

**EUONYMUS SCALE, *UNASPIS EUONYMI* (COMSTOCK): HOST
PREFERENCE, AND DISTRIBUTION OF NATIVE AND IMPORTED
NATURAL ENEMIES IN VIRGINIA**

by

David Kirk Jefferson

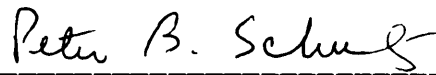
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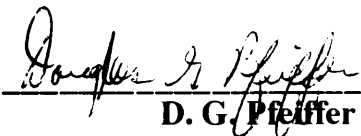
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by
David Kirk Jefferson
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Entomology
(Abstract)

Studies were conducted in 1992 and 1993 to determine which of six *Euonymus* species and cultivars would sustain the highest populations of *Unaspis euonymi* (Comstock), as a food source for *Chilocorus kuwanae* (Silvestri). Analysis of data collected from field-grown plants and container-grown plants indicates that *Euonymus japonicus* (Thunb.) 'Albo-marginatus' would sustain the highest populations of *U. euonymi*.

In 1992 and 1993, surveys were conducted to determine the presence of existing natural enemies in Virginia as well as to determine the establishment of the imported predator, *Chilocorus kuwanae*. In 1992, *C. kuwanae* and parasites from the genera *Aspidiotiphagus* and *Encarsia* was found at several locations in Virginia Beach. In 1993, *C. kuwanae* was recovered in seven of the twelve Virginia counties surveyed. Either *Aspidiotiphagus* or *Encarsia* were found in nine of the twelve counties. A single specimen of *Cybocephalus* sp. was recovered from a sample taken in Fredericksburg.

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Chapter 1

Introduction and Literature Review

Euonymus in the ornamental landscape

Euonymus ranks among the most commonly used ornamental landscape plants. It exhibits a wide variety of growth habits, including ground covers, climbing vines, and both evergreen and deciduous shrubs. *Euonymus* is one of 55 genera belonging to the family Celastraceae. Most of the 176 species belonging to the family are Asian in origin (Dirr 1983).

Euonymus spp. are popular in the ornamental landscape for several reasons. The deciduous species add color to the landscape with their fruit and colorful fall foliage. *Euonymus japonicus* (Thunb.) and *E. fortunei* (Turcz.) have been cultivated to produce variegated forms that have both attractive leaf color and texture. *Euonymus* spp. can withstand a wide range of soil conditions, although they perform better in a well-drained loam. *Euonymus* spp. also propagate easily from cuttings (Lancaster 1981).

There has been a declining use of *Euonymus* spp. in landscapes because of infestation by euonymus scale, *Unaspis euonymi* (Comstock) (Homoptera: Diaspididae) (Brewer & Oliver 1984). Raupp et al. (1985) found that in Maryland, 68% of the euonymus plants at 150 home sites had problems with *U. euonymi* infestation. In addition to draining plant sap from the host, feeding diaspidids inject a toxin with their saliva (Kosztarab 1990). Symptoms of heavy feeding by *U. euonymi* include necrotic and chlorotic foliage followed by leaf abscission, branch necrosis and eventual plant death. These symptoms are caused by enzymatic action of the saliva of the scale insect on the host

plant. *Unaspis euonymi* infestations generally remain unnoticed until populations have reached injurious levels.

Previous control methods of *U. euonymi* have included lime sulfur applied during plant dormancy or wettable sulfur applied in early spring. Dormant oil has largely replaced lime sulfur. Organophosphate insecticides have also proven useful in euonymus scale control (Davidson & Lyon 1987). Dennis (1969) reported that of the insecticides tested, only diazinon, dimethoate, and phorate had any significant effect. Efficacy was improved by the addition of oil. Dennis (1969) also noted that no effective method of biological control had been found .

Conventional control of *U. euonymi* with insecticides has been generally unsuccessful for several reasons. With exception of first instars and adult males, all life stages of the scale are covered with an oystershell-like test. This test keeps the pesticide from reaching the scale insect (Cohen et al. 1987). Additionally, females oviposit for an extended period (up to 60 days), resulting in an overlap of developmental stages. Plant coverage with insecticides is difficult to achieve since *U. euonymi* develops on all surfaces of the plant. *Euonymus* spp. are being eliminated from nursery production as well as established landscapes because of the difficulty in controlling *U. euonymi*. Therefore, to keep the desirable species of *Euonymus* in production and in the landscape, a suitable method of *U. euonymi* control must be found.

Biology and Ecology of *Unaspis euonymi* (Comstock) (Homoptera: Diaspididae)

Comstock first described *U. euonymi* as *Chionaspis euonymi*, from a sample submitted from Norfolk, VA in 1881 (Comstock 1881). Ferris (1937) placed *euonymi* in the genus *Unaspis* and revised the description .

Female *U. euonymi* have three instars while the males have five. The scale generally has two generations per year in the northern United States, including Virginia, and three in the south (Underhill 1943). The time required for a generation to mature varies from one generation to the next, and from one climatic zone to another. In Virginia, the first generation requires 6 weeks to mature and the second requires 4-5 weeks. In Virginia, the number of eggs laid per female varies from 60 to 157, with an average of 98 (Chapman et al. 1931). Emergence of the first instars (crawlers) occurs soon after eggs are laid. Crawler emergence in Virginia has been reported to be in early May for the first generation and mid July for the second (Underhill 1943).

The crawlers move around on the host for a few hours before settling. Since they are positively phototactic, their movement is generally upwards toward new shoot growth. The females settle primarily on the stems while the males mostly settle on the leaves (Cockfield & Potter 1990). Differences in site preference may reflect differential risks in mortality. Leaf abscission can be an important source of mortality in sedentary leaf feeders. Male scale insects do not feed after the second instar. Furthermore, alate males could possibly emerge from abscised leaves and fly to the females to mate. More than 50% of the leaves in heavily infested field plants may drop during the

winter, resulting in high mortality for any leaf-dwelling females (Cockfield & Potter 1990).

Although the spread of economically important diaspidids has been primarily through the transport of infested host plants, there are naturally occurring factors that influence scale dispersal. There are two zones of influence that affect the performance of scale populations: non-host plant or ambient conditions, and host plant conditions such as morphology and physiology (McClure 1990). "Ambient conditions" refers to climate and microclimate and includes temperature and relative humidity. Temperature has a great effect on scale performance and reproduction. For example, California red scale, *Aonidella aurantii* (Maskell), has up to five generations per year in warm dry climates such as that of Queensland, Australia. However, in the cool, coastal regions of California, California red scale averages two generations per year (Beardsley & Gonzalez 1975). Low winter temperatures affect the overwintering life stage. In central Europe, San Jose scale, *Quadraspidotus perniciosus* (Comstock) overwinters as first instars in the black cap phase of development. (Beardsley & Gonzalez 1975). In the southeastern United States San Jose scale overwinters as immature males and females (Johnson & Lyon 1988). Summer diapause occurs in purple scale, *Lepidosaphes beckii* (F.) on citrus grown in the Mediterranean region. These diapausing females resume normal activity in the fall (Beardsley & Gonzalez 1975).

Ambient, abiotic factors affect crawlers as well as adults. Extremes in relative humidity as well as in temperature can influence crawler emergence, dispersal and settling. High humidity can delay emergence and slow dispersal

(Greathead 1990). Low humidity will result in desiccation of the crawlers. Temperature can determine settling sites on a particular host plant. Florida red scale, *Chrysomphalus aonidum* (L.) will settle on the cooler shaded parts of trees during the summer months and on the sunny parts during the cooler seasons (Beardsley & Gonzalez 1975). What appears to be a negatively phototactic response may actually be a response to low humidity and high temperature. However, light does play a role in crawler dispersal. Crawler emergence occurs mostly in the morning; the positively phototactic crawlers will move upwards toward the light and onto new growth (Greathead 1972). Wind affects both mortality and dispersal of scale insects. Greathead (1972) demonstrated that air currents can dislodge and deposit crawlers in sufficient quantities downwind to establish new infestations. Nevertheless, not all crawlers are deposited on a suitable host.

In addition to ambient or atmospheric conditions, there are several external and internal host factors that affect scale performance. External factors include substrate texture, color and host growth-habit. Internal factors include nutrients available to the scale and the presence of alleochemicals (Beardsley & Gonzalez 1975).

Color of the substrate affects crawler wandering time. Crawlers of *A. aurantii* wander longer on a yellow surface than they do on a green surface (Willard 1973). The crawlers of many scale species exhibit thigmotaxis, a tendency to settle adjacent to surface irregularities or in leaf axils or other crevices. Surface dust also will induce a thigmotactic response.

Scales and other phloem feeding insects such as mealybugs (Homoptera: Pseudococcidea) and aphids (Homoptera: Aphididae), utilize amino acids and

amides found in host phloem as a primary source of dietary nitrogen (McClure 1979, Sadof & Raupp 1991). Nitrogen levels in the phloem can be increased by increasing soil fertility or by changes in host physiology. Variegation in plant tissue elevates available nitrogen levels (Sadof & Raupp 1991). Plants will increase nitrogen in the phloem in response to environmental stress. Temperature extremes, disease, physical injury, pollutants, pesticides and herbivory are all sources of environmental plant stress. Although herbivory will initially result in an increased flow of metabolic nitrogen, it will result in a net decrease in nutrients and subsequent generations of scale will not perform as well as earlier populations (McClure 1979).

