conversion was higher from 20.0 $\mu\text{g g}^{-1}$ (0.94 - 0.99 $\mu\text{g g}^{-1}$) than 10.0 $\mu\text{g g}^{-1}$ mancozeb (0.53 - 0.58 $\mu\text{g g}^{-1}$) at 30th day of incubation. Newsome (1975) recorded ETU of 0.35 $\mu\text{g g}^{-1}$ in soil 15 days after treatment with maneb at the rate 3.0 lbs per acre to tomato crop. Calumpang *et al.* (1993) also recorded ETU to the extent of 1.59 and 0.004 $\mu\text{g g}^{-1}$ from mancozeb treated soil after 0 and 21 days of application, however the ETU was not detected in soil after the application of mancozeb applied at the rate of 2.0 lbs per acre after 60 days (Blazquez, 1973) and maneb at 2.0 lbs per acre (Rhodes, 1977).

5.2.3 Ethylenethiourea: The ETU is one of the important degradation products of ethylene (bis) dithiocarbamate fungicides. The results of ETU converted from mancozeb were explained in the section 5.2.2. Since, the rate of ETU conversion and its degradation go simultaneously in soil and therefore, it is difficult to predict the degradation kinetics. Further, if the rate of conversion is greater than degradation of ETU, then the value persisted is higher or vice-versa. So, to know the exact degradation of ETU in soils, a separate experiment was carried out as in mancozeb

degradation studies. It was observed that degradation of ETU (Tables 18 and 19) in lower doses are maximum compared to higher doses. No ETU residues were detected on 60th day from $2.0 \mu\text{g g}^{-1}$ level compared to $4.0 \mu\text{g g}^{-1}$ level ($0.12 - 0.14 \mu\text{g g}^{-1}$). But the rate constants and half-life derived from the equation were same in both the level of ETU application in soils. With increase in moisture content the persistence of ETU in soils decreased. The value of rate constants decreased and half-life values were increased with decreasing in moisture content of the soils. The half-life values for air dry moisture ranged from 23.98 to 30.53 days and for field capacity moisture from 14.67 to 16.28 days in soils. It was also noticed that the degradation rate were lower and half-life values were higher at temperature of 15°C . The low content of ETU at field capacity and temperature of 25°C may therefore be due to higher rate of degradation. Blazequez (1973) reported the persistence of $86.0 \mu\text{g g}^{-1}$ and traces by 13th and 27th days after application respectively. Rhodes (1973) also observed similar half-life of less than four weeks under field conditions when applied at the rate of 2.0 lbs per acre. Miles and Doerge (1991) showed reaction rate of ETU in soils approximately doubled for every 10°C increase in temperature ($E_{\text{activation}} = 15.7 \text{ K Cal mol}^{-1}$). Nash and Beall (1980) observed a short half-life of 3 days in soil at pH 6.7 containing 5.2 per cent organic matter under temperate conditions. Similar observations were also made by Calumpang *et al.* (1991) that ETU degraded rapidly with 78 per cent reduction in 3 days after application of ETU.

The lower rate of degradation of ETU in sterile soils as compared to non-sterile soils suggest that the degradation of ETU in soil is micro-biological. The half-life value in sterile soil condition are almost two times in all the soils. Miles and Deorge (1991) concluded that degradation of ETU in soils was affected by sterilisation with gamma irradiation and autoclaving. The observed degradation rates were 0.354 and 0.007 day^{-1} for gamma irradiation and autoclaving of soils when compared to control soil (0.484 day^{-1}).

