# THE ALLELOPATHIC EFFECTS OF YAUPON ON ASSOCIATED HERBACEOUS SPECIES

by

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Submitted in partial fulfillment of the requirements of the University Undergraduate Fellows Program 1978-1979

Approved by:

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April 1979

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I would like to express my sincere gratitude for the diligent efforts and guidance extended by Dr. Fred Smeins throughout the entire course of this study.

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#### ABSTRACT

A field survey showed a definite decrease in species diversity of herbaceous vegetation beneath yaupon (<u>Ilex vomitoria</u>) canopy in comparison to vegetation in open areas between yaupon clumps. Several laboratory experiments were performed to test the possibility of inhibition due to excretion of toxic substances by yaupon into the environment.

The effects of aqueous extracts from plant materials and associated soils on germination and initial growth of cucumber (<u>Cucumis sativus</u> L.) and wheat (<u>Triticum aestivum</u> L.) were studied. The following materials were collected from mature yaupon stands for use in aqueous extract pre-paration: fresh yaupon leaves, leaf litter, yaupon roots, soil from within a yaupon canopy, soil from an open, grassy area, and yaupon berries. Strong inhibition of test seed germination and growth did not occur in potting dishes when water extracts were applied: There was no significant difference in germination or growth in leaf and stem of the test subjects after treatment with extract solutions when compared with the controls; there was no apparent difference among the extracts obtained from the various yaupon plant parts or soils.

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#### INTRODUCTION

Yaupon (<u>Ilex vomitoria</u>) is an evergreen, many-stemmed shrub that grows to a maximum height of 8 m with a trunk diameter up to 30 cm. It occurs throughout the Southeastern Coastal Plains of the United States from Virginia to Florida and South Central Texas (Vines 1960). Yaupon, a good browse plant for deer, is sometimes utilized by cattle. The fruit provides a food supply for birds. However, dense stands of this plant compete with more desirable and productive forage grasses (Baur, Bovey and Diaz-Colon 1970).

Overgrazing and other kinds of land disturbance, as well as the removal of the occurrence of natural fires has transformed the original open post oak savannah in Central Texas into brushy or dense woodlands. Yaupon and other woody species have formed thickets with the resultant decrease in the desireable forage grasses. The once wide-spread little bluestem (<u>Schizachyrium scoparium</u>) has been replaced in many areas by other less productive grasses and other herbaceous species. In addition, an overall decrease in diversity of herbaceous species and density of herbaceous growth is observed on the edge and beneath the yaupon canopy. It has been suggested that yaupon contains a germination or growth inhibitor which may contribute to the invasion of yaupon and disappearance of valuable grasses of the original post oak savannah.

Jensen (1955) detected the presence of a factor in the outer covering and pulp of yaupon fruit that inhibited elongation of wheat (<u>Triticum</u> <u>aestivum</u> L.) seedlings. He found that the inhibitory factor was nonvolatile and persisted in dry fruits. Bovey and Diaz-Colon (1969) found similar effects of yaupon fruit extracts on the elongation of cucumber (<u>Cucumis sativus</u> L.) seedlings.

The purpose of this study is to determine the relationship of the shrub yaupon to other plant components of its community and to further explore the allelopathic potential of yaupon. The knowledge gained from this type of research may enable us to control the detrimental effect of yaupon on both livestock and wildlife rangeland.

### LITERATURE REVIEW

A glance at the natural landscape is sufficient to show that plant species tend to be grouped in different combinations, forming more or less definite communities. In other words, the individuals of each plant species are not scattered at random, but are distributed in a pattern over the land.

A plant must respond to an environment that is conditioned by other plants. Two major ways in which plants may alter the environment, and therefore determine what other plants may live in an area are: 1) competition for scarce resources. The supply of water, nutrients, and solar energy is almost never adequate to permit maximum development of every individual plant in the community. 2) excretion of a toxic substance into the environment, a phenomenon referred to as allelopathy (Daubenmire 1947).

