CORE
Metadata, citation and similar papers at core.ac.uk
Provided by Stellenbosch University: SUNJournals

# Accelerated Microbial Degradation of Nematicides in Vineyard and Orchard Soils

H.J. Hugo[1*], C. Mouton[2], A.P. Malan[3]

(1) ARC Infruitec-Nietvoorbij[1], Private Bag X5026, 7599 Stellenbosch, South Africa
(2) Nemconsult, PO Box 877, Upington 8800, South Africa
(3) Department of Conservation Ecology and Entomology, Faculty of AgriSciences, Stellenbosch University, Private Bag X1, 7602 Matieland, Stellenbosch, South Africa

Submitted for publication: November 2013
Accepted for publication: March 2014

Key words: Accelerated microbial degradation, AMD, carbamates, nematicides, organophosphates, review

**Accelerated microbial degradation (AMD) of organophosphate and carbamate nematicides is a phenomenon whereby biodegradation in the soil is increased, leading to a dramatically shortened persistence of nematicides. More intensified agriculture practices in South Africa in response to the future demand for food may lead to increased pest and disease pressure, which in turn will lead to more frequent pesticide application. The same principle applies to plant-parasitic nematode control practices, and the overuse and misuse will have a pronounced effect on the enhancement of AMD. With limited management options available, the responsible use of nematicides becomes more pertinent. Producers should be aware of the problems associated with multiple soil applications of organophosphates and carbamates against plant-parasitic nematodes. This article reviews factors contributing to the AMD of carbamate and organophosphate nematicides in soil and makes practical recommendations to avoid the occurrence of AMD in vineyard and orchards.**

## INTRODUCTION

The most important plant-parasitic nematode species found in South African vineyards and deciduous fruit orchards include *Criconemoides xenoplax* (Raski, 1952) Loof & De Grisse, 1989 (ring nematodes), *Longidorus* spp. (needle nematodes), *Meloidogyne* spp. (root-knot nematodes), *Paratrichodorus* spp. (stubby-root nematodes), *Pratylenchus* spp. (root-lesion nematodes), spiral nematodes from different genera, *Tylenchulus semipenetrans* Cobb 1913 (citrus nematodes) and *Xiphinema* spp. (dagger nematodes) (Addison & Fourie, 2007), of which C. *xenoplax* and *Meloidogyne* spp. are known to be the most damaging to vines and deciduous fruits (Storey, 2007).

Nematicides are pesticides used to control a wide variety of plant-parasitic nematodes on many crops, including grapevines and fruit. There are two main classes of nematicides: fumigant and non-fumigant nematicides. Currently, the majority of the nematicides belong to the non-fumigant group (Table 1). Non-fumigant nematicides are all organophosphate and carbamate pesticides. In contrast to the fumigants, these chemicals depend heavily on initial mixing with the soil and local redistribution in solution in the soil water. Some are systemically absorbed and redistributed within plants. They are commonly formulated as granules, but many are also available in more concentrated emulsifiable or water-soluble spray liquids (Dunn & Noling, 2003).

Nematicides may be removed from soil through at least three routes: chemical degradation, leaching, and biological degradation (Dunn & Noling, 2003). Biological degradation occurs when living organisms, usually bacteria and fungi, metabolise the active ingredient and its breakdown products to access the carbon as an energy source. Accelerated microbial degradation (AMD), or the biodegradation of nematicides, is a term used to describe situations in which the normal process of biological degradation of soil-applied nematicides is drastically increased, causing premature loss of the nematicide activity from the target area. This results in poor levels of plant-parasitic nematode control, as the product is then not active in the soil long enough to give effective control. The continuously growing list of pesticides affected by AMD also includes insecticides, fungicides, herbicides and fumigants (Arbeli & Fuentes, 2007).

It is speculated that climate change will result in new and more aggressive plant-parasitic nematode populations (Coakley *et al*., 1999). Already, increased nematode

[1] The Fruit, Vine and Wine Institute of the Agricultural Research Council

*Corresponding author: e-mail address: hugoh@arc.agric.za
*Aknowledgements: The authors would like to thank Hortgro Science, for funding of the project.*

TABLE 1
Classification of non-fumigant nematicides and their registration with reference to accelerated microbial degradation (AMD).

| Type of nematicide | Active ingredient | Product name | Registered | Reference to AMD |
|---|---|---|---|---|
| Organophosphates | Fenamiphos*** | Nemacur® | Organophosphates | Fenamiphos*** |
| | Cadusaphos | Rugby®<br>**Sokker | Grapes; stone fruit; pome fruit | Karpouzas *et al*. (2004) |
| Carbamates | Aldicarb* | Temik® | Grapes, stone fruit | Smelt *et al*. (1987) |
| | Oxamyl | Vydate®<br>**Blockade | Stone fruit | Smelt *et al*. (1987); Haydock *et al*. (2012) |

* Currently not available, and prohibited from January 2014
** Trade name of generic products
*** Plums excluded

problems in South Africa over the past two decades are forcing grape and tree fruit producers to increase the use of nematicides. However, it is exactly this more frequent use of nematicides that has greatly induced AMD and has led to poor nematode control. In this review we concentrate on the accelerated degradation of nematicides in agricultural soils as reported in the literature, and aim to identify key problem areas regarding AMD, as well as present recommendations on how to avoid AMD in vineyards and fruit orchards.

## THE CAUSE OF ACCELERATED DEGRADATION

According to Harrison (1990), the fate of pesticides can be separated into three major types: adsorption, which is the binding of pesticides to mineral or organic matter; transfer processes, which move pesticides from one place to another in the environment; and degradation processes, which break down these pesticides. Pesticide degradation processes break down pesticide molecules into simpler and, generally, less toxic compounds. However, degradation can be problematic when a pesticide is destroyed before control of the target organism has been achieved.

A combination of factors such as soil characteristics, climate and the structure of the nematicide molecule determines the persistence of nematicides in the soil. Soil is a complex and variable environment that is difficult to standardise in relation to nematicide degradation. The most prominent feature of the fate of organophosphate and carbamate nematicides in soil is their decreased persistence as toxic molecules compared to chlorinated hydrocarbon insecticides. Most organophosphate and carbamate nematicides are very susceptible to degradation in the soil, as soil is an excellent medium for both non-biological (chemical breakdown) and biological modifications (microbial breakdown). Microbial metabolism of these two classes of nematicides seems to be the most important factor accounting for their degradation in soil (Laveglia & Dahm, 1977).

Chemical degradation is the breakdown of pesticides by processes that do not involve living organisms. The adsorptive capacity, pH, temperature, moisture content and mineralogy of the soil all affect the rate and type of chemical reaction that occurs. The principal chemical reactions are hydrolysis, reduction, oxidation and dealkylation, although not all reactions occur with any one nematicide (Laveglia & Dahm, 1977). The most important reaction is hydrolysis, a pH-affected cleavage reaction that many complex molecules undergo in the presence of water (Harrison, 1990).

Some organophosphate and carbamate compounds are especially susceptible to rapid hydrolysis (after several hours) under alkaline conditions, and chemical reactivity generally increases with temperature increase. Also, the mineral composition of the soil defines the chemical environment into which the pesticide is introduced (Harrison, 1990).

Fenamiphos is a non-volatile, systemic organophosphate nematicide. In soil, it is rapidly oxidised to fenamiphos sulphoxide, which is slowly oxidised into fenamiphos sulphone (Leonard *et al*., 1998). Both the primary oxidation products, fenamiphos sulphoxide and fenamiphos sulphone, have nematicidal activity and toxicity similar to that of fenamiphos (Waggoner & Khasawinah, 1974), with both products being more mobile (Bilkert & Rao, 1985) and persistent (Ou & Rao, 1986). Fenamiphos sulphone is further oxidised to the desisopropyl form, which has no nematicidal properties (Anderson, 1989). Fenamiphos sulphone phenol, which is formed by the hydrolysis of fenamiphos sulphone, has also been detected during fenamiphos degradation in soil (Ou, 1991).