The degradation pattern of ETU residues was different in the three soils as faster rate of degradation was noticed in Hiriyur soils as compared to Chettalli and Bangalore soils. This may be due to higher stability in ETU in Bangalore and Chettalli soils and increased degradation under alkaline condition of Hiriyur soil. This explained the low persistence of ETU in Hiriyur soil. However no wide variation in degradation pattern of ETU was noticed as reported by Miles and Deorge (1991):

5.3 Plant uptake and persistence of fungicides in tomato and their persistence in soil.

5.3.1 Plant uptake of fungicide by tomato

5.3.1.1 Metalaxyl : The translocation of metalaxyl in tomato plants following uptake from soil increased up to 15 days to attain maximum concentration after application irrespective of the soil application rates used in the study. The absorption of metalaxyl in tomato plants reached the maximum concentration of $10.38 \mu\text{g g}^{-1}$ on 15th day and thereafter decreased to $0.62 \mu\text{g g}^{-1}$ by the 40th day of metalaxyl-mancozeb application to soil at $225.0 \text{ mg a.i pot}^{-1}$. Plant absorbed metalaxyl moved to fruits with water through xylem tissues. However, only very insignificant amount of $0.48 \mu\text{g g}^{-1}$ residues were detected in fruits from the first harvest (after 30 days). The quantities of residues were within safe limits of $0.5 \mu\text{g g}^{-1}$ prescribed by Anon. (1993). Since metalaxyl is a systematic fungicide, its translocation is mainly apoplastic but trace amount of downward translocation has also been reported by Cohen *et al.* (1979). He has further reported that metalaxyl (Metalaxyl 50 WP) at the rate of $17.5 \text{ mg per plant}$ provided full protection of tomato leaves against *Phytophthora infestans* 1.0 hour after soil drench application. Rowe *et al.* (1982) applied metalaxyl to soil at the rate of $68.0 \text{ mg per plants}$ (3.4 kg ha^{-1}) to potatoes grown under field conditions that effectively controlled *Phytophthora infestans* disease up to 60 days after application.

Rapid plant absorption and translocation of metalaxyl in sun flower was reported by Murucchini *et al.* (1983) when incorporated into soil that controlled disease caused by *Plasmopora helianthi* effectively. Metalaxyl fungicide reached the maximum concentration in the leaves within 20 days of application. Then, there was a progressive decrease in residue and 34 days after planting the concentration of the metalaxyl in the leaves was 70 per cent lower than the maximum concentration regardless of the treatments and its presence in the leaves was still detected 90 days after planting. The decrease in the concentration of the fungicide in the plants has to be attributed to the variation in the ratio between weight of the metalaxyl and weight of the plant rather than to a degradation of the active principle.

The metalaxyl was readily absorbed by roots and translocated throughout young cabbage, red raspberry and straw berry plants as observed by Carries and Bristow (1987). They further explained that the movement in the transpiration stream

appears to account for the distribution of metalaxyl within the plants. The absorption of metalaxyl progressed mainly due to less degradation and continued supply of the metalaxyl by soil (Table 24).

5.3.1.2 Ethylenethiourea : Plant uptake pattern of ETU following soil application of metalaxyl-mancozeb has been presented in Table 23. It was observed that the absorption of ETU increased up to 20 days after application and afterwards the concentration of ETU was reduced (Figure 19). The plant absorption of ETU attained maximum concentration of 1.45 and 2.47 $\mu\text{g g}^{-1}$ residues from 112.5 and 225.0 mg a.i. metalaxyl- mancozeb treatments per pot respectively within 20 days. The plant absorbed ETU made their way to fruits and a meager amounts of less than 0.06 $\mu\text{g g}^{-1}$ residues could be detected in fruits from the harvests made after 30 and 40 days. The recovery of small quantities of ETU from fruits reflected the dissipation of residues in plant system simultaneous to conversion.