Position of the host plant in the landscape also influences scale performance. *Euonymus* spp. grown in protected portions of the ornamental landscape have higher scale densities than those in unprotected areas (Brewer & Oliver 1984). Kosztarab (personal communication 1992) postulated that the protected parts of a landscape are regions of higher plant metabolic stress. Plants adjacent to buildings receive protection from wind and rain. This protection prevents rain from reaching the plants roots in addition to washing off accumulated dust on foliage and stems. Furthermore, buildings are a source of reflected as well as radiated heat.

Sampling and Census Taking

Sampling is important, not only in the study of insect ecology and population dynamics, but also in the pest management decision-making process. There are essentially two kinds of population estimating methods, absolute and relative. Absolute estimates measure actual numbers of insects

and are expressed in terms of actual numbers of insects, i.e., population densities. Absolute estimates are useful in researching population dynamics. However, because of the cost involved, they are not often used in pest management decision-making. Relative estimates are used to compare populations over time and space. Since the comparison is relative, the same sampling procedure must be used for each comparison. Population indices may also be used as a relative estimate of the population. A population index is a measure of insect by-product; for example, the amount of crop injury or the number of frass pellets, etc. (Pedigo 1989).

Sampling systems should be capable of gathering information about a complex of species, as well as a single species within a complex of species. They should also have the sensitivity to measure pest density or injury potential at levels at and below the economic threshold or injury level (Westigard & Calvin 1977).

Kozár (1990) reviewed several sampling techniques used by Hungarian and Russian authors. A summary of that review is presented here. Field or orchard distribution sampling methods vary greatly. Several methods of scale sampling have been developed. However, the methods chosen by investigators have often been arbitrary and in accordance with their purposes. Various kinds of methods are discussed, including the examination of 10 trees per orchard. Some authors suggest walking through the orchard at diagonals or examining five alternate trees. Kozár cited one of his own studies wherein 10 cm branch sections taken from four sides of a tree were used as the sampling unit. In a study of sampling of *Q. perniciosus*, Westigard and Calvin (1977) removed

fruit spurs from the top 5 m and the lower 2 m of pear trees. Every tree in the orchard was sampled.

Gulmahamad and DeBach (1978) found populations of *Q. perniciosus* to be contiguously distributed. Both interplant and intraplant variations were very large. One or two limbs of a host plant might be infested while the rest of the plant remained free of scale. Therefore, the sampling was restricted to infested branches. The situation found by Gulmahamad and DeBach is similar to what would be expected in euonymus scale infestations in the ornamental landscape. *Euonymus* spp. are not always planted adjacent to each other nor do they all come from the same source. Therefore not all the *Euonymus* spp. in an ornamental landscape would have an equal chance of becoming infested with scale.

Kozár (1990) also discussed scoring or methods of evaluating percentage of scale infestation. This evaluation is done by assigning a score to a specific population density. For example, Borchsenius (1950) assigned scores based on population densities ranging from scattered specimens to continuous infestations. This system, according to Kozár, was later quantified by Goanaca et al. (1974) by giving numerical values to each category. In his own writings, Kozár developed a somewhat complex scoring system (Table 1). Table 1 originally appeared in a work by Kozár and Victorin (1978) and was reproduced in Kozár's 1990 review. The United States Department of Agriculture, Animal and Plant Health Inspection Service (1992) has developed a fairly simple method of evaluating *U. euonymi* infestation. This system ranks scale infestations by physical appearance of the plant rather than actual numbers of scales (Table 2).

Table 1. Degree of infestation scoring system of population density of armored scale insects (From Kozár 1990).

Degree (score) of infestation	Specimens of scale insects on unit plant sample	Nymphs
	Trunk, branch, leaf ,fruit	All plant parts
0	0	0
1	0-5	1-10
2	5-20	11-50
3	21-100	51-100
4	100+	100+

System of scores: 0= no infestation, 1= a few individuals, 2 = small colonies, 3= large colonies, 4 = continuous infestation.

Table 2. Description of physical appearance and corresponding infestation level (from USDA, APHIS 1992).

High	<ol style="list-style-type: none"> 1. Yellowish or whitish spots on the upper surface of leaves. 2. Leaves and stems are heavily covered with a brown crust which are female scales. 4. Defoliated branches which are covered with scale in mid-summer.
Medium	<ol style="list-style-type: none"> 1. Leaves and stems covered with elongated white male scales. 2. No yellow or white spots on leaves. 3. No stem encrustation.
Low	<ol style="list-style-type: none"> 1. No white specks seen when approaching plant, but after spreading foliage apart there are one or two scattered white specks on the underside of leaves and on the stems. 2. No yellow or white spots on leaves. 3. No stem encrustation.
None	<p>No white specks on the plant after a few minutes of looking at undersides and lower parts of the stems.</p>

In addition to sampling to establish locations and densities of scale populations, sampling for natural enemies may also be desirable. Sampling for natural enemies is conducted to determine population composition and relative abundance of existing natural enemies both over time and space. Natural enemy sampling is purposefully biased. Only viable hosts from a specific age cohort are sampled to determine the composition and relative abundance of natural enemies. Random sampling is also desirable for collecting population data. However, sampling low population densities would require an enormous number of samples to be taken.

There are certain problem areas both in the field and in the laboratory which can make sampling and data collection of natural enemies difficult. Ants often interfere with host feeding and oviposition of parasites. Scale populations are often greater on branches where ants are active than on branches where they are not. Weather can adversely affect the activity of parasites. Heat buildup can severely depress parasite activity. Furthermore, dust is lethal to parasites (Beardsley & Gonzalez 1975). Presumably, female wasps are killed simply by abrasion of constant contact with the dust particles as they search for oviposition sites (Davidson & Miller 1990). Pesticides applied to the scale insects are lethal to parasites for an extended period, the lipophilic component of the test stores the pesticide so that it kills any emerging parasites. These factors do not necessarily affect all scales equally, therefore data gathered from sites affected by such factors would have great variability (Gulahamad & DeBach 1978).

Economic Injury Level and the Ornamental Landscape

A common definition of economic injury level (EIL) is the minimum number of insects that will cause economic damage or yield loss (Pedigo et al. 1986). Pedigo (1989) described the main ideas behind the concept of economic injury. There are distinctions among injury, damage, and economic damage. Injury is the effect of pest activities on host physiology. Damage is the measurable loss of host utility including loss of quality or aesthetic value. Economic damage is the amount of damage that will justify control measures.

The economic threshold or injury level is a complex value based on economics, injury potential and the potential growth rate of the pest population. Specifically, pest management decisions are based on the following factors: crop value, cost of management, degree of injury, and crop susceptibility to injury (Pedigo 1989).

As appealing as the concept of economic injury may seem, it does have some limitations. In crops that take years to mature, such as lumber and some field-grown nursery crops, economic injury levels are very difficult to determine. Market values fluctuate as do management costs. Furthermore, the parameters used in pest management decisions in agronomic crops do not easily lend themselves to the ornamental landscape (Pedigo 1989).

In place of the use of EIL, aesthetic injury levels (AIL) can be incorporated in the pest management decision-making process (Raupp et al. 1988). Coffelt and Schultz (1990) developed aesthetic injury levels for control of the orangestriped oakworm, *Anisota senatoria* (J. E. Smith) in Norfolk, VA. The injury level was developed by surveying the residents of Norfolk to determine how much defoliation (damage) they would accept. Acceptance levels were

combined with the value of the trees to determine action threshold. Similar methods have been used to develop AIL for other landscape pests. Virtually all insect populations are affected to some extent by natural enemies. For many species, natural enemies are a primary regulating force in population dynamics. The ability to manipulate natural enemies is significant to pest management. The object of biological control is either to introduce natural enemies or to manipulate existing ones, so as to cause the pest population to fluctuate (regulate) below the EIL or AIL.

Population regulation below injury levels can be achieved even if there is a large initial pest population. A large initial pest population provides an abundant food supply for the natural enemy. The predator or parasite population expands and consumes the pest species, thereby decreasing the food supply. As the food supply decreases, the natural enemy population drops and allows an increase in pest density, followed by an increase in the number of predators (Pedigo 1989).

Biological Control and *Chilocorus kuwanae* (Silvestri) (Coleoptera: Coccinellidae)

Taylor (1935) listed the following characteristics of the ideal biological control agent: (1) it is present during the entire growing season, (2) it has no serious natural enemies, (3) it is a voracious feeder at all mobile life stages and is not generally polyphagous, (4) it has a long adult life and (5) it has a high rate of dispersion, and, when its main food source is depleted, it will feed on other prey.

Chilocorus kuwanae meets many but not all the above criteria. Climate (especially high temperatures) has an adverse effect on the establishment of *C. kuwanae* populations. *Chilocorus kuwanae* will not establish on euonymus grown in

hot micro-climates where temperature are raised by reflection from bare soil or buildings (Hendrickson et al. 1991). In spite of these apparent drawbacks, *C. kuwanae* has proven to be a useful biological control agent and is becoming established in the United States.