After the application of aldicarb by soil incorporation, soil water rapidly dissolves and releases aldicarb from the granule. Once in solution, the degradation processes, including oxidation and hydrolysis, commence immediately. Aldicarb is degraded mainly by means of oxidation and hydrolysis, during which it is rapidly oxidised to aldicarb sulphoxide, which turns into aldicarb sulphone (Ou *et al*., 1985).

### Biological factors

Microbial degradation occurs when microorganisms such as algae, bacteria and fungi utilise a pesticide as a food substrate. Microbial degradation can be rapid under soil conditions favouring microbial activity. Such conditions include warm temperatures, favourable pH levels, adequate soil moisture, aeration (supply of oxygen), and fertility. The degree of adsorption also affects microbial degradation, because pesticides must generally be in solution in order to be absorbed and metabolised by microorganisms. The frequency of pesticide applications is also an important factor that can affect such degradation (Harrison, 1990). Nevertheless, the degradation of xenobiotic compounds by members of the soil microflora is an important means by which said compounds

are removed from the environment, and thus prevented from becoming pollution-related problems (Karns *et al*., 1986). The involvement of microorganisms in the degradation of fenamiphos was proven in a study conducted by Stirling *et al*. (1992).

Another study showing the involvement of microorganisms in the degradation of nematicides was conducted by Suett (1986), who examined the degradation of carbofuran in soil. The microbial populations were found to be the lowest in soil that had been treated regularly with a soil sterilant, and it was significant that the persistence of carbofuran was greater in such soil, despite many previous applications of carbofuran, compared to the higher degradation rates in any of the untreated soils. Several soil microorganisms have been reported to degrade carbofuran (Williams *et al*., 1976; Felsot *et al*., 1981).

***Bacteria***

The biodegradation of aromatic organophosphate pesticides by microorganisms isolated from the soil is well documented in the literature. Individual bacteria such as *Pseudomonas* spp. (Serdar *et al*., 1982; Sheela & Pai, 1983) and *Flavobacterium* sp. (Sethunathan & Yoshida, 1973) have been associated with the degradation of various pesticides.

Several genera of bacteria have been linked to the enhanced degradation of carbofuran (Felsot *et al*., 1981; Karns *et al*., 1986). Carbofuran degradation by *Arthrobacter, Bacillus* and *Pseudomonas* species has been reported by Cain and Head (1991). All of the isolated strains metabolised carbofuran to carbofuran phenol, which was then degraded further to an undetectable compound. An *Achromobacter* species, isolated by Karns *et al*. (1986), was shown to be capable of removing the carbamate side chain of carbofuran at a rapid rate, and of utilising the nitrogen portion of the N-methylcarbamate side as a source of cellular nitrogen. Read (1987) found bacterial isolates from water mixtures that were capable of degrading aldicarb in soil/water mixtures. This included species of *Arthrobacter*, *Nocardia* and *Pseudomonas*, and smaller numbers of *Achromobacter* and *Bacillus*.

Singh *et al*. (2003) observed that pesticide-degrading genes are normally associated with plasmids that can move between bacterial strains, thus enhancing the diversity of the degraders. It is also well known that the degradation of several chemicals is carried out by two or more microorganisms (Nelson *et al*., 1982; Read, 1986), or by communal interaction between different components of consortia (Ou & Thomas, 1994; Sutherland *et al*., 2000).

Rajagopal *et al*. (1984) studied the metabolism of carbaryl and carbofuran in a mineral salts medium by soil-enrichment cultures, which led to a shift in the population of dominant bacteria. The selective stimulation of *Bacillus* sp. in a carbaryl-supplemented medium, and of *Arthrobacter* sp. in a carbofuran-supplemented medium, from a primary inoculum of carbaryl-enrichment culture, merits discussion, because *Bacillus* sp. and *Arthrobacter* sp. were both found to be equally efficient in degrading both carbaryl and carbofuran. Moreover, hydrolysis was the major pathway followed in the bacterial degradation of both insecticides. The reason for the substrate-specific stimulation of *Bacillus* sp. and *Arthrobacter* sp., despite a common pathway of hydrolysis for both insecticides, was not clear.

Ou (1991) concluded that microorganisms (in either axenic or mixed cultures) that were capable of degrading fenamiphos could not be isolated from field-treated soil samples. However, a mixed bacterial culture obtained from a field-treated sample can mineralise the chemical when in the presence of a small amount of soil. Ou also found that the addition of biodegradable chemicals such as glucose serves to enhance the biodegradation of fenamiphos. Ou and Thomas (1994), during further investigation of the necessity for the presence of soil, suggested that enzymes responsible for the mineralisation involved must be induced, and that certain organic constituents that are released from soil organic matter can serve as inducers. Phenolic compounds, and especially simple phenols, are likely candidates.

***Fungi***

Results obtained from a study conducted by Jones (1976) indicated that the ability to metabolise aldicarb and its sulphoxide is common in soil fungi. Pure cultures of five common soil fungi were tested for the degradation of aldicarb, with three of the five being tested against aldicarb sulphoxide, the major toxic metabolite. The group of soil fungi included three genera that are early colonisers or decomposers of organic materials, namely *Cunninghamella*, *Penicillium* and *Trichoderma*, and two (*Gliocladium* and *Rhizoctonia*) that are often associated with root surfaces (Dickinson & Pugh, 1974).

The oxidative metabolism of aldicarb to aldicarb sulphoxide was most rapid in the *Cunninghamella elegans* and *Penicillium multicolour* cultures, with *Trichoderma harzianum* showing an appreciable conversion. *Gliocladium catenulatum* had one of the higher percentages of residual aldicarb. The major organosoluble metabolites found were aldicarb sulphoxide, oxime sulphoxide and nitrile sulphoxide, with only small amounts of the corresponding sulphones. Such production indicates that the metabolic pathway in the above-mentioned fungi is similar to that which is found in higher plants and animals (Jones, 1976).

Read (1987) studied fungal development in treated soil and found that the first laboratory treatments of aldicarb applied to field-collected soil did not result in visible fungal growth on the surface of the soil. After two or more retreatments with aldicarb, however, different strains of microorganisms developed, causing rapid breakdown of aldicarb in soils with a pH higher than 6. Following the discovery of this phenomenon, data were recovered to indicate the extent of fungal growth on the soil at different times after treatment with low and high concentrations of aldicarb.

***Other organisms***

The degradation of insecticides by algae has been observed. An alga, *Chlorella pyrenoidosa* Chick, was found to be capable of metabolising parathion (Mackiewicz *et al*., 1969; Zuckerman *et al*., 1970). Zuckerman *et al*. (1970) exposed algal cultures to parathion and found that the compound was converted into aminoparathion within the algal cells, and this, in turn, was rapidly released. Mackiewicz *et al*. (1969)

conducted a study in which bean plants were exposed to parathion or parathion plus *C. pyrenoidosa* and incubated for seven days under aseptic conditions. Plants exposed to parathion showed only parathion, whereas the plants that were exposed to parathion and *C. pyrenoidosa* contained, in addition to parathion and aminoparathion, an unidentified sulphur-containing metabolite. Parathion metabolites found in plants were suggested to be products of microbial metabolism rather than of metabolism by the plant.

Actinomycetes have also been reported as being capable of enhancing the degradation of carbofuran (Felsot *et al*., 1982) and of aldicarb (Kuseske *et al*., 1974) after repeated use. Actinomycetes ('ray fungi') are defined as a group of morphologically diverse, but usually filamentous, gram-positive bacteria that sometimes have been classified as mitosporic fungi (Hawksworth *et al*., 1995).

**Non-biological factors**

It is important to realise that factors such as soil properties, climatic variations and cropping histories can affect the bioavailability of pesticides. This affects pesticide availability, either directly by affecting the adsorption, or indirectly by affecting the composition and activities of soil microbial populations. The net effects of the other factors concerned must also be considered when assessing the broader implications of accelerated degradation (Suett *et al*., 1993).