Hoagland and Frear (1976) showed that the radioactivity from ^{14}C -ETU was rapidly taken up from an aqueous solution by the roots of seedlings of corn, lettuce, pepper and tomato plants that further transported to leaves. The plant species tested can readily degrade ETU once it is absorbed and translocated in the plant. Nash (1976) observed ETU content in soybeans from EBDC fungicide treated to soil remained low ($<9.2 \mu\text{g g}^{-1}$) throughout 15 day monitoring period when applied at $10.0 \mu\text{g g}^{-1}$. The rapid rise of ETU residues in lettuce leaves within 4 days by absorption from the soil application at 10.0 mg per pot and then remaining virtually constant for 3 to 4 days was reported by Smith *et al.* (1988). The residues were however declined rapidly to non-detectable levels within 20 days. Further if the daily addition of ETU (2.0 mg) continued the level of ETU in the lettuces stabilised at about $0.36 \mu\text{g g}^{-1}$. At this level the rate of loss from the leaves must balance the rate of uptake from the soil. However, the concentration in the leaves never reached the level ($0.7 \mu\text{g g}^{-1}$) found in the 10.0 mg pot^{-1} treatment, suggesting that the level in the lettuce depends on the concentration rather than the amount of ETU available in the soils and additional ETU is not cumulative. The authors further showed that the ETU content increased rapidly during the first 3 days reaching a maximum level after 10 to 12 days. Once the maximum level had been reached, the decline was much slower and ETU did not fall to an undetectable level until 20 to 35 days after initial application of zineb or maneb. It therefore appears that additional ETU was being formed in the soil

(Table 25) by the breakdown of fungicides and the same was being absorbed by the plants (Tables 24). The rate of ETU formation was however not sufficient to produce a steady level presumably as the fungicide itself declined.

5.3.2. Persistence of fungicide residues in soil

5.3.2.1 Metalaxyl : The persistence of metalaxyl retained in soil was decreased drastically in tomato grown soils compared to laboratory incubation studies using soil alone. The residues of metalaxyl left in soil after the plant absorption degraded at the half-life of 6.52 and 9.57 days from the two respective treatments at 112.5 and 225.0 mg a.i. per pot. The less persistence of metalaxyl in the soil may be attributed to the partition of residues in plant uptake (Table 23), leaching in soil and degradation (chemical, microbiological and photo chemical) as also volatilization of the metalaxyl from soil surface. Sharom and Edgington (1986) observed the similar dissipation pattern of the metalaxyl in the soil under field conditions where half-life of residues were approximately 15 days at a soil depth of less than 30 cm.

5.3.2.2 Mancozeb and Ethylenethiourea : Substantial loss of mancozeb residues to the extent of 90.4 per cent has taken place in the soil for the left over residues after plant uptake from the two treatments of 112.5 and 225.0 mg per pot respectively within 30 days. As explained earlier, this period coincided with the major processes of fungicide residues partitioning into plant uptake, metabolism and soil degradation that caused the rapid loss in such soil. The rate constants were higher compared to laboratory studies and accordingly short half-life were observed. Similar observations were recorded by Blazquez (1973), Nash and Beall (1980), Doneche (1983) and Calumpang *et al.* (1993).

The ETU formation from the mancozeb treated pots were increased with increase in concentration and incubation period up to 20 days at lower dose and 10 days in higher dose of application. The ETU content increased up to certain period signifying the completion of metabolism process and then it decreased due to degradation of its residues. This phenomenon may be attributed to the conversion and degradation acting simultaneously in the soil. The results of the finding are in conformity with the finding of Blazquez (1973), Newsome (1975), Nash and Beall (1980) and Calumpang *et al.* (1993). However contradictory report of Rhodes (1977) shows no traces of ETU in soil treated with mancozeb at the rate of 2.0 lbs per acre.