An allelopathic effect is defined as any direct or indirect harmful effect by one plant on another through the production of chemical compounds that escape into the environment. Allelopathy is thus separated from competition which involves removal of, or reduction of, some factor from the environment that is required by another plant sharing the area. Allelopathy depends upon a chemical compound being added to the environment by a plant (Rice 1974).

Studies of competition have for a long time ignored the possibility of chemical inhibition. But if two plants are in such close proximity that they can draw upon the same supplies of water and mineral nutrients, they are close enough to be immersed in, or touched by, one another's biochemical products. As allelopathy is recognized in growing numbers of vegetational situations, it becomes obvious that many of the early studies of

competition for minerals, sunlight, and water need to be re-evaluated.

Reviews concerning the entire field of allelopathy have been published by Loehwing (literature until 1937), Evenari (literature until 1949), Bonner (literature until 1950), Martin (literature to 1957), Woods (literature to 1960), and Winter, Garb (literature to 1961), Moreland (literature to 1966) and Holm (with special consideration on agricultural lands, literature to 1969) (Diaz-Colon unpublished).

The possibility that plants secrete substances which affect the growth of other plants was apparently recognized by DeCandolle in 1832. Interest in toxic secretions of plants arose from a consideration of "soil sickness". It was observed in the nineteenth century that as one piece of ground was continuously cropped to one plant, the yields decreased and could not be improved by additional fertilizer. His postulate was that the deleterious effect of continuous one-crop agriculture might be due to toxic excretions from roots. He considered that such substances were important in plant distribution (Kreb 1972).

A variety of agricultural problems of economic importance, such as "soil sickness" in apple cultures and the "peach-replant" problem, have been attributed to allelopathic effects. Very often it is impossible to replant apple trees on the same soil, and growers are compelled, therefore, to plant apple seedlings in soils never before used for apple growing. Besides an impoverishment of the soil regarding microelements and damage to the roots by nematodes, root residues become increasingly important. The assumption that root residues have an effect on apple replants is based on the results of Fastabend, cited from Börner (1960). He revealed that if water was leached through sick soil containing apple tree roots, a soil sickness developed if the leachate was added to healthy soil. These results were confirmed with apple seedlings, cultured in water solution containing root bark. As little as 1 g of air dried root bark of a 16 year old apple tree per 500 ml nutrient solution reduced the root and stem growth to 50% within 30 days (Diaz-Colon unpublished).

The peach replant problem was explored by Proebsting (1950). In 1922, at Davis, California, peach and apple orchards were planted; in 1942, these trees were removed, and the whole area was planted in Faye Elberta peach trees in the spring of 1943. Within one year, it was clear that the peach trees succeeding apples were growing better than the peach trees succeeding peaches. Proebsting points out one notable characteristic of the replant problem: It is highly specific; not all subsequent crops do poorly (Kreb 1972).

In the early 1900's, several agronomists commented on the effect of black walnut trees (Juglans nigra) on nearby grass and alfalfa plants. Massey (1925) observed that the zone of dead alfalfa around a walnut tree extended over an area 2 to 3 times greater than that covered by the tree canopy and suggested that this zone was determined by the outer limits of walnut roots. The roots were suspected of secreting a toxin to which some plants, for example alfalfa (lucerne) and tomatoes, were susceptible; other plants, for example corn (maize) and beets, showed no ill effects (Kreb 1972).

Davis (1928) extracted from the roots and hulls of the black walnut, a crystalline substance called juglone (5-hydroxy-1,4-napthaguinone) and showed that this chemical would kill tomatoes and alfalfa plants. Brooks (1951) found no evidence of antagonism between walnut and other timber tree species that normally occurred together with black walnut in forest stands, and he emphasized the selective effects of the walnut toxin: Some

species like alfalfa are killed; others like Kentucky blue grass become more abundant than usual near walnut trees (Kreb 1972).

Allelopathy is a wide spread phenonomen in nature and found in a variety of communities, taking form in a variety of ways. The distribution of annual plants in desert areas is not random and early observations indicated that some shrubs were rarely associated with annual herbs, whereas other shrub species harbored large number of annuals. Went (1952) observed this pattern in the deserts of the southwestern United States: The shrub <u>Encelia farinosa</u> A. is very rarely associated with annual plants. This observation gave rise to the assumption that toxic substances excreted by these plants are responsible for this situation.