Pesticide adsorption can cause a pesticide to adhere so tightly to soil particles that it cannot be taken up by the target organisms. Too much leaching can cause the pesticide to move away from the target organism, which can lead to the injuring of non-target species or to groundwater contamination. Absorption or uptake, on the other hand, involves the movement of pesticides into plants and animals. Uptake by plants, volatilisation and the leaching of nematicides lead to a loss in the efficacy of nematicides. Such losses are dependent on the nature of the chemical compounds and on environmental conditions (D. Kiezebrink, personal communication, 1998). Many soil characteristics affect pesticide adsorption, including sunlight, pH, texture, temperature, soil moisture content and depth.

***Sunlight/ultraviolet light***

Photodegradation, the breakdown of pesticides by the action of sunlight, can destroy pesticides on foliage, on the surface of the soil, and even in the air. Pesticides, once applied, vary considerably in their degree of stability when exposed to natural light. Factors that affect photodegradation include intensity of sunlight; characteristics of the application site, such as soil type or vegetation; application method, such as broadcast versus in-furrow; and chemical and physical properties of the pesticide (Harrison, 1990).

***Soil pH***

Soil pH influences the reactivity of the chemical and certain soil functions, such as microbial metabolism (Harrison, 1990). Several studies have shown that an increase in soil pH results in an increase in soil microbial biomass and enzymatic activity (Acosta-Martinez & Tabatabai, 2000; Singh *et al*., 2003). Edwards (1973), in his review of the biological aspects of pesticide degradation, emphasised the greater activity of bacteria and actinomycetes that occurs in near-neutral acid soil, while Singh *et al*. (2003) observed that the second and third treatment with fenamiphos resulted in much more rapid degradation than the first treatment in two alkaline soils. These observation suggest that an alkaline pH in soils supports higher microbial biomass and enzymatic expression, which in turn helps the microbial community to adapt and to develop gene-enzyme systems for the enhanced degradation of pesticides.

There have been previous reports of a non-specific relationship between a high pH and the rapid biodegradation of carbamate insecticides (Suett & Jukes, 1988; Chapman & Harris, 1990; Smelt *et al*., 1996). Read (1987) found that repeated applications of aldicarb or carbofuran to a sandy loam soil led to accelerated degradation at pH 6.4, but not at pH 5.6. It was also demonstrated that carbofuran breaks down quickly by means of chemical hydrolysis in an alkaline environment (pH 7.9) (Getzin, 1973). Transformation rates of oxamyl and aldicarb were also found to be higher in sandy loam and loam soils with a pH of around 7, than in humic or peaty soils with a low pH (Smelt *et al*., 1979; Bromilow & Leistra, 1980; Smelt *et al*., 1981a, 1981b).

Microbial activity has been found to be low or negligible in low-pH soils (Siddaramappa *et al*., 1978; Read, 1986). Increased breakdown of nematicides at higher pH values can be explained partly by the increased hydrolysis of the compounds concerned, as hydrolysis is more rapid under alkaline conditions. At acid pH values, hydrolysis of the compounds will not occur, with the nematicides then mainly being oxidised to their sulphoxides and sulphones. As active ingredients in the organophosphate and carbamate families are acidic, they generally degrade much faster at a pH > 7, while most nematicides degrade at least twice as fast in lime-based soils (pH 7.5 to 8.5) than in to acidic soils (Dunn & Noling, 2003).

***Soil texture***

Terbufos is adsorbed rapidly by soil, with adsorption being correlated positively with organic matter content (Felsot & Dahm, 1979). Soils containing high amounts of clay and organic matter adsorb pesticides relatively well, possibly causing some active ingredients to be adsorbed to organic matter so tightly that they are less available for nematode control (Ou *et al*., 1985). In very sandy soils with little organic matter, low binding capacity permits excessive leaching and rapid loss of the active ingredient from the root zone. The weakly adsorbed compounds oxamyl and aldicarb, together with the oxidation products aldicarb sulphoxide and aldicarb sulphone, are easily transported downwards or upwards with water in soils that are low in organic matter content (Leistra *et al*., 1980; Smelt *et al*., 1983a, 1983b). Therefore, the products concerned are often more effective in heavier soils than in lighter soils, as a result of adsorption capacity.

Soil type also plays a key role in the rate of degradation of pesticides. Pesticides volatise readily from coarse-textured soils; the texture of soil also affects the amount of pesticide that is lost due to run-off (Harrison, 1990). Smelt *et al*. (1995) found the longest half-lives to occur in humic sandy soils, whereas the shortest were found in sandy loam soils.

***Temperature***

Microbial activity increases with increasing temperature (Hamaker, 1972), therefore an increase in temperature accelerates the degradation of pesticides, with the most frequent range for temperature studies being from ± 15 to 35°C (Laveglia & Dahm, 1977; Ou *et al*., 1982). Numerous studies have indicated that there is a positive correlation between temperature and the rate of pesticide degradation (Ramanand *et al*., 1988a; Jones & Norris, 1998). Results obtained from laboratory studies performed by Bromilow and Leistra (1980) demonstrate that higher temperatures benefited the microbes responsible for the degradation of aldicarb and fenamiphos. However, in the field, soil temperatures fluctuate both daily and seasonally. It therefore is unlikely that soil temperature has an effect on degradation rates under temperate field conditions (Johnson *et al*., 1981).

According to the available evidence, pesticides that persist in the cooler conditions of temperate countries do not necessarily persist in the hot and humid conditions of the tropics and subtropics (Arbeli & Fuentes, 2007). One of the major factors responsible for such a difference in pesticide persistence between temperate and tropical conditions is soil temperature (Talekar *et al*., 1977). The results obtained by Ramanand *et al*. (1988a) indicated that intensive use of the same pesticide might lead to the more rapid build-up of a very active pesticide-degrading microbial population under the hot-humid conditions of a tropical environment than it would under temperate conditions. From the evidence presented by Ramanand *et al*. (1988b), the bacterium, which was isolated from the soil at 35°C, appears to possess a greater capacity to mineralise carbofuran at 35°C than it does at 20°C. The above suggests that the biodegradation of carbofuran and other pesticides might proceed at a faster rate under the hot conditions of tropical environments than in more temperate conditions.

***Soil moisture***

Increased rates of pesticide degradation have been observed in soils with increased moisture content (Jones & Norris, 1998). Blumhorst (1996) suggested that the increase might be due to increased microbial activity in such soils. Ou and Rao (1986) reported that the total toxic residue of fenamiphos disappeared more rapidly in moist soil than in dry soil. At high soil-moisture levels, nematicidal compounds were more easily dissolved and dispersed, and therefore were more available for both chemical and microbial breakdown. This is the case particularly if granulated formulations are used, as the active compound needs to dissolve off the granule. Low moisture content in soil is associated with a decrease in microbial metabolic activity (Orchard & Cook, 1983), which has a direct effect on the microbial degradation of nematicides.

The primary degradation process appears to be hydrolysis (either microbial or chemical). Therefore the degradation rate of pesticides would be expected to be reduced in low-moisture soils (Bull *et al*., 1970; Jones & Norris, 1998). Bromilow *et al*. (1980) reported that the oxidation and hydrolysis rate of aldicarb in sandy loam soils containing 5% water was lower than the rate in soils containing 10% and 15% water. In completely dry conditions, aldicarb or its toxic derivatives were relatively resistant to chemical decomposition (Bull *et al*., 1970). However, such a situation would probably never occur in the field. With normal moisture levels, aldicarb would probably be degraded fairly rapidly, and the rate of decomposition would be accelerated at higher moisture levels, especially in heavier soils. In addition to chemical degradation, the residual life of aldicarb would be shortened by volatilisation enhanced by the evaporation of water from the soil adjacent to the treated area. Wet soils tend to adsorb less pesticide than do dry soils (Danielson *et al*., 1961), either because the water molecules compete for the binding sites, or because some of the pesticide molecules remain dissolved in the soil water. Therefore, pesticides volatise readily from soils with a high moisture content (Harrison, 1990).

Earlier work stressed the importance of soil water (Schuldt *et al*., 1957) and of irrigation or rainfall (Rohde *et al*., 1980) in the dispersion of non-volatile nematicides. With excessive rain or irrigation there is a risk of premature loss of an active ingredient from the root zone and of water contamination due to leaching or run-off (Dunn & Noling, 2003). The low water solubility of fenamiphos (0.04% to 0.07%) prevents rapid leaching from sandy loam soil in the absence of excess amounts of water, and allows sufficient time for adsorption by the plant of nematicidal concentrations (Rohde *et al*., 1980).