5.4 Dissipation of fungicide residues in/on tomato and soil under field condition

5.4.1 Dissipation of fungicide residues in/on tomato

5.4.1.1 Metalaxyl : The perusal of residue data of metalaxyl on tomato foliage and fruits (Table 25 and 26) showed dissipation of residues followed the first order kinetics ($R^2 > 0.9$). The degradation rate constants were 150.85×10^{-3} and 186.49×10^{-3} for foliage and 85.07×10^{-3} and $73.04 \times 10^{-3} \text{ day}^{-1}$ for fruits at 0.2 and 0.4 per cent metalaxyl-mancozeb respectively. The dissipation rate of residues was however more from higher application rate resulting in the lower half-life of 3.13 days as compared to 4.14 days for lower application rate in foliage. The rate of residue decay in fruits followed the half-life of 7.02 and 8.75 days respectively from the two spray treatments. A distinct pattern of residue dissipation was followed in fruits and foliage (Figures 22 and 23). The faster rate of loss of metalaxyl in foliage may therefore be due to its rapid absorption followed by movement to fruits along with photosynthates. The waiting period for tomato fruits were worked out to be 10.63 and 17.39 days for 0.2 and 0.4 per cent spray of metalaxyl-mancozeb.

Cabras *et al.* (1985) explained the disappearance of acylanilide (metalaxyl, furalaxyl and benalaxyl) residues in tomato where they did not visualize linear or a pseudo-first order kinetics. Further they reported that metalaxyl residues were detected up to 20 days (0.16 and $0.21 \mu\text{g g}^{-1}$) after the spray of 380 and 760 g ha^{-1} to tomato crop. Narasimha Rao *et al.* (1993) reported that metalaxyl present in metalaxyl- mancozeb after third spray of 0.2 and 0.4 per cent recorded the initial deposits of 9.7 and $12.0 \mu\text{g g}^{-1}$ on grape berries which dissipated at the half-life of 2.7 and 3.3 days to below tolerance limit of $2.0 \mu\text{g kg}^{-1}$ in 6.6 and 8.6 days. These observations were close to present findings on tomato.

5.4.1.2 Mancozeb and ethylenethiourea : The dissipation pattern of mancozeb residues on tomato foliage and fruits is presented in Tables 25 and 26 and the dissipation curve in Figures 13 and 14. The initial deposit of 40.40 and $99.05 \mu\text{g g}^{-1}$ on foliage resulting from 0.2 and 0.4 per cent spray concentration of metalaxyl-mancozeb, dissipated at the half-life of 13.37 and 13.56 days reflecting similar pattern of residue decay irrespective of application rate. Further the loss of residues over a period of time show a slow and steady dissipation from 17.4 and 10.3 per cent loss within 5 days to over 90 per cent of 40 days of the spraying.

Fruits accumulated comparatively small quantities of mancozeb that dissipated at the half-life of 5.23 and 6.95 days. The dissipation rate was therefore higher in fruits as compared to foliage. The per cent loss in residues over a period of time also reflect the higher reduction of residues from the initial stage itself and finally more than 86 per cent loss was recorded within 20 days as against 60 per cent in foliage. It may be attributed to the fast rate of fruit growth to help the dilution of residues.

The highest recovery of ETU residues following the metabolism/conversion of mancozeb to ETU, immediately after the second spray was also found to decay progressively over a period of time with an increase in residues after 15 days reflecting the carry over of ETU residues from the metabolism of first application as well as the maximum conversion from the second spraying by 15 days and therefore an increase in residues. However this phenomenon could not be noticed in fruits for the ETU residues where a progressive dissipation was noticed throughout from the highest buildup of residues of 2.15 and 2.85 $\mu\text{g g}^{-1}$ after second application.

The dissipation pattern of mancozeb residues and its metabolism to ETU in present studies agree with the findings of Rhodes (1977) with respect to the dissipation and metabolism of radio labeled ^{14}C -maneb on tomato and beans. The recovery of ^{14}C -residues of ETU in tomato and beans all through was evidenced to the steady rate of metabolism. Similar dissipation pattern of mancozeb and maneb on tomato was noticed by Vonstryk and Jarvis (1978). Ripley *et al.* (1978) also concur these results in grape for zineb with the record of half-life values 15 to 20 days.