Biology of *Chilocorus* spp.

In general, the diaspidid-feeding lady beetles have very similar life histories. Adult *Chilocorus* spp. mate approximately five days after emergence (Hodek 1967). Oviposition occurs shortly after mating. Greathead and Pope (1977) reported average pre-ovipositional periods ranging from 23 days in *Chilocorus nigritis* (Mulsant) to 50 days in *Chilocorus distigma* (Say). The eggs are laid singly or in small clusters, in cracks or crevices in the substrate or under scale covers. Females will lay eggs until shortly before death, providing there is a male present for repeated matings. Females may deposit hundreds of eggs; as many as 600 in some species. *Chilocorus nigritis* has been recorded laying an average of 2.2 eggs per day with a lifetime maximum of about 292 (Greathead & Pope 1977). Under laboratory conditions, the egg stage lasts about one week but varies under field conditions. The larval stage lasts from 1-3 weeks and consists of four or five instars. When five instars occur, instars 2-4 are shorter in duration (Drea & Gordon 1990).

The larvae of most species of *Chilocorus* will feed on all stages of prey. However, the immature larvae of some species are unable to feed on mature scales. The size of the larva relative to the size of the scale determines the stage of scale on which the larva will feed. The larvae of some species attack only the vulnerable egg and crawler stages, whereas the adults will also attack the mature scale. The larvae feed by one of two methods, either lifting the scale cover or chewing holes in it to expose the prey beneath. When food is scarce they will often cannibalize their own unhatched eggs. Mature larvae enter a 1-2 day pre-pupal state within the last larval cuticle. The cuticle then splits and the pupal stage appears. Many *Chilocorus* spp. congregate in small groups or clusters to pupate. These pupal aggregations may be more a haphazard result of larval feeding aggregations than of gregarious behavior (Samways 1984, Drea & Gordon 1990).

Predaceous coccinellids are parasitized and preyed upon by a host of vertebrate, invertebrate and disease organisms. The impact of these antagonists varies with the species of natural enemy, time of year, geographic region and the species of coccinellid involved (Drea & Gordon 1990). In the US, four species of Formicidae and one species of Eulophidae have been reported as serious pests of *C. kuwanae*. The species of ants are: *Camponotus ferrugineus* (F.), *C. nearcticus* (Emery), *Formica pallidefulva* (Latreille), *Lasius alienus* (Foerster) and *Taponoma sessile* (Say). The eulophid parasite is *Aprostocetus neglectus* (Domenichini). The effects and spread of the parasite can be prevented by releasing only adult *C. kuwanae* (Hodek 1967, Hendrickson et al. 1991).

The foraging behavior of *C. kuwanae* is typical of that of most predators. Before an encounter with prey, *C. kuwanae* moves along a relatively smooth course. With each successful prey encounter, the rate of travel and turning is increased. *Chilocorus kuwanae* maintains the increase in speed and turning during the search within the patch; that is, the area in which prey items are found (Luck 1984). The increase in turning is considered an adaptation of predators that feed on aggregated prey. This efficient search behavior apparently compensates for the lack of chemical or visual sensory apparatus used to detect prey at long distances (Podoler & Henen 1986). This lack of distance sensory apparatus may be a consideration in rearing *Chilocorus* spp. on host plants with morphological features that might offer protection to the scale.

Bull et al. (1993) concluded that in the laboratory, *C. kuwanae* will perform equally well on *Q. perniciosus* and *U. euonymi*. *Unaspis euonymi* has not been reared on an unnatural host. An alternative method to laboratory rearing would be the use of a field insectary that would provide *C. kuwanae* in sufficient quantities to meet the demands of distribution. The choice of an insectary host for *C. kuwanae* is based on several factors. In addition to availability of land for *Euonymus* spp. for rearing *U. euonymi* versus availability of and storage space for squash for rearing *Q. perniciosus*, there are other considerations. The same factors that adversely affect establishment of *C. kuwanae* in the landscape would have a similar effect in a field insectary. Parasitism by *A. neglectus* and antagonism by ants are potential problems. In addition to being inhibited by insect antagonists, *C. kuwanae* is also inhibited by high temperatures (Hendrickson et al. 1991). Parasites, other antagonists, and warm micro-

climates can be eliminated from laboratory cultures, but there are other factors to consider. The temperature and humidity of the culture must encompass the tolerances of the host substrate, the host insect, and the predator (Fisher & Finney 1973). When predators do not have enough food or space, cannibalism can occur, especially when overlapping life stages are present (Fisher & Finney 1973, Drea & Gordon 1990). Culture chambers must be adequately ventilated to prevent the growth of both plant and insect pathogens. The chamber may need to be illuminated to meet any light requirements of the host medium or insects (Fisher & Finney 1973).

Introduction of *Chilocorus kuwanae*

Any life stage of *Chilocorus* spp. can be introduced in the field. The introduction of eggs would avoid the problem of a forced dietary change and require less work in the insectary. Moreover, many more eggs can be released than adults. However, eggs appear to be highly susceptible to adverse conditions, including handling during the rearing process. Since eggs are securely cemented to the surface, movable artificial substrates such as cotton or frayed linen can be used to minimize handling damage. However, some species such as *Chilocorus infernalis* (F.) will not accept artificial substrates as oviposition sites (Hattingh & Samways 1991). Releases of eggs do not usually lead to successful establishment. Of 30 eggs observed by Hattingh and Samways (1991), only five reached fourth instar. None of the larvae pupated, and after 3 wk, no live individuals were found. Drea and Carlson (1987) were unable to recover any larvae from *C. kuwanae* eggs introduced from a laboratory culture. Release of third and fourth instar larvae has been

successful. Of the third and fourth instars of *C. infernalis* released, 21% pupated by the end of the second week, whereas the survival rate for second instar larvae was 9% (Hattingh & Samways 1991). Since egg and first instar larval mortality is high and adults tend to disperse, third and fourth instars may be best suited for introduction. However, in a comparison of three species, Hattingh and Samways (1991) found that adults were the most suitable in field introductions. A colony initiated with adult *C. nigrilis* survived through the summer and the coldest part of winter, albeit in small numbers. Conversely, Drea and Carlson (1987) contended that because adults tend to disperse, larvae are better suited for release. Among the diaspidid-feeding coccinellids, migration or dispersal does not occur in large numbers. When dispersal does occur, it is on an individual or small group basis resulting from a search for more favorable microhabitat (Drea & Gordon 1990). Greathead and Pope (1977) reported that *Chilocorus schioedtei* (Mulsant) prefers moist conditions and that *C. distigma* (Klug) prefers arid regions. *C. kuwanae* has not become established in Israel because of its intolerance of the high temperatures of the region (Drea & Gordon 1990). Even in temperate regions, high temperatures in the microhabitat can prohibit establishment of *C. kuwanae*. Hendrickson et al. (1991) reported releasing 60 *C. kuwanae* adults on 22 small specimens of *Euonymus fortunei* in Newark, Delaware; heat reflected by the bare soil surrounding the plants was believed to have repelled the predators.

The status of *U. euonymi* as a pest has clearly been established, as has its potential as a target of biological control by natural enemies. By understanding the biology and ecology of both *U. euonymi* and its natural enemies, *U. euonymi* populations should be able to be reduced to levels below that of

aesthetic injury. The objectives of this research were to facilitate increased production of natural enemies and to monitor the distribution of both native and imported natural enemies of *U. euonymi*.

Chapter 2

Rearing of *Unaspis euonymi* (Comstock)

Euonymus scale, *Unaspis euonymi*, utilizes evergreen *Euonymus* spp. as its primary host (Sadof & Raupp 1991). There are several factors that influence scale performance on a host plant. These include environmental factors, such as light, temperature and humidity (Beardsley & Gonzalez 1975), and host morphology and physiology (McClure 1979, Brewer & Oliver 1987, Sadof & Raupp 1991). Of the internal or physiological host factors, probably the most important is the amount of nitrogen in the phloem. Phloem feeders such as scales (Coccoidea) and aphids (Aphididae) obtain their dietary nitrogen from amides and amines found in plant sap in the phloem tissue. Therefore, any increase in nitrogen in the phloem would benefit scale performance and survival. There are several factors leading to increased soluble nitrogen in plant phloem. Increased soil fertility and environmental plant stress will result in temporary nitrogen increase (McClure 1990). Variegation in plant tissue is a cause of long term nitrogen increase. Variegation, or the unequal distribution of chlorophyll in plant tissues, causes the plant to produce less photosynthate per leaf. This reduction in photosynthetic output causes the plant to produce more leaves than a non-variegated plant per unit of photosynthetic output (Sadof & Raupp 1991). This added leaf production results in increased nitrogen movement in the phloem. The excess nitrogen is, in turn, intercepted by phloem feeders resulting in their improved performance and growth. Other host factors that affect mortality include bark color and texture (Beardsley &

Gonzalez 1975). Surface irregularities and leaf axils that offer protection and camouflage will elicit a thigmotactic response (Greathead 1990). In addition to surface irregularities, stem color also influences the wandering time of scale crawlers. Any factor that reduces crawler wandering time reduces crawler mortality (Beardsley & Gonzalez 1975, McClure 1979). The purpose of these experiments was to examine natural and unnatural hosts with differing physiology and morphology in order to gain further data regarding a suitable host for optimal *U. euonymi* production.