***Soil depth***

Degradation rates in subsurface layers might differ significantly from those in the surface layer because of changes in soil conditions such as organic matter content, microbial activity, moisture content and temperature (Fomsgaard & Albaiges, 1995; Smelt *et al*., 1995). Decomposition rates in subsoils are generally much slower than they are in surface soils, because microbial activity is much less deep in the soil (Bromilow *et al*., 1986). *In situ* degradation rates under field conditions are difficult to measure and in modelling studies are often assumed to decrease with soil depth (Smelt *et al*., 1978; Ou *et al*., 1985; Bromilow *et al*., 1986; Di *et al*., 1995).

Smelt *et al*. (1978) reported that the disappearance of aldicarb sulphone in subsoil samples was found to be considerably slower than it was in soil samples from corresponding surface horizons. Di *et al*. (1995) found fenamiphos to have a half-life in subsurface soil from two to more than four times the half-life that it had in the surface soil, probably because of the lower levels of microbial activity in the subsoil (Bromilow *et al*., 1986). However, another four pesticides were found to have shorter half-lives in the subsoil. The non-uniform trend that was detected in degradation rates with soil depth may be attributable partly to the dual role that soil organic matter plays in affecting microbial activity on the one hand, and pesticide sorption on the other (Rao *et al*., 1993). A decrease in the content of soil organic matter with soil depth may reduce soil microbial activity, and thus the pesticide degradation rate; however, a decrease in the content of soil organic matter also reduces pesticide sorption, which might result in an increase in the leaching rate (Hamaker, 1972). The pesticides that showed higher degradation rates in the subsoil were generally less water-soluble and had higher sorption coefficients.

Therefore, as the amount of organic matter in the subsoil decreases, the positive effect on the degradation rate due to decreased sorption might outweigh the negative effect on the degradation rate due to increased microbial activity, particularly for those pesticides with lower water solubility and a higher sorption coefficient (Di *et al*., 1995).

The rapid oxidation and hydrolysis of aldicarb in surface soils is associated with a high level of aerobic microbial activity. As the soil sampling depth increased, the amounts of aldicarb sulphoxide steadily decreased, especially in the case of samples that were taken from deeper than 150 cm (Ou *et al*., 1988). Considerable amounts of aldicarb sulphone were also detected in all surface samples, whereas negligible amounts of aldicarb sulphone were detected in all subsurface samples. The results confirmed that the oxidation and hydrolysis of aldicarb in surface soil occurred more rapidly than it did in the subsurface soil (Ou & Rao, 1986). A given pesticide's likelihood of reaching the groundwater is affected by the degradation rates in the subsurface, as well as in the surface layers, of the unsaturated soil zone (Bromilow *et al*., 1986; Pothuluri *et al*., 1990; Bergstrom, 1996).

## AGRICULTURAL PRACTICES LEADING TO THE DEVELOPMENT OF AMD

Pesticide degradation in soil is affected by both biotic and abiotic factors, which complement one another in the micro-environment (Singh *et al*., 2003). Repeated exposure of a soil microbial population to the same active ingredient often stimulates reproduction of the organisms that are able to degrade the chemical concerned, due to it being an energy or mineral nutrient source that is inaccessible to the other microorganisms that are present in the same soil (Dunn & Noling, 2003). Therefore, the degradation rate accelerates, and insufficient pesticide remains available to control the pest (Harrison, 1990).

The results of tests that were conducted by Read (1983) on field soils with a history of use of carbofuran strongly indicated that (a) repeated soil treatments of the insecticide for insect control can result in the selection of microorganisms that rapidly break down future application of the compound; (b) resting stages of the selected degradative microorganisms persist in the soil after a treatment of the carbofuran has been broken down; (c) the resting stages can survive prolonged periods of freezing, air-drying, or highly acidic conditions in the soil; and (d) populations can build up within a short time and rapidly degrade a new application of carbofuran, when adequate conditions of temperature, soil moisture and pH are provided.

Ou *et al*. (1994) conducted laboratory and field studies on the microbial degradation of fenamiphos in soils. They concluded that (a) the degradation of fenamiphos in soils was enhanced after one field application of the nematicide at 4.48 kg/ha, and that such enhancement lasted between three to four years; and (b) the half-lives for total toxic residue in soil were shortened after just one application of fenamiphos. However, most nematode-infested soils generally have been shown to require more than one treatment per year (Anderson, 1989). Data obtained by Anderson and Lafuerza (1992) indicated that, if fenamiphos were to be applied to the soil twice or three times per year, doing so would lead to a gradual increase in the rates of degradation of fenamiphos, which, in turn, would lead to a gradual decrease in the efficacy of fenamiphos. Data obtained by Ou (1991) indicated that the enhanced degradation of fenamiphos gradually declined between two and four years after a single field application of the chemical had been administered. The above suggests that the population of fenamiphos degraders in the field-treated soil gradually declined over a four-year period to the same level as was present before the application.

Cross-degradation of pesticides in soils is a problem of great concern (Chuang, 2000). 'Cross-conditioning' and 'cross-adaptation' are terms used to describe the accelerated breakdown of a chemical after repeated application of another chemical to the soil. The two chemicals involved usually are structurally related analogues (Felsot, 1989). In a study conducted by Anderson *et al*. (1998), *in situ* soil microorganisms that were adapted to degrading one organophosphate pesticide were indicated as being quite specific to that particular organophosphate pesticide. The exceptions to the rule in said case are structural analogues such as methylparathion and ethylparathion, isofenphos-methyl and -ethyl, or propyl and isobutyl ethoprophos. During the above-mentioned study, it was found that the soils adapted to degrade isofenphos-ethyl could also degrade isofenphos-methyl, and vice versa. However, neither the isofenphos-methyl-adapted soil nor the isofenphos-ethyl-adapted soil could degrade fenamiphos, prothiophos, ethrofolan or isazophos analogues at higher rates than the control rates.

Many studies have demonstrated that soil microorganisms that are adapted to degrade organophosphate pesticides do not attack carbamate pesticides at high rates (Racke & Coats 1988a, 1988b; Smelt *et al*., 1996). Similarly, soils that are adapted to degrade carbamates at accelerated rates have not been found to attack organophosphates (Racke & Coats, 1988a; Moorman, 1994). According to Anderson *et al*. (1998), cross-degradation reaction among organophosphates is rare. However, the cross-accelerated degradation of carbamate nematicides in soils is well known (Racke & Coats. 1988b). Previous studies have shown that soils that are adapted to degrade aldicarb could cause the accelerated degradation of oxamyl, and vice versa (Smelt *et al*., 1987). Similarly, soils that are adapted to degrade carbofuran can degrade aldicarb at accelerated rates and vice versa (Suett & Jukes, 1988; Suett, 1989).

## PREVENTION OF ACCELERATED MICROBIAL DEGRADATION

Microbial adaptations are natural processes used to rid the soil of applied chemicals that cannot be eliminated completely. Decreasing the degradation capacity to a point where the soil can again be treated safely with the same pesticide depends on the structure of the chemical used, as well as on sufficient application of it, the frequency of the treatment, climatic variables, and the ability of microorganisms to use parts, or all, of the chemical as nutrient and/or energy sources (Suett, 1986; Anderson *et al*., 1998). The best prescription for avoiding problems of accelerated degradation is to apply appropriate compatible pesticides and to use pesticides only when doing so becomes necessary (Suett, 1986; Harrison,

1990; Anderson *et al*., 1998). The sterilisation of machinery after use on a treated field would also minimise the danger of spreading the degrading microorganisms from one field to another (Read, 1986).

Felsot (1984) proposed two general kinds of strategies for coping with enhanced biodegradation: operational and technological. *Operational strategies* include the conservation of pesticides; crop and chemical rotation; the proper calibration of equipment; and the altered timing of applications. Pesticides are conserved by means of monitoring pest populations through scouting, soil sampling, and by applying pesticides only when economic thresholds are reached.