The work of Nash and Beall (1980) suggested that dissipation rate was following the first order equation $\ln(C_t) = \ln(C_0) - Kt$ and the half concentration ($C_{1/2}$) values of the EBDC fungicides and ETU in fruits and vegetables ranged from 3 to 35 days and 3 to 28 days respectively. Udaykumar and Agarwall (1990) reported the half-life of 15 and 10 days for mancozeb and maneb on brinjal plants respectively. Both these compounds were most persistent in leaves and least in fruits. Besides fungicides, ETU also detected in the brinjal fruits as degradation product. The amount of ETU was 0.03 $\mu\text{g g}^{-1}$ on zero day which increased to 0.51 $\mu\text{g g}^{-1}$ on 20th day after treatment, however a gradual decline was observed thereafter and this suggests the formation of ETU from EBDC fungicides and then its breakdown to certain other

compounds. Contradicting the present investigations, a short half-life of 3.1 to 3.3 days on grapes was reported by Narasimha Rao *et al.* (1993).

Present studies suggest waiting periods of 10.0 and 15.93 days for mancozeb residues based on the persistence pattern of residues and prescribed MRL of $3.0 \mu\text{g g}^{-1}$. However, a very short period of only 1.0 day was recommended by Sharma *et al.* (1994) and Patil *et al.* (1993).

5.4.2 Persistence of fungicide residues in soil under crop cover

5.4.2.1 Metalaxyl : The persistence of metalaxyl residues in soil received during foliar applications of the crop at 0.2 and 0.4 per cent spray concentration is presented in Table 28 and the degradation of residues in Figure 23. The perusal of the data reflect the persistence of residues for a period of 30 days only. The residues degraded at the half-life of 8.77 and 9.60 days reflecting faster loss of residues by 10 - 20 days coinciding with the maximum plant uptake. The rate of loss of residues in the beginning may therefore be due to soil degradation as well as plant absorption. Similar short half-life values of metalaxyl in soil were also reported by Bailey and Coffey (1985), Sharom and Edgington (1986) and Drobey and Coffey (1991).

5.4.2.2 Mancozeb and ethylenethiourea : The soil buildup of mancozeb and resultant ETU has been presented in Table 28 and graphical presentation of degradation of residues in Figure 23. The perusal of the data on persistence of mancozeb residues shown meager quantities of residues spilled on soil surface that degraded at the half-life of 6.1 and 6.5 days to non-detectable levels within 30 days following breakdown of residues as well as transforming into ETU and other metabolites. Accordingly, ETU residues were also detected all along the persistence of mancozeb. The metabolism of mancozeb to ETU was however increased ETU concentration to its maximum quantities after 5 days and thereafter degraded progressively to non-detectable levels within another 25 days.

It is assumed that the degradation pattern of mancozeb in soil under crop cover is also influenced by root activity, rhizosphere micro-organisms, environmental and other associated factors. Blazquez (1973), Newsome (1975), Rhodes (1977), Nash and Beall (1980), Doneche (1983), Zhang (1991) and Calumpang *et al.* (1993) also

observed short half-life and faster degradation of mancozeb and the formation of ETU in the soil when EBDC fungicides applied at recommended doses.

5.5 Decontamination of fungicide residues from tomato fruits

The reduction in fungicide residues from fruit surface following washing of fruits in tap water was to the extent of 25 to 28 per cent for metalaxyl, 40 to 60 per cent for mancozeb and 16.4 to 39.4 per cent for ETU. Washing followed by cooking of fruits however increased the loss to a great extent for all the fungicides to the extent of about 80 per cent of metalaxyl and mancozeb but only about 40 per cent for ETU. The dislodging of the surface residues was higher for mancozeb by washing as compared to metalaxyl and ETU residue due to their movement in the fruit (Table 30).

All the fungicide residues in the present investigation were reduced upon washing and washing+cooking except ETU residues (Figure 24). This may be attributed that the metalaxyl and ETU are soluble in water, while mancozeb are easily dislodgeable from the fruits. The continued persistence of ETU even after cooking of fruits confirms the continuance of the metabolism of mancozeb under influence of heat during cooking.