Materials and Methods

Unnatural Host Rearing

On 16 July 1992, one pad of prickly pear cactus, *Opuntia vulgaris* (Mill) and one fruit from of each of nine squash, *Cucurbita* spp., varieties were placed randomly in plastic trays. The squash fruit and cactus pads were grown at the Hampton Roads Agricultural Experiment Station (HRAES) in Virginia Beach Va. The following nine varieties were used: 'Early Acorn', 'Table Ace', 'Butternut', 'Little Dumpling', 'All Seasons', 'Chirimen', 'Mooregold', 'Buttercup' and 'Taki Midget'. These varieties were chosen because they do not decompose rapidly during storage.

Branches of *Euonymus japonicus* (Thunb.) were collected from an infested shrub at the author's home in Virginia Beach and examined for active crawlers. Upon confirmation of crawler presence, the scale-infested branches were placed on the new host material. The trays were then placed in a growth chamber at 27°

C and 70% r. h. After seven days, the branches were removed and the squash and cactus examined for the presence of settled first instars of *U. euonymi*.

Natural Host Rearing.

In September 1990, 24 plants of each of six *Euonymus* species and cultivars of different growth habits, foliage colors and variegation patterns were planted in a field at HRAES. The field was planted in a randomized complete block design with four replications and six plants per block. The following species were used: *Euonymus japonicus* (Thunb.), *E. japonicus* 'Microphyllus', *E. japonicus* 'Aureus', *E. japonicus* 'Albo-marginatus', *E. fortunei* (Turcz.), and *E. kiautschovicus* (Losen.) 'Manhattan' (Dirr 1983). Table 3 provides a description of the *Euonymus* species and cultivars used. Since *U. euonymi* has two generations per year in Virginia (Underhill 1943), this experiment was divided into two parts, each coinciding with one of the crawler emergence periods.

Part 1

On 10 May 1992, in conjunction with the first crawler emergence period, scale-infested twigs were removed from a hedge of euonymus located near HRAES. The twigs were cut to a length of 10 cm and the ends of the twigs were dipped in paraffin wax to retard desiccation. Two of the 10 cm twigs were then tied to current season's growth on each of the *Euonymus* species and cultivars in the field plots. On 17 May, the twigs were removed and the crawlers that had transferred from the infested twigs to the field host plants were counted. In November 1993, the plants in the field plots were rated according to scale population densities and assigned the following scores: 0 =

none, 1 = low infestation, 2 = medium infestation, 3 = heavy infestation, and 4 = continuous infestation.

Table 3. List and description of *Euonymus* species and cultivars.

Species	Description¹
<i>E. fortunei</i>	Procumbent or climbing shrub or ground cover. Leaves elliptic, variegated or solid green. Roots develop along lower surface of stem.
<i>E. japonicus</i>	Large shrub or small tree up to 7.6 m. Leaves obovate to oval, commonly used as an evergreen hedge.
<i>E. japonicus</i> 'Albo-marginatus'	Shrub or small tree; leaves smaller than above and are commonly misshapen with gray-green markings and a white a margin.
<i>E. japonicus</i> 'Aureus'	Upright in growth habit, leaves are large and golden yellow with a dark green margin. Variegation easily reverts to green.
<i>E. japonicus</i> 'Microphyllus'	A slow growing dwarf shrub up to 90 cm; small closely-spaced dark-green leaves. Frost sensitive.
<i>E. kiautschovicus</i> 'Manhattan'	Large shrub to 3 m, spreading habit with bright green oval to obovate leaves. Cold tolerant.

¹ Descriptions from Dirr (1983).

These ratings were based on the USDA, APHIS (1992) scoring system of *U. euonymi* infestations (Table 1). An analysis of variance (ANOVA) was performed to determine if there were any significant differences among species and cultivars and the degree of infestation ($\alpha = 0.05$). Tukey's honestly significant difference (HSD) test was performed to determine where the significant difference occurred (SAS Institute 1985).

Part 2

During June 1992, 12 container-grown plants of each of the six varieties (Table 3) were collected from commercial nurseries in Virginia for the second part of the experiment and placed in a randomized block design, in a wooded area that would provide protection from desiccation and heat stress. On 3 July, branches from heavily infested *E. japonicus* shrubs at HRAES were used to inoculate the plants during the second crawler emergence period. The branches were cut into 10 cm sections and the ends of the twigs were dipped in paraffin wax to minimize desiccation. The twig pieces were tied to current season's growth on the container plants and left in place for 7 d. Following crawler transfer, counts of settled crawlers and the surviving scale on each plant were taken every two weeks. Initially all settled first instars were counted. After scale tests began to develop, males and females were counted separately. Mortality was determined by counting the scale that failed to develop a covering (Stoetzel 1976). Analysis of variance (ANOVA) for a randomized complete block design (Lentner & Bishop 1986) was carried out for each cultivar to determine differences in percent change of scale densities.

On 1 July 92, stem cuttings were taken from each of the six *Euonymus* species and cultivars in preparation for the 1993 season (Table 3). Each

cutting was inserted in a 15 cm pot containing a mixture of peat moss and pine bark (1:2 by volume). The containerized cuttings were then placed in a high humidity propagator (Humidifan, Jaybird Manufacturing, Centre Hall, PA) to promote rooting. After the roots had developed, the plants were placed under shade-cloth that permitted 45% light penetration for one month to facilitate acclimation to ambient conditions. Following acclimation, the containerized *Euonymus* spp. were transferred to a growing area where they received daily overhead irrigation. On 20 September, the *Euonymus* species and cultivars were moved to an unheated greenhouse to protect them from winter injury. On 5 May 1993, the plants were removed from the greenhouse and returned to the shade to protect plants from heavy rain and desiccation.

Beginning 1 May 1993, female *U. euonymi* infesting the plants at HRAES and the author's home in Virginia Beach were examined daily to determine the onset of crawler emergence. On 11 May 1993, branches of previous year's growth from a heavily infested *E. japonicus* located at the author's home were used to inoculate the plants. After removing the branches from the infested plant, the branches were cut into 10 cm sections. Two freshly cut 10 cm twig sections were tied to 72 of the year-old container-grown *Euonymus* plants and replaced at 2d intervals for 6 days (12 plants of each of six species and cultivars [Table 3] in a randomized complete block design with 12 blocks). During the inoculation process, the plants were placed in darkness at 27° C and 70% r.h. On 18 May 1993, following crawler transfer, the newly infested plants were placed under shade and settled first instars were counted. Subsequent counts of surviving scale were made 1 June and 15 June. On 17 May 1993, a second group of *Euonymus* species and cultivars was inoculated

using the above methods, using five blocks instead of 12. The surviving scales on these plants were counted 24 May, 7 June and 21 June. The density of scale insects (scales / cm of stem length) was recorded at each counting date. A two-way analysis of variance (ANOVA) for a randomized complete block design (Lentner & Bishop 1986) was carried out using block and cultivar as class variables. Where statistically significant differences were found at $\alpha = 0.05$, Tukey's HSD mean comparison procedure was performed to determine where the differences occurred.

Results and Discussion

Unnatural Host Rearing

Although other scale insects have been successfully reared on the host material used, there were no living *U. euonymi* found on any of the *Cucurbita* spp. or the *Opuntia vulgaris*. There have been no published reports of success in rearing *U. euonymi* on an unnatural host. Therefore, if *U. euonymi* is to be reared in an insectary, it will most likely be necessary to rear it on its natural hosts. In addition to *Euonymus* spp., *U. euonymi* also utilizes other plant species as hosts. These other hosts include *Buxus*, *Camellia*, *Ilex*, *Lonicera* and *Pachysandra* spp. (Johnson & Lyon 1988).

Natural Host Rearing

Of the 144 field-grown *Euonymus* species and cultivars, initial observable crawler transfer was successful on only a few plants. Therefore, a scale survival study was not conducted. However, analysis of the infestation ratings after the completion of four generations (2 years) of *U. euonymi* did show

statistically significant differences in *U. euonymi* infestation levels among the six different *Euonymus* species and cultivars ($F = 11.19$, $df = 121$, $P < 0.001$). *E. japonicus* 'Albo-marginatus' and *E. japonicus* 'Microphyllus' had the two highest means; the former being significantly greater than the remaining four species and cultivars, and the latter significantly greater than the three of the remaining four species and cultivars (Table 4).

Table 4. Mean scale density rating for 1992 - 1993 infestation trials.

<i>Euonymus</i> species and cultivars	Mean Score ¹	Sample Size
<i>E. japonicus</i> 'Albo-marginatus'	1.82 a	11
<i>E. japonicus</i> 'Microphyllus'	1.66 ab	24
<i>E. japonicus</i> 'Aureus'	0.87 bc	23
<i>E. fortunei</i>	0.47 c	19
<i>E. kiautschovicus</i> 'Manhattan'	0.09 c	23
<i>E. japonicus</i>	0.04 c	22

¹ Mean scores followed by the same letter are not significantly different; ($P > 0.05$; Tukey's test [SAS Institute 1985])
Scoring system- 0 = none, 1 = light, 2 = medium, 3 = high.