The use of appropriate agronomic practices can greatly reduce the need for pesticides. The prevalence of crop rotation systems in the United Kingdom may explain why few pest control problems have been associated with the enhanced biodegradation of pesticides in the country (Suett, 1986). *Technological strategies* require alterations in the formulation chemistry or the structural chemistry of the nematicide concerned. The alternatives include extenders and inhibitors, new formulation technology, and directed chemistry. Extenders and inhibitors are additives to pesticide formulations that improve residual bioactivity by preventing, or at least slowing down, biodegradation. Inhibitors, which are well known from enzymology, increase chemical persistence by means of inhibiting specific degradative enzymes. Extenders increase chemical persistence by means of imposing generalised adverse effects on soil microbial populations.

The concept of a threshold concentration that is necessary to stimulate microbial enrichment suggests that the significant lowering of initial rates of chemical applications could help to alleviate degradation problems (Alexander, 1985). The inability of low carbofuran concentrations (Chapman *et al*., 1986) and low-rate field applications of aldicarb (Suett & Jukes, 1988) to induce microbial enrichment supports such hypothesis. These operational strategies are the most desirable for the long-term management of biodegradation problems. They are more compatible with environmental goals, and require less research and development than do technological strategies. Furthermore, with the exception of chemical class rotations they can be implemented immediately (Felsot, 1984).

Rotation schemes with nematicides might avoid the problems that are otherwise likely to be present with enhanced biodegradation. Alternating an annual application of a pesticide with the application of another pesticide, which is not broken down by degrading microorganisms that are capable of degrading the first pesticide, can, at least, delay development, or help to maintain low populations of the degrading microorganisms in the soil (Read, 1986). Microorganisms that have been found to adapt to degrading the organophosphates isofenphos, prothiophos, terbufos and cadusafos, and the carbamates aldicarb and carbofuran, have been found not to degrade the organophosphate fenamiphos at high rates (Anderson *et al*., 1998). Therefore, the microorganisms that degrade fenamiphos are highly specific. No evidence of cross-degradation between fenamiphos and ethoprophos has been found to date (Anderson & Lafuerza, 1992; Smelt *et al*., 1996), indicating that the two organophosphate nematicides could be alternated successfully. The carbamate aldicarb can also be alternated with the organophosphate nematicides (Smelt *et al*., 1987), but the potential implication of cross-enhancement with other carbamates, like carbofuran (Suett & Jukes, 1988; Felsot, 1989; Chapman & Harris, 1990) and oxamyl (Smelt *et al*., 1987), must be considered. Alternating the use of carbofuran with a compound such as terbufos could reduce the hazard of developing degrading microorganisms (Read, 1986). Measures for controlling accelerated degradation therefore should form an integral part of the general strategy of crop and pesticide management.

Dunn and Noling (2003) suggest that, if turf managers should observe that the period of satisfactory control achieved with fenamiphos is much less than was experienced previously, the use of fenamiphos should be ceased immediately. A period of complete avoidance of fenamiphos for at least one year, and perhaps for a period of two years, might be necessary for the soil microbial population that is responsible for this phenomenon to revert back to normal rates of fenamiphos metabolism. Little information is available regarding the period required for nematicide degradation rates in previously treated soils to be restored to the rates in previously untreated soils. The results obtained (Read, 1986; Chapman & Harris, 1990; Suett *et al*., 1993) suggest that an interval of at least three, and preferably four, years should be left before carbofuran is reapplied to a previously treated soil. Ou (1991) reported that three to four years were necessary before the accelerated degradation rate of fenamiphos declined in a sandy soil in a temperate region. In tropical regions, fenamiphos degradation rates returned to levels found in previously untreated soils when sequential field treatments were interrupted for 12 to 16 months (Anderson & Lafuerza, 1992). In contrast, the accelerated degradation of the soil-applied pesticides carbofuran, aldicarb and oxamyl was found to persist for longer than five years (Suett *et al*., 1993; Smelt *et al*., 1996). It seems, therefore, that the duration of the accelerated degradation capacity of a soil might differ between soils, as well as between pesticides. There might be systematic differences between the organophosphate and carbamate pesticides (Chapman & Harris, 1990; Anderson & Lafuerza, 1992). Based on Johnson's (1998) results, fenamiphos application for the control of *M. incognita* should not exceed three consecutive years. Thereafter, the crop should be rotated and/or the nematicide should be changed. Similar results were previously reported with the use of ethoprop (Hall *et al*., 1988). For growers developing crop rotation schemes using alternating pesticide treatments, it is clearly important to quantify the differences in rates of recovery (Smelt *et al*., 1996).

Large concentrations at, and adjacent to, injection sites tend to kill nematodes within a few days of application, so that accelerated degradation seldom results in the complete failure of the treatment. Nevertheless, it can significantly reduce the effectiveness of some applications. Reduced efficacy can be overcome by increasing the dose rates used, and by covering the soil with plastic sheeting after treatment, although the increased costs of such treatments are only

economic viable with high-value crops. Thus, during the past decade, the phenomenon of accelerated transformation has led to a gradual increase in the doses of fumigants applied to such crops in the Netherlands (Suett *et al*., 1996). By altering the use of the two fumigants, which have no cross-adaptation, and by using resistant varieties, it is possible to limit the need for the use of each fumigant to no more than once in five years (Suett *et al*., 1996).

Racke and Coats (1988a) determined in their study that ethoprop has a greater persistence in soil than does terbufos. Read (1987) indicated that high concentrations of aldicarb apparently retard rapid degradation by the microbes. Read (1987) further indicated that the efficiency of aldicarb as a soil insecticide/nematicide is unlikely to be affected adversely by aldicarb-degrading microorganisms in acid soils with a pH of less than 5.6. Under such acidic conditions, however, the parent chemical can persist for long periods, and it can be oxidised to the highly water-soluble aldicarb sulphoxide metabolite. The soluble metabolite could then be eroded readily and leached into well water. In near-neutral acidic or in alkaline soils, aldicarb can be broken down by microorganisms to metabolites of lower toxicity (oximes and nitriles) more readily, and finally be mineralised into carbon dioxide ($CO_2$). Under such conditions, toxicity residues are less likely to be leached into groundwater or well water.

Read (1987) noticed that the application of the fungicide chlorothalonil (Bravo R) to aldicarb-treated soil at 50 mg/kg inhibited fungal growth and resulted in slight retardation in the rate of loss of aldicarb toxicity. The 100 mg/kg application resulted in a slightly longer persistence of aldicarb. High rates of 205, 500 and 1 000 mg/kg of chlorothalonil did not delay the breakdown of aldicarb for more than three to four days. Pesticide losses from photodegradation can be reduced by incorporating the pesticide into the soil during or immediately after application (Harrison, 1990).

Since the results obtained by Le Roux *et al*. (2001) in citrus orchards indicated that cadusafos becomes more prone to AMD when it is applied by means of drip irrigation rather than by means of broad application, it suggests that nematicides should rather not be applied by means of drip irrigation in order to prevent AMD from developing.

Studies on the genetic basis of carbofuran degradation by a bacterium reported by Karns *et al*. (1986), and those isolated elsewhere, will help to elucidate the manner in which the organisms evolved a new biodegradative function in response to the presence of a man-made chemical. Such studies might lead to the development of molecular probes for use in examining newly reported aggressive soils for the presence of organisms containing genes that are homologous to those found in organisms elsewhere. In such a manner it might be possible to trace the evolution and spread of degradative organisms (Karns *et al*., 1986).

## PREVENTING AMD IN VINEYARDS AND ORCHARDS

Currently, cadusaphos and fenamiphos are registered for use on grapevines and stone fruit. Plums are excluded from the use of fenamiphos, while oxamyl is registered for use on stone fruit only. Aldicarb is in the process of being phased out by 2014, and is no longer commercially available in South Africa (Table 1).

Notably, the soil application of a nematicide should only be curative, and not preventative. To determine the need for a nematicidal application, a representative soil and root sample from a vineyard or orchard should be taken and analysed for the occurrence of economically important nematodes (Kleynhans *et al*., 1996). Should the numbers of a specific nematode, or of a nematode combination, exceed the threshold damage level, a registered nematicide (Table 1) should be applied at the recommended dosage, as prescribed on the label. Within a period of six months, a second representative soil sample should again be drawn and, if the results indicate the need, another nematicide application should be done. The most important factor to bear in mind is that the same nematicide should never be used consecutively, as doing so would be likely to lead to cross-degradation.