Marshal and Jarvis (1979) have reported the reduction of mancozeb and ETU residues to the extent of 50 per cent following the processing of treated tomato fruits into fruit juice. Trotter and Pardue (1982) reported the effectiveness of washing on reduction of EBDC residues from spinach and found no indication that ETU residues are similarly affected perhaps as a result of their being incorporated into the spinach. Sharma *et al.* (1994) suggested simple processing of freshly harvested vegetables effective in decreasing the amount of mancozeb as a result of washing.

Most of the researches observed lower residues of EBDC fungicide upon cooking for 10 to 15 minutes (Sharma *et al.*, 1994) and the continued persistence of ETU in tomato (Newsome 1976); grapes (Ripley *et al.*, 1978) and apples (Hajslova *et al.*, 1986).

SUMMARY

VI. SUMMARY

The present investigation on “Behaviour of metalaxyl and mancozeb residues in soils and tomato” was taken up with the main objectives of finding the extent of adsorption and degradation of fungicide residues in soils, their uptake and persistence in tomato plants. The studies were conducted on a systemic fungicide metalaxyl and a contact fungicide mancozeb along with its major metabolite ETU interacting with tomato plant and three main types of soil from Bangalore, Chettalli and Hiriya representing the different textural classes. The salient features of the investigation are summarised as follows:

I. Adsorption of metalaxyl and ETU in soils

1) Adsorption of metalaxyl was mainly influenced by the clay content of the soils. Metalaxyl adsorption in soils decreased in the order of Hiriya > Chettalli > Bangalore, which is also the order of decreasing clay content. The adsorption of ETU in soils was found to be influenced by organic carbon content and it followed the order: Chettalli > Bangalore > Hiriya. The influence of texture or clay content of the soils was not evident. The per cent adsorption of metalaxyl was higher than ETU in all the soils.

2) Freundlich equation provided a good fit ($R^2 > 0.9$) for the adsorption data on metalaxyl and ETU in all the soils. The Freundlich constant K values for metalaxyl were higher than those of ETU in all the soils and ranged from 5.870 to 7.281 for metalaxyl and from 1.860 to 2.306 for ETU. The values for $1/n$ (energy of adsorption) were less than unity ($1/n < 1$) for both metalaxyl and ETU and indicated non-linear relationship between the fungicide or metabolite concentration and adsorption and therefore can be characterised as L-type of adsorption.

3) The sorption coefficients for organic carbon (K_{oc}) were higher for metalaxyl than ETU in all the soils and ranged from 362 to 2275 $\mu\text{g ml}^{-1}$ for metalaxyl and 128 to 581 $\mu\text{g ml}^{-1}$ for ETU. The Gibb's free energy (dG°) were always negative indicating that the adsorption is exothermic.

II. Degradation of fungicides residue in soils

1) The degradation pattern of metalaxyl, mancozeb and ETU residues indicated a close correspondence to first order exponential degradation kinetics in soils and mainly

influenced by soil moisture and temperatures. The rate of degradation was faster at lower compared to higher application rate. Among the factors studied, the soil moisture at field capacity and temperature of 25°C were found to accelerate the degradation of fungicides or metabolite in the soils. However, the persistence of fungicides residues were higher from higher dose of application, air-dry moisture, temperature of 15°C and sterile soil conditions. The degradation of fungicides in soils was predominantly biological as well as chemical in nature.

2) Slower degradation of metalaxyl was noticed in the soils and their half-life values were higher than mancozeb and ETU as evident by wide range of half-life values from 41.24 to 165.11 days. In case of metalaxyl Hiriur soil was found to be superior in degrading the metalaxyl.