During the second crawler emergence period of 1992, there was successful scale transfer with 58 of the 72 container-grown *Euonymus* species and cultivars, albeit very few scales actually settled on each. The average number transferred was 31. The range for all plants was 0-140. There was a large amount of variation within each of the cultivar groups as well. Analysis of variance (ANOVA) showed no statistical significance among species and cultivars. This second generation inoculation, although it did not provide results on species and cultivar differences, proved to be a valuable tool in the development of infestation and counting techniques as well as revealing sources of variability which could be eliminated or greatly reduced.

Analysis of scale survival data from the container grown *Euonymus* species and cultivars showed a parallel trend to the survival data from the field-grown *Euonymus* spp. and cultivars. Initially, *E. japonicus* 'Albo-marginatus' and *E. kiautschovicus* 'Manhattan' showed highest settled crawler densities. However, by the final count, *E. kiautschovicus* 'Manhattan' had dropped in rank and *E. japonicus* 'Albo-marginatus' and *E. japonicus* 'Microphyllus' had the two highest average scale densities (Tables 5-7). Although *E. kiautschovicus* 'Manhattan' had the second highest initial scale density, the scale on *E. kiautschovicus* sustained 80% mortality, the highest level of mortality between the initial and final counts of all six *Euonymus* species and cultivars. *E. japonicus* 'Aureus' showed the lowest initial settled crawler density of the six species and cultivars.

However, the scale on the *E. japonicus* 'Aureus' showed the least amount of mortality (7%) (Fig. 1).

In addition to scale survival, the 1993 container *Euonymus* spp. and cultivar experiment was intended to measure overall scale performance on six different species and cultivars of *Euonymus*. Unfortunately, most of the test plants died of root disease at the end of the first scale generation. Therefore, no population increase data could be collected.

Table 5. Mean scale density on each of the six *Euonymus* species and cultivars, initial count (18 May).

Cultivar	Mean density (scale / cm)	N	Standard error
<i>E. japonicus</i> 'Albo-marginatus'	5.90 a	11	0.31
<i>E. kiautschovicus</i> 'Manhattan'	5.65 a	12	0.38
<i>E. japonicus</i> 'Microphyllus'	4.34 ab	10	0.31
<i>E. fortunei</i>	2.78 ab	12	0.27
<i>E. japonicus</i>	2.71 ab	11	0.20
<i>E. japonicus</i> 'Aureus'	1.40 b	9	0.13

Mean densities followed by the same letter are not significantly different; ($P > 0.05$, Tukey's test [SAS Institute 1985]).

Table 6. Mean scale density on each of the six *Euonymus* species and cultivars, second count (1 June).

Cultivar	Mean density (scale/cm)	N	Standard error
<i>E. japonicus</i> 'Albo-marginatus'	4.90 a	11	0.32
<i>E. kiautschovicus</i> 'Manhattan'	4.80 a	12	0.32
<i>E. japonicus</i> 'Microphyllus'	3.30 ab	10	0.26
<i>E. fortunei</i>	2.74 ab	12	0.19
<i>E. japonicus</i>	2.16 ab	11	0.13
<i>E. japonicus</i> 'Aureus'	1.51 b	9	0.14

Mean densities followed by the same letter are not significantly different ($P > 0.05$, Tukey's test [SAS Institute 1985]).

Table 7. Mean scale density on each of the six *Euonymus* species and cultivars, final count (15 June).

Cultivar	Mean density (scale/cm)	N	Standard error
<i>E. japonicus</i> 'Albo-marginatus'	3.32 a	11	0.20
<i>E. japonicus</i> 'Microphyllus'	1.78 ab	10	0.15
<i>E. kiautschovicus</i> 'Manhattan'	1.66 ab	12	0.13
<i>E. japonicus</i>	1.16 ab	11	0.09
<i>E. japonicus</i> 'Aureus'	1.10 b	9	0.11
<i>E. fortunei</i>	0.92	12	0.12

Mean densities followed by the same letter are not significantly different ($P > 0.05$, Tukey's test [SAS Institute 1985]).

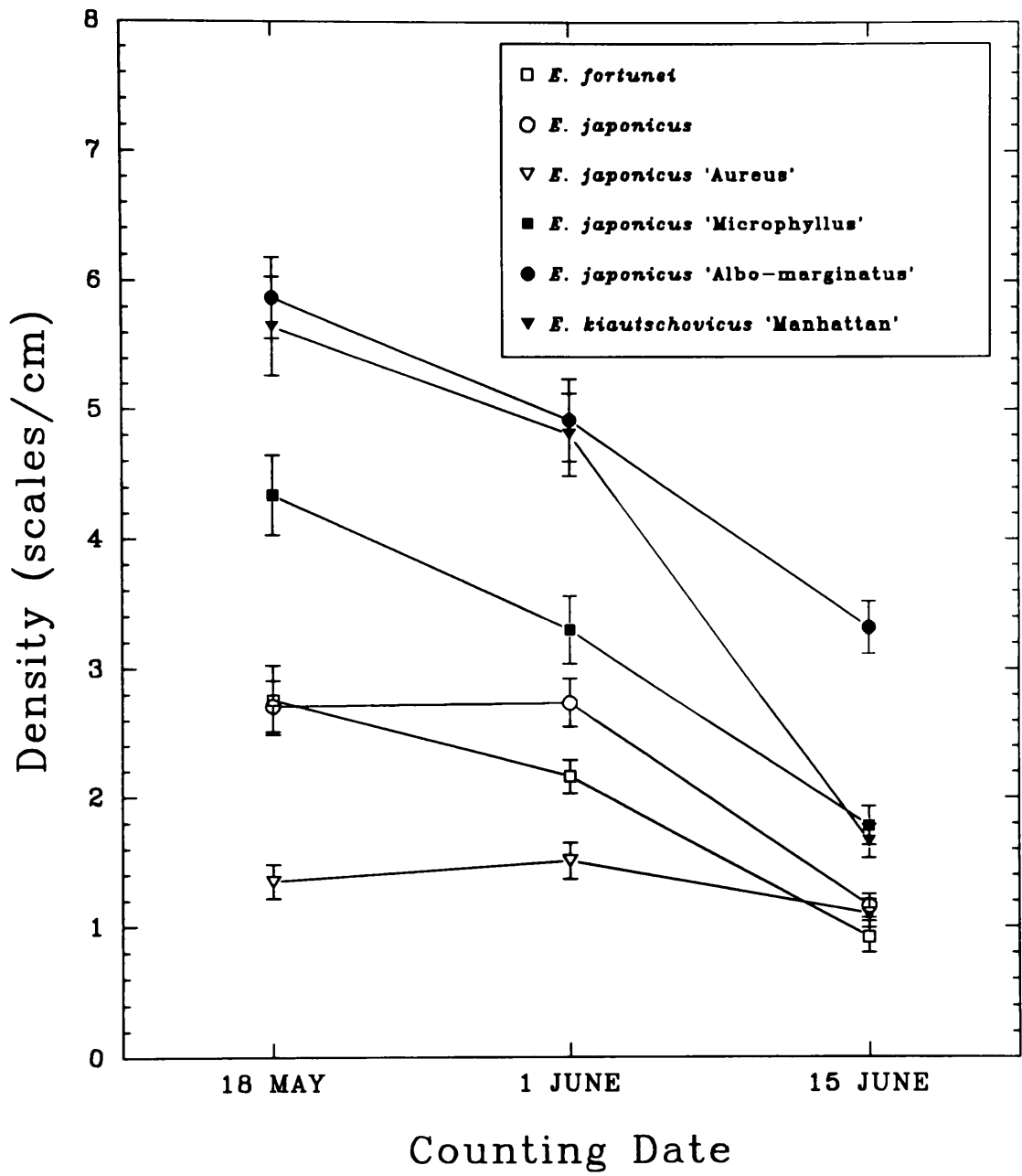


Fig. 1. Scale densities on the six species and cultivars of *Euonymus*, 1993.

Chapter 3

Survey of Establishment of the Introduced *Chilocorus kuwanae* (Silvestri) (Coleoptera: Coccinellidae) and Primary Existing Natural Enemies of *Unaspis euonymi* (Comstock) in Virginia

Unaspis euonymi (Comstock) is a serious pest of evergreen *Euonymus*. Even though a number of natural enemies have been found that utilize *U. euonymi*, none have proven to be useful biological agents.

In 1983-84, The United States Department of Agriculture, Agricultural Research Service (USDA, ARS) imported and established two predators of *Euonymus* scale (Drea & Carlson 1987). The USDA Animal and Plant Health Inspection Service (APHIS), in cooperation with the Virginia Department of Agriculture and Consumer Services (VDACS), released *Chilocorus kuwanae* (Silvestri) in Virginia Beach, VA, in 1987. A *C. kuwanae* culture was also initiated in 1987 at the Hampton Roads Agricultural Experiment Station (HRAES), in Virginia Beach. In addition to the Virginia Beach release, *C. kuwanae* was released in the following Virginia counties in 1992: Henrico, Clarke, Roanoke, Fairfax and Spotsylvania. In 1993, *C. kuwanae* was released in Loudoun, Campbell, Pittsylvania and Prince George counties.