If the occurrence of AMD against a nematicide is suspected in agricultural soil, a biological soil test is available for confirming the presence of high populations of the specific microbial complex that is responsible for utilising the active ingredients of organophosphates and carbamates as a carbon source. For this test, a nematode, *Aphelenchus avenae* Bastian, 1865 is commercially used to test for nematicidal activity in the soil. This nematode feeds on a wide variety of fungi and, when the numbers of the nematode increase dramatically in a suspected soil sample compared to in the control (untreated soil), it is assumed that the microbial populations responsible for AMD are present in the soil (Stirling *et al*., 1992).

From the information that is available in the literature, it can be surmised that the microbial complex build-up of organophosphate nematicides in the soil is very specific for particular molecules, meaning that different organophosphates can be alternated with each other. In contrast, however, carbamate nematicides are not molecule specific and different carbamate nematicides cannot be used alternatively. Carbamate nematicides should only be alternated with an organophosphate nematicide, or with another chemical with a different molecular structure. Many studies have also shown that accelerated degradation with carbamate nematicides can develop rapidly in soils with a pH higher than 6.5. If the microbial populations have established themselves in the soil, they might last for up to two years in soils with a low pH, and for longer than five years in soils with a high pH. In the case of organophosphate nematicides, the soil tends to recover more quickly at a pH below 6, and more slowly at a higher pH. Generally, soil will recover within a period of 12 to 18 months before the same organophosphate can be used again with good nematicidal results and persistence (Smelt *et al*., 1987; Racke & Coats, 1988b; Suett & Jukes, 1988).

In the case of grapevines, only fenamiphos and cadusaphos are available for alternative use in established vineyards. Based on the literature, organophosphates can be used alternatively, since the same microorganisms that utilised one organophosphate do not tend to utilise another organophosphate (Anderson *et al*., 1998). According to Storey (2007), yearly treatment against ring and dagger nematode in South African grapevines is insufficient, as these

nematodes are difficult to control under local conditions. Should a third application be necessary, a totally different registered biological or chemical compound option should be considered for use, if such an option is available. There is not yet evidence of cross-degradation between carbamate and organophosphate nematicides in the literature, but a one-to-one alteration programme of fenamiphos with any of the organophosphate nematicides is not recommended (Anderson & Lafuerza, 1992).

In the case of pome fruit, with only oxamyl being registered for treatment, no alternative option with regard to organophosphate and carbamate nematicides is available. A totally different registered nematicide should then be considered for alteration if a second application is needed within a six-month period, in a one-to-one alteration. According to Anderson *et al*. (1998), in this type of alternation, soils should be monitored routinely for the development of biodegradation.

For stone fruit, if fenamiphos is used as the first application, the second application can be either cadusaphos or oxamyl. In the case of carbamates, different carbamates cannot be used alternatively, since microorganisms that degrade one carbamate will also degrade all others. For stone fruit, for example, aldicarb and oxamyl should not be used alternatively, as the same microorganisms that are responsible for the breakdown of aldicarb are also responsible for the breakdown of oxamyl.

The scope of organophosphate and carbamate nematicides for the control of plant-parasitic nematodes is very small, and if these products are not used responsibly they might have a serious impact on humans, animals, the environment and the sustainability of agriculture. It is of vital importance to prevent the occurrence of AMD, so as not to jeopardise the use of these nematicides in the long run. This will only be achieved if all the producers from the different industries maintain the highest level of responsibility and safety when using either nematicides or any other pesticides.

## LITERATURE CITED


Acosta-Martinez, V. & Tabatabai, M.A., 2000. Enzyme activities in a limed agricultural soil. Biol. Fert. Soils 31, 85-91.

Addison, P. & Fourie, J.C., 2007. Cover crop management in vineyards of the Lower Orange River region, South Africa: 2. Effect on plant parasitic nematodes. S. Afr. J. Enol. Vitic. 29, 26-32.

Alexander, M., 1985. Biodegradation of organic chemicals. Environ. Sci. Technol. 18, 106-111.

Anderson, J.P.E., 1989. Accelerated microbial degradation of nematicides and other plant-protection chemicals in soils. Nematropica 19, 1.

Anderson, J.P.E. & Lafuerza, A., 1992. Microbial aspects of accelerated pesticide degradation in soils. In: Anderson, J.P.E., Arnold, D.J.F., Lewis, F. & Torstensson, L. (eds). Proc. Internat. Symp. on Environ. Aspects of Pesticide Microbiol., Sigtuna, Sweden. Available: http://www.bj.refer.org/benin_ct/edu/isysphyt/english/indust/bayer/amd98.htm (accessed 25 May 2013).

Anderson, J.P.E., Nevermann, K. & Haidt, H., 1998. Accelerated degradation of nematicides in soils: Problem and its management. In: Hidalgo, L. (ed). Proc. XIII Acorbat Meeting Ecuador, November 1998, Guayaquil, Ecuador. pp. 568 – 579.

Arbeli, Z. & Fuentes, C.L., 2007. Accelerated biodegradation of pesticides: An overview of the phenomenon, its basis and possible solutions; and a discussion on the tropical dimension. Crop Prot. 26, 1733-1746.

Bergstrom, L., 1996. Model predictions and field measurements of chlorsulfuron leaching under non-steady-state flow conditions. Pestic. Sci. 48, 37-45.

Bilkert, J.M. & Rao, P.S.C., 1985. Sorption and leaching of three nonfumigant nematicides in soils. J. Environ. Sci. Health B20, 1-26.

Blumhorst, M.R., 1996. Experimental parameters used to study pesticide degradation in soil. Weed Technol. 10, 169-173.

Bromilow, R.H. & Leistra, M., 1980. Measured and simulated behaviour of aldicarb and its oxidation products in fallow soils. Pestic. Sci. 11, 389-395.

Bromilow, R.H., Baker, R.J., Freeman, M.A.H. & Görög, K., 1980. The degradation of aldicarb and oxamyl in soil. Pestic. Sci. 11, 371-378.

Bromilow, R.H., Briggs, G.G., Williams, M.R., Smelt, J.H., Tuinstra, L.G.M.T. & Traag, W.A., 1986. The role of ferrous ions in the rapid degradation of oxamyl, methomyl and aldicarb in anaerobic soils. Pestic. Sci. 17, 535-547.

Bull, D.L., Stokes, R.A., Coppedge, J.R. & Ridgway, R.L., 1970. Further studies on the fate of aldicarb in soil. J. Econ. Entomol. 63, 1283-1289.

Cain, R.B. & Head, I.M., 1991. Enhanced degradation of pesticides: Its biochemical and molecular biological basis. In: Walker, A. (ed). Pesticides in soils and water: Current perspectives. BCPC Monograph No. 47, British Crop Protection Council, Farnham. pp. 23 – 40.

Chapman, R.A. & Harris, C.R., 1990. Enhanced degradation of insecticides in soil: Factors influencing the development and effects of enhanced microbial activity. In: Racke, K.D. & Coats, J.R. (eds). Enhanced biodegradation of pesticides in the environment. American Chemical Society, Washington, DC. pp. 82 – 96.

Chapman, R.A., Harris, C.R. & Harris, C., 1986. Observations on the effect of soil type, treatment intensity, insecticide formulation temperature and moisture on the adaptation and subsequent activity of biological agents associated with carbofuran degradation in soil. J. Environ. Sci. Health, part B 17, 355-361.

Chuang, C., 2000. Enhanced degradation of pesticides. Environ. Eng. Res. 1, 47-61.

Coakley, S.M., Scherm, H. & Chakraborty, S., 1999. Climate change on plant disease management. Ann. Rev. Phytopathol. 37, 399-426.

Danielson, L.L, Gentner, W.A. & Jansen, L.L., 1961. Persistence of soil-incorporated EPTC and other carbamates. Weed Sci. 9, 463-476.