3) Lower persistence of mancozeb and ETU was observed in the soils resulting in rapid rate of degradation at smaller half-life values as compared to metalaxyl indicating the faster degradation of mancozeb and ETU. In mancozeb treated soils, the ETU formation was increased up to 30 days of incubation and thereafter it declined. Among soils, the degradation of either mancozeb or ETU is not influenced by soil types and only small variations were observed. However mancozeb persistence was higher in Hiriur soils than Chettalli and Bangalore soils.

III. Plant uptake of fungicides by tomato

1) The plant uptake studies have indicated that Maximum concentration of metalaxyl and ETU residues in tomato plants following uptake from soil was attained after 15 and 20 days of soil application respectively. The residues were however reduced further and reached below the prescribed MRL in fruits by first harvest itself.

2) The degradation of soil retained fungicide residues followed the first-order exponential reaction ($R^2 > 0.9$) and metalaxyl persistence was higher than mancozeb. The half-life values for metalaxyl were in the range of 6.52 to 9.57 days, while mancozeb ranged from 9.74 to 12.41 days for different treatments. The rapid disappearance of metalaxyl and mancozeb residues to the extent of 59.2 to 72.4 and 3.81 to 48.3 per cent was found within 10 days of fungicide applications at the two application rates.

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3) The ETU residues were also found in metalaxyl-MZ treated soils indicating the metabolism of mancozeb. Higher amount of ETU residues were noticed on 10 and 20 days after application. The maximum amounts of ETU were recorded in higher dose within 10 days as compared to 20 days in lower dose (112.5 mg pot⁻¹).

IV. Dissipation and persistence of fungicides in/on tomato and soil under crop cover

1) Foliage accumulated comparatively higher initial residues of fungicides than fruits following foliar applications. Metalaxyl residues dissipated at higher rate of residue decay in foliage as compared to fruits. A reverse order on dissipation of mancozeb residues was found in foliage and fruits.

2) The half-life of metalaxyl in foliage ranged from 3.13 to 4.14 days and 7.82 to 8.75 days for tomato fruits as compared to the mancozeb half-life of 13.87 and 13.56 days for foliage and 5.23 and 6.95 days for fruits from 0.2 and 0.4 per cent metalaxyl-MZ spray respectively.

3) Maximum concentration of ETU was recovered on the zero day of second spray compared to other days indicating the carry over of ETU residues metabolised from mancozeb of the first spray application. The persistence of ETU in foliage continued beyond 40 days but fruits were devoid of any detectable residues.

4) Safety constants worked out in terms of waiting period required 18 days after the last spray application of metalaxyl-MZ to bring residues down the prescribed MRL of 0.5 µg g⁻¹ for metalaxyl and 3.0 µg g⁻¹ for mancozeb.

5) The degradation of fungicide residues in soils under the field conditions were faster than laboratory incubation studies. The degradation rate constants were accordingly higher in field conditions (75.63×10^{-3} to $127.85 \times 10^{-3} \text{ day}^{-1}$) compared to laboratory incubation studies (10.24×10^{-3} to $46.99 \times 10^{-3} \text{ day}^{-1}$). The half-life values were also less than 10 days for metalaxyl, while 7 days for mancozeb. The residues of the both the fungicides received by the soil from foliar applications to tomato crop did not persist beyond 30 days.

V. Decontamination of fungicide residues from tomato fruits

1) Fungicide residues in general were decreased upon washing and cooking of tomato fruits from the aged residues after 5 and 10 days of spray application. The rate of loss of residues were almost same from the two application rates upon washing and washing followed by cooking while the intensity of dislodging the residues being severe from washing followed by cooking. Further the dislodging effects were more pronounced on the less aged residues.

2) Washing effected reduction of 25.3 to 28.9 per cent metalaxyl, 40.3 to 60.4 per cent mancozeb and 16.4 to 39.4 per cent ETU residues.

3) Decontamination processes of washing followed by cooking removed as high as 78.3 to 78.6 per cent metalaxyl; 74.6 to 78.4 per cent mancozeb and 44.5 to 48.3 per cent ETU residues.

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