Natural enemies, other than Coccinellidae, have been reported on *U. euonymi*. Gill et al. (1982) reported the mite *Hemisarcoptes malus* (Shimer) (Acari: Hemiscaroptidae) and the aphelinid parasite *Aspidiotiphagus citrinus* (Crawford) (Hymenoptera: Aphelinidae) in Maryland. In addition to

Aspidiotiphagus sp., Kosztarab (1963) reported *Prospaltella* sp. (Hymenoptera: Aphelinidae) and the predaceous mite *Thyreophagus entomophagus* (Laboulbene) (Acari: Acaridae) and other unidentified mites in the families Phytoseiidae and Tarsonemidae. Kosztarab and Kozár (1988) listed the following nine parasite species: *Aenasioidea hispanica* (Girault) (Hymenoptera: Encyrtidae), *Alaptus excisus* (Westwood) (Hymenoptera: Mymaridae), *Aphytis proclia* (Walker) (Hymenoptera: Aphelinidae), *Aspidiotiphagus citrinus* (Crawford) (Hymenoptera: Aphelinidae), *Coccidencyrtus poutiersi* (Ashmead) (Hymenoptera: Encyrtidae), *Coccophagoides similis* (Masi) (Hymenoptera: Aphelinidae), *Dicopus citri* (Girault) (Hymenoptera: Mymaridae), *Prospaltella gigas* (Howard) (Hymenoptera: Aphelinidae) and *Thysanus merceti* (Malenotti) (Hymenoptera: Signiphoridae). *Encarsia* spp. have also been found in *Unaspis* infestations (Bryan, personal communication). A few members of the Cecidomyiidae, particularly the genus *Lestodiplosi*, also feed on scale (Harris 1990). Additionally, Neuroptera in the families Raphidiidae, Coniopterygidae and Chrysopidae prey on scale insects (Drea 1990). Despite the occasional abundance of these parasites and predators, they have been ineffective as biological control agents.

MATERIALS AND METHODS

Distribution Survey, 1992

The 1992 distribution study was confined to twenty-five sampling sites within a 10 mile radius of HRAES (Table 8). Only *Euonymus* spp. with

medium to high scale populations were sampled. The original sites were located using 1991 survey data from the *Euonymus* Scale Biological Control Project, of USDA, APHIS. During the 1992 sampling process, some of the sites were sprayed by the landscape managers, destroying the local scale population. Alternate sites were selected by searching for *Euonymus* spp. infested with healthy scale near the sprayed sites. The viability of the scale at the replacement sites was confirmed by dissecting 150 scales from each location to ensure a majority of living scale insects.

Sampling at each of the locations was accomplished by two methods. Sampling for parasites and slow moving predators was achieved by the removal of a 10 cm twig from each compass quadrant of the scale infested region of the *Euonymus* spp. The twigs were of current season's growth. Leaves were removed and the twig was placed in a culture tube with a cotton stopper. Larger, faster moving predators were sampled by using a beating sheet. The predators were removed from the plant by either beating or shaking the branches over the collection sheet. Following collection, twig samples were returned to the laboratory and the scales on each twig were counted. Predators, such as *Chilocorus* larvae and other large insects, were removed. Culture tubes were then placed in darkness for 4 weeks until the parasites had emerged. After 4 weeks the dead parasites were removed from the culture tubes and representative samples were sent to the USDA, APHIS laboratory in Niles, Michigan for identification. Sites where *C. kuwanae* were recovered were recorded.

Table 8. List of 1992 Virginia Beach Sampling sites

Site Number	Address	Positive for <i>C. kuwanae</i>	Positive for <i>Aspidiotiphagus</i> spp. or <i>Encarsia</i> spp.
1.	5632 Zinia Court	YES	YES
2.	5628 Zinia Court	NO	NO
3.	5540 Indian River Road	NO	YES
4.	1236 General Avenue	YES	YES
5-7.	5656 General Avenue	YES(all)	YES(all)
8-10.	5549 Indian River Road	NO	NO
11.	6680 Indian River Road	NO	NO
12-14.	1749 LaCrosse Drive	NO	YES(14)
15.	1596 Appleton St.	YES	NO
16-17.	1581 Princes Anne Road	NO	YES
18.	667 Aragona Blvd.	YES	NO
19.	756 Aragona Blvd.	YES	NO
20-21.	800 Aragona Blvd.	YES	NO
22-23.	5561 Bayside Avenue	YES	YES
24-25.	1506 Air Rail Avenue	YES (24)	NO

Distribution Survey 1993

Following the 1992 study, the natural enemy survey was expanded into a state-wide distribution study. With the aid of many extension agents and home owners, 41 sites in 12 counties in Virginia were chosen as survey sites (Table 9). Since *Euonymus* spp. are native to Asia and do not occur naturally in Virginia, sites were chosen according to distribution of population centers where *Euonymus* plantings would likely be found. In addition to cities sampled, Table 9 also lists the biotic or life zones found in Virginia (Hoffman 1969). This information has been included so that any natural enemies that might be found could be associated with their corresponding biotic region. Collection methods for the 1993 survey were the same as those used in the 1992 survey. The Danville and Emporia sites were dropped from the survey route after 12 July because of the lack of parasites at the Danville sites and the distance to those sites.

RESULTS AND DISCUSSION

1992 Distribution Survey:

There were both *Aspidiotiphagus* spp. and *Encarsia* spp. (Hymenoptera: Aphelinidae) in each of the samples sent to the USDA, APHIS laboratory in Niles. Additionally, some of the samples contained parasites from more than one location. Therefore the specific location of each genus in Virginia Beach could not be established. Of the 25 Virginia Beach sites sampled, *Aspidiotiphagus* or *Encarsia* spp. were collected from 11 sites and *C. kuwanae*

was collected at nine sites (Table 8). Figure 3 shows the locations for natural enemies in Virginia Beach.

1993 Virginia Survey

As in the 1992 survey, *Aspidiotiphagus* spp. and *Encarsia* spp. were the primary indigenous natural enemies found. These two genera, as with most endoparasites, do not parasitize *U. euonymi* at a rate sufficient to keep the scale population below plant injury levels. Of the 12 cities sampled, *C. kuwanae* was recovered from eight. Aphelinid parasites were found in all 12 cities. Aphelinid parasites were found at 18 of the 41 locations (44%) on 25 May, whereas none was found during the 12 July sampling period (Tables 10, 13, Figure 4). Variation in the results from each sampling period is most likely due to changes in the relative abundance of third instar *U. euonymi* from which the parasites would be emerging.

Chilocorus kuwanae was recovered from 11 of 36 sites (31%) on 10 August and from six of 36 sites (16%) on 27 July. These percentages would have 31% and 19%, respectively if Danville and Emporia had been sampled and no changes in insect populations at the sites had occurred (Tables 10,13, Figure 4). Two of the five sites in Fredericksburg were positive for *C. kuwanae*. These were not the same two locations for each date (Tables 11,12). Variation in the numbers of *C. kuwanae* collected might be explained by the above average temperatures during July, 1993. The beetles may have moved to cooler micro-climates during the July sampling trips.

The establishment of *C. kuwanae* in Virginia is not surprising since it has become established in both Maryland and North Carolina (Drea & Carlson

1987, Nalepa et al. 1993). Even though *Euonymus* spp. and *U. euonymi* are not distributed uniformly throughout the landscape, *C. kuwanae* has little difficulty finding new food sources. In Charlottesville, *C. kuwanae* larvae were found in mid-August on a euonymus hedge where they had not been found previously. The nearest known location of *C. kuwanae* was 3 miles away. Similar sudden appearances of *C. kuwanae* occurred in Virginia Beach during 1992.

Table 9. Distribution of 1993 sampling sites by city, county and life zone

City	Number of sites	County	Life zone
Mechanicsville	1	Hanover	Carolinian
Richmond	4	Henrico	Carolinian
Fredericksburg	5	Stafford	Carolinian
Leesburg	5	Loudoun	Alleghanian
State Arboretum	3	Clarke	Alleghanian
Charlottesville	4	Albemarle	Alleghanian
Roanoke	3	Roanoke	Alleghanian
Lynchburg	4	Campbell	Carolinian
Danville	3	Pittsylvania	Carolinian
Emporia	2	Greensville	Austroriparian
City of Chesapeake	1	Incorporated	Austroriparian
City of Virginia Beach	4	Incorporated	Austroriparian

Life zones as described by Hoffman (1969).

Table 10. Survey for *Chilocorus kuwanae* and aphelinid parasites in Virginia, 25 May 1993.