Di, H.J., Kookana, R.S. & Aylmore, L.A.G., 1995. Application of a simple model to assess the ground water contamination potential of pesticides. Aus. J. Soil Res. 33, 1031-1040.

Dickinson, C.H. & Pugh, G.J.F., 1974. Biology of plant litter decomposition, vol 1. Academic Press, London.

Dunn, A.R. & Noling, J.W., 2003. Characteristics of principal nematicides. RFNG009, Entomology and Nematology Department, Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida, Gainesville, FL.

Edwards, C.A., 1973. Biological aspects of the degradation and behaviour of pesticides in soil. In: Gladstone, M.N. (ed). Proc. 7th British Insecticide Fungicide Congress, British Crop Protection pp. 811-823.

Felsot, A.S., 1984. Coping with enhanced biodegradation of agricultural chemicals. Presented at 187th National Meeting of the American Chemical Society, St Louis, MO, April 1-13.

Felsot, A.S., 1989. Enhanced biodegradation of insecticides in soil: Implications for agroecosystems. Ann. Rev. Entomol. 34, 453-476.

Felsot, A. & Dahm, P.A., 1979. Sorption of organophosphorus and carbamate insecticides by soil. J. Agric. Food Chem. 27, 557-563.

Felsot, A.S., Maddox, J.V. & Bruce, W., 1981. Enhanced microbial degradation of carbofuran in soils with histories of carbofuran use. B. Environ. Contam. Tox. 26, 781-788.

Felsot, A.S., Wilson, J.G., Kuhlman, D.E. & Steffey, K.L., 1982. Rapid dissipation of carbofuran as a limiting factor in corn rootworm control in fields with histories of continuous Furadan® use. J. Econ. Entomol. 75, 1098-1103.

Fomsgaard, I. & Albaiges, J., 1995. Degradation of pesticides in subsurface soils, unsaturated zone, a review of methods and results. Int. J. Environ. Analyt. Chem. 58, 231-245.

Getzin, L.W., 1973. Persistence and degradation of carbofuran in soil. Environ. Entomol. 2, 461-467.

Hall, M.R., Johnson, A.W. & Smittle, D.A., 1988. Nematode population densities and yield of sweet potato and onion as affected by nematicides and time of application. Ann. Appl. Nematol. 2, 15-21.

Hamaker, J.W., 1972. Decomposition: Quantitative aspects. In: Goring, C.A.I. & Hamaker, J.W. (eds). Organic chemicals in the soil environment, vol 1. Marcel Dekker, New York. pp. 253 – 340.

Harrison, S.A., 1990. The fate of pesticides in the environment. Adapted from "The fate of pesticides in the environment and groundwater protection," by C.L. Brown and W.K. Hock. Agrochemical Factsheet #8, Penn State Cooperative, USA Extension.

Hawksworth, D.L., Kirk, P.M., Sutton, B.C. & Pegler, D.N., 1995 (8th ed). Ainsworth & Bisby's dictionary of the fungi. International Mycological Institute, CAB International, Wallingford, UK.

Haydock, P.P.J., Ambrose, E.L., Wilcox, A. & Deliopoulos, T., 2012. Degradation of the nematicide oxamyl under field and laboratory conditions. Nematology 14, 339-352.

Johnson, A.W., 1998. Degradation of fenamiphos in agricultural production soil. J. Nematol. 30, 40-44.

Johnson, A.W., Rohde, W.A., Dowler, C.C., Glaze, N.C. & Wright, W.C., 1981. Influence of water and soil temperatures on the concentration and efficacy of phenamiphos or control of root-knot nematodes. J. Nematol. 13, 148-153.

Jones, A.S., 1976. Metabolism of aldicarb by five soil fungi. J. Agric. Food Chem. 24, 115-117.

Jones, R.L. & Norris, F.A., 1998. Factors affecting degradation of aldicarb and ethoprop. J. Nematol. 30, 45-55.

Karns, J.S., Mulbry, W.W., Nelson, O.J. & Kearny, P.C., 1986. Metabolisms of carbofuran by a pure bacterial culture. Pestic. Biochem. Physiol. 25, 211-217.

Karpouzas, D.G., Karanasios, E. & Menkissoglu-Spiroudi, U., 2004. Enhanced microbial degradation of cadusafos in soils from potato monoculture: Demonstration and characterization. Chemosphere 56, 549-559.

Kleynhans, K.P.N., Van den Berg, E., Swart, A., Marais, M. & Buckley, N.H. 1996. Plant nematodes in South Africa. Handbook No. 8, Plant Protection Research Institute, Biosystematics Division, Pretoria.

Kuseske, D.W., Funke, B.B. & Schultz, J.T., 1974. Effects and persistence of Baygon (Propoxur) and Temik (Aldicarb) insecticides in soil. Plant Soil 41, 255-269.

Laveglia, J. & Dahm, P.A., 1977. Degradation of organophosphorus and carbamate insecticides in the soil and by soil microorganisms. Ann. Rev. Entomol. 22, 483-513.

Leistra, M., Bromilow, R.H. & Boesten, J.J.T.I., 1980. Measured and simulated behaviour of oxamyl in fallow soil. Pesticide Sci. 11, 379-388.

Leonard, R.A., Shirmohammadi, A., Johnson, A.W. & Marti, L.R., 1998. Pesticide transport in shallow groundwater. Trans. Am. Soc. Agric. Engineers 31, 776-788.

Le Roux, H., Pretorius, M.C. & Huisman, L. 2001. Accelerated degradation of nematicides used on citrus in South Africa. How Degrading, no 3, 1.

Mackiewicz, M., Deubert, K.H., Gunner, H.B. & Zuckerman, B.M., 1969. Study of parathion biodegradation using gnotobiotic techniques. J. Agric. Food Chem. 17, 129-130.

Moorman, T.B., 1994. Pesticide degradation by soil microorganisms: Environmental, ecological, and management effects. In: Hatfield, J.L. & Stewart, B.A. (eds). Soil biology: Effects on soil quality. CRC (Lewis), Boca Raton, Ann Arbor, MI. pp. 121 – 165.

Nelson, L.M., Yaron, B. & Nye, P.H., 1982. Biologically-induced hydrolysis of parathion in soil: Kinetics and modelling. Soil Biol. Biochem. 14, 223-227.

Orchard, V.A. & Cook, F.J., 1983. Relationship between soil respiration and soil moisture. Soil Biol. Biochem. 15, 447-453.

Ou, L.T., 1991. Interaction of microorganisms and soil during phenamiphos degradation. Soil Sci. Soc. Am. J. 55, 716-722.

Ou, L.T., Gancarz, D.H., Wheeler, W.B., Rao, P.S.C. & Davidson, J.M., 1982. Influence of soil temperature and soil moisture on degradation and metabolism of carbofuran in soils. J. Environ. Qual. 11, 293-298.

Ou, L.T. & Rao, P.S.C., 1986. Degradation and metabolisms of oxamyl and phenamiphos in soils. J. Environ. Sci. Health B21, 25-40.

Ou, L.T., Rao, P.S.C., Edvardsson, K.S.V., Jessup, R.E., Hornsby, A.G. & Jones, R.L., 1988. Aldicarb degradation in sandy soils from different depths. Pestic. Sci. 23, 1-12.

Ou, L.T., Sture, K., Edvardsson, V., Thomas, J.E. & Rao, P.S.C., 1985. Aerobic and anaerobic degradation of aldicarb sulfone in soils. J. Agric. Food Chem. 33, 545-548.

Ou, L.T. & Thomas, J.E., 1994. Influence of soil organic matter and soil surfaces on a bacterial consortium that mineralises fenamiphos. Soil Sci. Soc. Am. J. 58, 1148-1153.

Ou, L.T., Thomas, J.E. & Dickson, E.W., 1994. Degradation of fenamiphos in soil with a history of continuous fenamiphos applications. Soil Soc. Am. J. 58, 1139-1147.

Pothuluri, J.V., Moorman, T.B., Obenhuber, D.C. & Wauchope, R.D., 1990. Aerobic and anaerobic degradation of alachlor in samples from a surface-to-groundwater profile. J. Environ. Qual. 19, 525-530.