City	Number of sites	County	Sites positive for <i>C. kuwanae</i> / sites inspected	Sites positive for <i>Aspidiotiphagus</i> or <i>Encarsia</i> / sites inspected
Mechanicsville	1	Hanover	1/1	0/1
Richmond	4	Henrico	1/4	1/4(A) ¹
Fredericksburg	5	Stafford	2/5	4/5(A)
Leesburg	6	Loudoun	0/6	1/6(A)
Blandy Farm	3	Clarke	1/3	2/3(A)
Charlottesville	4	Albemarle	2/4	3/5(A)
Roanoke	3	Roanoke	0/3	3/3(A)
Lynchburg	4	Campbell	0/4	1/4(A)
Danville	3	Pittsylvania	0/3	0/3
Emporia	2	Greensville	2/2	1/2(E)
City of Chesapeake	1	Incorporated	0/1	0/1
City of Virginia Beach	4	Incorporated	3/4	2/4(A,E)
TOTALS	41		12/41 (30%)	18/41 (44%)

¹ A = *Aspidiotiphagus* sp., E = *Encarsia* sp.

Table 11. Survey for *Chilocorus kuwanae* and aphelinid parasites in Virginia, 14 June 1993.

City	Number of sites	County	Sites positive for <i>C. kuwanae</i> / sites inspected	Sites positive for <i>Aspidiotiphagus</i> or <i>Encarsia</i> / sites inspected
Mechanicsville	1	Hanover	1/1	0/1
Richmond	4	Henrico	2/4	2/4(E,A ¹)
Fredericksburg	5	Stafford	2/5	5/5(A,C)
Leesburg	6	Loudoun	0/6	2/6(E,U)
Blandy Farm	3	Clarke	0/3	0/3
Charlottesville	4	Albemarle	2/4	1/4(A)
Roanoke	3	Roanoke	0/3	2/3(A,U)
Lynchburg	4	Campbell	0/4	1/4(A0)
Danville	3	Pittsylvania	0/3	0/3
Emporia	2	Greensville	2/2	0/2
City of Chesapeake	1	Incorporated	0/1	0/1
City of Virginia Beach	4	Incorporated	3/4	0/4
TOTALS	41		9/41 (22%)	12/41 (29%)

¹ A = *Aspidiotiphagus* sp., E = *Encarsia* sp., U = Unknown C = *Cybocephalus* sp.

Table 12. Survey for *Chilocorus kuwanae* and aphelinid parasites in Virginia, 1 July 1993.

City	Number of sites	County	Sites positive for <i>C. kuwanae</i> / sites inspected	Sites positive for <i>Aspidiotiphagus</i> or <i>Encarsia</i> / sites inspected
Mechanicsville	1	Hanover	1/1	0/1
Richmond	4	Henrico	2/4	0/4
Fredericksburg	5	Stafford	2/5	4/5(A) ¹
Leesburg	6	Loudoun	0/6	3/6((E)
Blandy Farm	3	Clarke	0/3	1/3(E)
Charlottesville	4	Albemarle	2/4	2/4(A)
Roanoke	3	Roanoke	0/3	2/3(A)
Lynchburg	4	Campbell	0/4	1/4(A)
Danville	3	Pittsylvania	0/3	0/3
Emporia	2	Greensville	2/2	0/2
City of Chesapeake	1	Incorporated	0/1	1/1(E)
City of Virginia Beach	4	Incorporated	3/4	1/4(E)
TOTALS	41		8/41 (20%)	16/41 (39%)

¹ A = *Aspidiotiphagus* sp., E = *Encarsia* sp.

Table 13. Survey for *Chilocorus kuwanae* and aphelinid parasites in Virginia, 12 July 1993.

City	Number of sites	County	Sites positive for	
			<i>C. kuwanae</i> / sites inspected	<i>Aspidiotiphagus</i> or <i>Encarsia</i> / sites inspected
Mechanicsville	1	Hanover	1/1	0/1
Richmond	4	Henrico	1/4	0/4
Fredericksburg	5	Stafford	1/5	0/5
Leesburg	6	Loudoun	0/6	0/6
Blandy Farm	3	Clarke	0/3	0/3
Charlottesville	4	Albemarle	2/4	0/4
Roanoke	3	Roanoke	0/3	0/3
Lynchburg	4	Campbell	0/4	0/4
Danville	3	Pittsylvania	0/3	0/3
Emporia	2	Greensville	2/2	0/2
City of Chesapeake	1	Incorporated	1/1	0/1
City of Virginia Beach	4	Incorporated	2/4	0/4
TOTALS	41		10/41 (24%)	0/41 (0%)

Table 14. Survey for *Chilocorus kuwanae* and aphelinid parasites in Virginia, 27 July 1993.

City	Number of sites	County	Sites positive for <i>C. kuwanae</i> / sites inspected	Sites positive for <i>Aspidiotiphagus</i> or <i>Encarsia</i> / sites inspected
Mechanicsville	1	Hanover	0/1	1/1(A) ¹
Richmond	4	Henrico	1/4	1/4(E)
Fredericksburg	5	Stafford	0/5	2/5(A)
Leesburg	6	Loudoun	0/6	5/6(A,E)
Blandy Farm	3	Clarke	1/3	3/3(A,E)
Charlottesville	4	Albemarle	1/4	2/4(A)
Roanoke	3	Roanoke	0/3	1/3(A)
Lynchburg	4	Campbell	0/4	2/4(A)
City of Chesapeake	1	Incorporated	0/1	0/1
City of Virginia Beach	4	Incorporated	3/4	1/4(A)
TOTALS	36		6/36 (16%)	17/36 (22%)

¹ A = *Aspidiotiphagus* sp., E = *Encarsia* sp.

Table 15. Survey for *Chilocorus kuwanae* and aphelinid parasites in Virginia, 10 August 1993.

City	Number of sites	County	Sites positive for <i>C. kuwanae</i> / sites inspected	Sites positive for <i>Aspidiotiphagus</i> or <i>Encarsia</i> / sites inspected
Mechanicsville	1	Hanover	0/1	0/1
Richmond	4	Henrico	1/4	0/4
Fredericksburg	5	Stafford	1/5	0/5
Leesburg	6	Loudoun	0/6	3/6(A,E ¹)
Blandy Farm	3	Clarke	1/3	2/3(A)
Charlottesville	4	Albemarle	3/4	2/4(A)
Roanoke	3	Roanoke	0/3	0/3
Lynchburg	4	Campbell	0/4	2/4(A)
City of Chesapeake	1	Incorporated	1/1	1/1(A)
City of Virginia Beach	4	Incorporated	4/4	0/4
TOTALS	36		11/36 (36%)	10/36 (27%)

¹ A = *Aspidiotiphagus* sp., E = *Encarsia* sp.

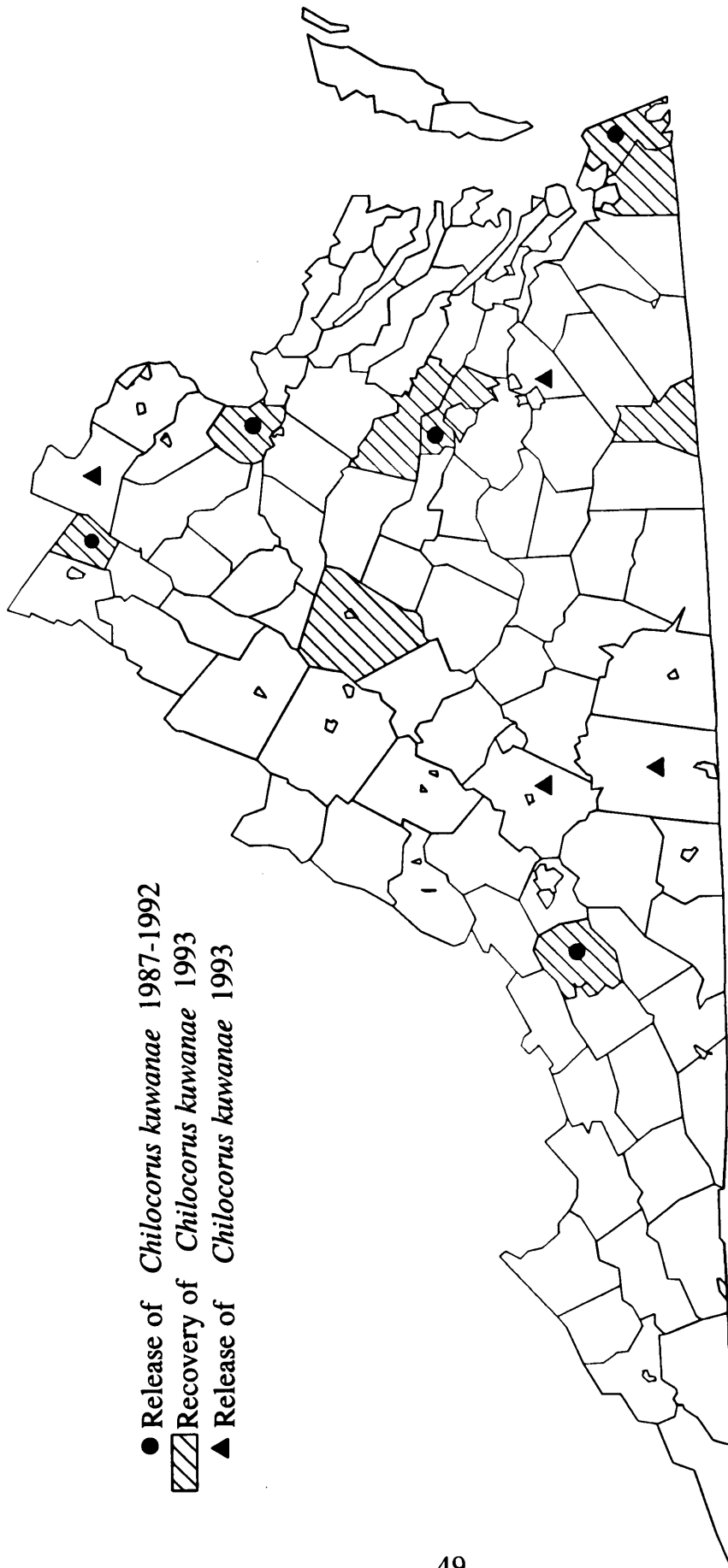


Fig. 2. Spatial distribution of releases and recoveries of *Chilocorus kuwanae* in Virginia 1987-1993.