Racke, K.D. & Coats, J.R., 1988a. Comparative degradation of organophosphorus insecticides in soil: Specificity of enhanced microbial degradation. J. Agric. Food Chem. 36, 193-200.

Racke, K.D. & Coats, J.R., 1988b. Enhanced degradation and the comparative fate of carbamate insecticides in soil. J. Agric. Food Chem. 36, 1067-1072.

Rajagopal, B.S., Rao, V.R., Nagendrappa, G. & Sethunathan, N., 1984. Metabolism of carbaryl and carbofuran by soil-enrichment and bacterial cultures. Can. J. Microbiol. 30, 1458-1466.

Ramanand, K., Panda, S., Sharmila M., Adhya, T.K. & Sethunathan, N., 1988a. Development and acclimatization of carbofuran-degrading soil enrichment cultures at different temperatures. J. Agr. Food Chem. 36, 200-205.

Ramanand, K., Sharmila, M. & Sethunathan, N., 1988b. Mineralization of carbofuran by a soil bacterium. Appl. Environ. Microbiol. 54, 2129-2133.

Rao, P.S.C., Bellin, C.A., Brusseau, M.L. & Linn, D.L., 1993. Coupling biodegradation of organic chemicals to sorption and transport in soils and aquifers: Paradigms and paradoxes. In: Chang, F.H. (ed). Proc. Sorption and Degradation of Pesticides and Organic Chemicals in Soil, October 1991, Denver, CO, 30. SSSA, Madison, WI. pp. 1 – 26.

Read, D.C., 1983. Enhanced microbial degradation of carbofuran and fensulfothion after repeated applications to acid mineral soil. Agr. Ecosyst. Environ. 10, 37-46.

Read, D.C., 1986. Accelerated microbial breakdown of carbofuran in soil from previously treated fields. Agric. Ecosyst. Environ. 15, 51-64.

Read, D.C., 1987. Greatly accelerated microbial degradation of aldicarb in re-treated field soils in flooded soil, and in water. J. Econ. Entomol. 80, 156-163.

Rohde, W.A., Johnson, A.W., Dowler, C.C. & Glaze, N.C., 1980. Influence of climate and cropping patterns on the efficacy of ethoprop, methyl bromide, and DD-MENCS for the control of root-knot nematodes. J. Nematol. 12, 33-39.

Schuldt, P.H., Burchfield, H.P. & Bluestone, H., 1957. Stability and movement studies on the new experimental nematicide 3,4-dichlorotetra-hydrothiophene-1,1-dioxide in soil. Phytopath. 17, 534 (Abst.).

Serdar, C.M., Gibson, D.T., Munnecke, D.M. & Lancaster, J.H., 1982. Plasmid involvement in parathion hydrolysis by *Pseudomonas diminuta*. Appl. Environ. Microbiol. 44, 246-249.

Sethunathan, N. & Yoshida, T., 1973. A *Flavobacterium* sp. that degrades diazinon and parathion. Can. J. Microbiol. 19, 873-875.

Sheela, S. & Pai, S.B., 1983. Metabolism of fensulfothion by a soil bacterium *Pseudomonas alcaligenes* $C_1$. Appl. Environ. Microbiol. 19, 873-875.

Siddaramappa R., Tirol, A.C., Seiber, J.N., Heinrichs, E.A. & Watanabe, I. 1978. The degradation of carbofuran in paddy water and flooded soil of untreated and retreated rice fields. J. Environ. Sci. Health B13, 369-380.

Singh, B.K., Walker, A., Morgan, J.A.W. & Wright, D.J., 2003. Role of soil pH in the development of enhanced biodegradation of fenamiphos. Appl. Environ. Microbiol. 69, 7035-7043.

Smelt, J.H., Crum, S.J., Teunissen, W.J. & Leistra, M., 1987. Accelerated transformation of aldicarb, oxamyl and ethoprophos after repeated soil treatments. Crop Prot. 6, 295-303.

Smelt J.H., Dekker, A. & Leistra, M., 1979. Effect of soil moisture condition on the conversion rate of oxamyl. Netherlands J. Agric. Sci. 27, 191-198.

Smelt, J.H., Dekker A., Leistra, M. & Houx, N.W.H., 1983a. Conversion of four carbamoyloximes in soil samples from above and below the soil water table. Pestic. Sci. 14, 173-181.

Smelt, J.H., Leistra, M., Dekker, A. & Schut, C.J., 1981a. Movement and conversion of ethoprophos in soil in winter: 1. Measured concentration patterns and conversion rates. Soil Sci. 131, 242-248.

Smelt, J.H., Leistra, M., Houx, N.W.H. & Dekker, A., 1978. Conversion rates of aldicarb and its oxidation products in soils. III. Aldicarb. Pestic. Sci. 9, 293-300.

Smelt, J.H., Schut, C.J. & Leistra, M., 1983b. Movement and conversion of aldicarb and its oxidation products in columns of grassed and fallow soil. J. Environ. Sci. Health B18, 645-665.

Smelt, J.H., Schut, C.J., Dekker, A. & Leistra, M., 1981b. Movement and conversion of aldicarb and its oxidation products in columns of grassed and fallow soil. J. Environ. Sci. Health B18, 645-665.

Smelt, J.H., Van de Peppel-Groen, A.E. & Leistra, M., 1995. Transformation of aldicarb sulfoxide and aldicarb sulfone in four water-saturated sandy subsoils. Pestic. Sci. 44, 323-334.

Smelt, J.H., Van de Peppel-Groen, A.E., Van der Pas, L.J.T. & Dijksterhuis, A., 1996. Development and duration of accelerated degradation of nematicides in different soils. Soil Biol. Biochem. 28, 1757-1765.

Stirling, A.M., Stirling, G.R. & Macrae, I.C., 1992. Microbial degradation of fenamiphos after repeated application to tomato-growing soil. Nematologica 38, 245-254.

Storey, S.G., 2007. Nematodes in vines: Practical guidelines for the short- and long-term control of nematodes. Wineland 60-63.

Suett, D.L., 1986. Accelerated degradation of carbofuran in previously treated field soils in the United Kingdom. Crop Prot. 5,165-169.

Suett, D.L., 1989. Accelerated degradation of soil-applied insecticides: Implications for insect pest control. Mede. Fac. Landbouwweten. Rijksuniv. Gent 54, 983-991.

Suett, D.L. & Jukes, A.A., 1988. Accelerated degradation of aldicarb and its oxidation products in previously treated soils. Crop Prot. 7, 147-152.

Suett, D.L., Jukes, A.A. & Parek, H., 1996. Non-specific influence of pH on microbial adaptation and insecticide efficacy in preciously treated field soils. Soil Biol. Biochem. 28, 1783-1790.

Suett, D.L., Jukes, A.A. & Phelps, K., 1993. Stability of accelerated degradation of soil-applied insecticides: Laboratory behaviour of aldicarb and carbofuran in relation to their efficacy against cabbage root fly (*Delia radicum*) in previously treated field soils. Crop Prot. 12, 431-442.

Sutherland, T.D., Horne, I., Lacey, M.J., Harcour, T.R., Russell, R.J. & Oakeshott, J.G., 2000. Enrichment of and endosulfan-degrading mixed bacterial culture. Appl. Environ. Microbiol. 66, 2822-2828.

Talekar, N.S., Sun, L.T., Lee, E.M. & Chen, J.S., 1977. Persistence of some insecticides in subtropical soil. J. Agric. Food Chem. 25, 348-352.

Waggoner, T.B. & Khasawinah, A.M., 1974. New aspects of organophosphorus pesticides. VII. Metabolism, biochemical, and biological aspects of Nemacur and related phosphoramidate compounds. Residue Rev. 53, 79-97.

Williams, I.H., Pepin, H.S. & Brown, M.J., 1976. Degradation of carbofuran by soil microorganisms. B. Environ. Contam. Tox. 15, 244-248.

Zuckerman, B.M., Deubert, K., Mackiewicz, M. & Gunner, H., 1970. Studies on the biodegradation of parathion. Plant Soil 33, 273-283.