Cybocephalus sp. (Coleoptera: Nitidulidae) was introduced from Asia, by USDA, at three sites in the greater Washington D.C. metropolitan area in 1984 (Drea & Carlson 1988). A single specimen of *Cybocephalus* sp. was recovered from a site in Fredericksburg during the 1993 survey. Even though only one specimen of *Cybocephalus* sp. was recovered during the sampling period, its presence may be significant. The number of *Cybocephalus* sp. at the Fredericksburg site, as well as their distribution, may be greater and more widely spread than the recovery of one specimen would indicate. *Cybocephalus* sp. are very small and easily overlooked in visual searches. Most *Cybocephalus* sp. have been recovered accidentally while sampling for parasites (Bryan, personal communication). Their size as well as their life cycle makes them difficult to locate. Female *Cybocephalus* sp. oviposit in hidden areas such as in cracks and crevices or under scale coverings. The larvae then develop under the scale covering. The scale cover not only provides concealment, but also offers protection from pesticides (Blumberg & Swirski 1982). Therefore, there may be more *Cybocephalus* sp. at the Fredericksburg site than the evidence indicates.

Conclusion

Most ornamental landscape environments are subject to frequent human contact. Moreover, as the public becomes increasingly sensitive or hostile to the use of pesticides in the landscape, other avenues of pest control will need to be explored. The use of integrated pest management is the best method of reducing pesticide usage. Armored scale insects are well suited for integrated control methods. Their populations can be suppressed by number a of methods, including biological control.

The goal of this research was to continue the collection of information useful in the suppression of *U. euonymi* populations to below aesthetic injury levels. The use of *C. kuwanae* as a biological control agent in combination with native parasites would enable *Euonymus* spp. to remain in the ornamental landscape and in nursery production without the excessive use of chemical pest control methods. This goal was reached by three methods. The first was the determination of a suitable plant host for *U. euonymi* for the field insectary rearing of *C. kuwanae*, in sufficient quantities that would meet the needs of a distribution program. Of those species and cultivars of *Euonymus* evaluated, *E. japonicus* 'Albo-marginatus' proved to be the best host as demonstrated in both scale crawler survival and in population increase over an extended period. The second was determination of the widespread distribution of the native parasitic species of *Aspidiotiphagus* and *Encarsia* and the relative abundance of adults over time. Populations of parasites were not of sufficient density to reduce *U. euonymi* populations to below injury levels. The third area was the confirmation of the establishment and distribution of *C. kuwanae* in Virginia. This confirmation was accomplished through a survey that showed *C. kuwanae* to be present in all life zones in Virginia where euonymus is grown. Given these data and that by other authors, it is safe to assume that as *C. kuwanae* becomes established, *U. euonymi* will become less of a problem in the ornamental landscape.

Appendix 1.

1993 Virginia survey site list

No.	County / City	Location or Address
1.	Hanover / Mechanicsville	Prairie Schooner restaurant 600 Mechanicsville Pike (<i>E. japonicus</i> 'Albo-Marginatus')
2.	Richmond	1501 Honey Tree Apts. (<i>E. japonicus</i>)
3.		2303 Carrollwood Court (<i>E. japonicus</i>)
4.		2300 Carrollwood Court (<i>E. japonicus</i>)
5.	Stafford / Fredericksburg	Dottie's Den 3701 Lafayette Blvd., left side (<i>E.</i> <i>japonicus</i>)
6.		Dottie's Den 3701 Lafayette Blvd., right side (<i>E.</i> <i>japonicus</i>)
7.		Mary Washington physical plant entrance, right side (<i>E. japonicus</i>)
8.		Mary Washington physical plant entrance, left side (<i>E. japonicus</i>)
9.		Spotsylvania Shopping Center, Taco Bell (<i>E.</i> <i>japonicus</i>)
10.	Loudoun / Leesburg	Shopping center in 200 block of Rt. 7 (Market St.), near Days Inn next to Errol's Video, left parking lot island (<i>E. japonicus</i>)
11.		Shopping center in 200 block of Rt. 7 (Market St.), near Days Inn next to Errol's Video, right parking lot island (<i>E. japonicus</i>)
12.		Townhouse Shopping Center at the corner of Market and South St., front entrance (<i>E. japonicus</i>)
13.		Townhouse Shopping Center at the corner of Market and South St., side of convenience store (end closest to Market St.) (<i>E. japonicus</i>)
14.		Woodburn estate off Lewis Lane, near house by driveway (<i>E. japonicus</i>)
15.	Woodburn estate off Lewis Lane, near bend in driveway (<i>E. japonicus</i>)	

16. Clarke / Va. State arboretum (Blandy Farm) Wahoo euonymus near end of gravel road near fence (*E. atropurpureus*)
17. Wintercreeper euonymus on first tree on right side of gravel road (*E. fortunei*)
18. Wintercreeper euonymus on second tree on right side of gravel road (*E. fortunei*)
19. Albemarle / Charlottesville Albemarle Square Shopping Center, Virginia Federal Bank (*E. japonicus*)
20. Hardees at Rio road and Rt. 29 (*E. japonicus* 'Aureus')
21. Fountain Court Apts. off of Rio Road, large hedge at end of complex near pool (*E. japonicus*)
22. Fountain Court Apts. on hill behind complex (*E. fortunei*)
23. 804 Davis St. right side of house (*E. japonicus* 'Aureus')
24. Roanoke / Roanoke Motel near intersection of Rt. 11 and Plantation Rd., sign planting. (*E. japonicus*)
25. Pepsi plant on Rt. 11, hedge by wall (*E. japonicus*)
26. Hollins College, hedge by French House (*E. japonicus*)
27. Campbell / Lynchburg 101 Deerfield, left of front door (*E. japonicus* 'Aureus')
28. Lynchburg city hall, north entrance (*E. fortunei*)
29. 2116 Edinburgh Ave., rear of house (*E. japonicus* 'Albo-marginatus')
30. Red Lobster at River Ridge Mall, rear of parking lot (*E. japonicus*)
31. Pittsylvania / Danville 307 Holbrook St., by fence near Roberts St. (*E. japonicus* 'Albo-marginatus')
32. 307 Holbrook St., front yard middle shrub. (*E. japonicus* 'Albo-marginatus')
33. 307 Holbrook St., large shrub by driveway (*E. japonicus* 'Albo-marginatus')

34. Greenville / Emporia McDonalds, front shrub bed (*E. japonicus* 'Albo-marginatus')
35. McDonalds, back of parking lot (*E. japonicus* 'Albo-marginatus')
36. Chesapeake 1448 Eva Dr., rear of house (*E. japonicus* 'Albo-marginatus')
37. Virginia Beach 5632 Zinia Court near garage (*E. japonicus* 'Albo-marginatus')
38. 1444 Diamond Springs Rd., euonymus field, rear of property (*E. japonicus* 'Albo-marginatus')
39. UPS warehouse sign at entrance on Air Rail Ave. (*E. japonicus* 'Albo-marginatus')
40. Norfolk International Airport, overflow parking, Miller's Store Rd. (*E. japonicus*)

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VITA

David Kirk Jefferson was born in June of 1959, in Chicago, Illinois. He grew up mostly in Athens, Georgia, and Lancaster, Pennsylvania. He graduated in 1977 from Connastoga Valley High School in Lancaster, Pennsylvania. After high school, he worked in the greenhouse tropical foliage production industry before starting college at Millersville State college in Millersville, Pennsylvania. He graduated, in 1985, from the University of Georgia with a Bachelor of Science in Ornamental Horticulture. While at the University of Georgia, he spent a summer as an intern working in plant interiorscapes. He has worked in various capacities in greenhouse and landscape management including, most recently, a position as gardener with the Colonial Williamsburg Foundation. He began work on a Master of Science in Entomology in 1991 under the direction of Dr. Peter B. Schultz. His work involved the biological control of *Euonymus* scale. He is currently employed as a pest management technician at Regent University in Virginia Beach, VA, and is an adjunct instructor of pest management at Tidewater Community College.

A handwritten signature in black ink, appearing to read "David Kirk Jefferson". The signature is written in a cursive, flowing style with a long horizontal flourish at the end.