Gray and Bonner(1948) demonstrated that <u>Encelia farinosa</u> leaves contain a chemical that inhibits the growth of tomato plants growing in sand in the laboratory. Water extracts of leaves were also inhibitory, and Gray and Bonner were able to isolate the toxic substance, 3-acetyl-6 methoxybenzaldehyde. This inhibitor was shown to have no effect on <u>Encelia</u> <u>farinosa</u> itself. Dried leaves retained their toxicity for several months. Gray and Bonner noted that this toxic effect was less striking on tomato plants grown in rich garden soil, perhaps because soil microorganisms destroyed the chemical inhibitor (Kreb 1972).

Vegetation patterning in other communities has also been attributed to allelopathic effects of dominant species. The chaparral of southern California is a mixture of shrubs with a sparse understory of herbaceous vegetation. Recurrent fires at intervals of 10 to 40 years destroy this dense shrub cover, and in the first growing season after the fire a luxuriant growth of herbs is produced. The shrubs regenerate principally by root sprouting. As the shrubs regenerate, seeds stop germination, usually about 5 to 6 years after the fire, and herbs become sparce again. Even in open stands of shrubs with 50 percent of the ground bare and adequate rainfall, there is no seed germination. Muller, Hanawalt, and McPherson (1968) cleared some chaparral stands by clipping close to the soil surface and removing the shrubs without disturbing the soil litter. In the next growing season, these clipped plots showed rapid germination of 30 herb species and appeared like an area one year after a fire; adjoining uncleared areas again produced no germination. Thus the fire cycle of the California chaparral is a sequence of events that follow from the destruction of toxic chemicals and the shrubs that produce them (Muller, Hanawalt and McPherson 1968).

In southern California, shrubs such as the aromatic <u>Salvia</u> <u>leucophylla</u> and <u>Artmisia</u> <u>californica</u> are often separated from adjacent grassland by a bare area 1 to 2 m wide. Apparently volatile terpenes, which are released from the leaves of these aromatic shrubs, are able to inhibit growth in nearby grasses (Rice 1974).

Other chaparral shrubs, such as the common "chamise", <u>Adenostoma</u> <u>fasciculatum</u>, secrete allelopathic agents from their leaves; this produces the noticeable lack of herbs in these chaparral stands, while adjacent grasslands and woodlands have dense herbaceous populations. McPherson and Muller (1969) reported a series of experiments designed to explain why chamise stands harbored few herbs. Competition for light, mineral nutrients, soil moisture, and oxygen were eliminated as possible causes by field experiments. Another suggestion was that grazing by small rodents and rabbits might be responsible for the shortage of herbs. To test this idea, McPherson and Muller placed small screened enclosures in <u>Adenostoma</u> stands and found that some of the effect was in fact caused by animal damage, but most of it was produced by leaf toxins.

The leaves of mature Adenostoma fasciculatum shrubs accumulate toxin on their surfaces during the dry summer months. This toxin is produced as a result of normal metabolism and is water soluble. When the rains of each new growing season commence, the toxin is dissolved and carried to the soil. Additional increments of toxin, proportional to the length of interrain periods, are added to the soil with each rain. It is highly probable that the toxin is concentrated in the upper 1 to 3 cm of soil by absorption to soil colloids and so becomes localized in the horizon where it must function. The first significant rains each season bring about germination of annual plants in the region. The seeds of these plants are also localized in the uppermost soil and litter horizons. Nearly all seeds in the soil of mature Adenostoma stands are prevented from germinating by the toxin which is most abundantly present during the normal germination period. Limited numbers of seeds of a few resistant species do germinate under these circumstances, but almost invariably the resulting seedlings fail to reach reproductive maturity. Root growth of these herbs is probably suppressed to such an extent that the slightest drought and resultant drying of the uppermost soil layers, kills the seedling. It is unlikely that the toxin kills seedlings directly at the concentrations which exist in the soil. Thus, although the physical factors of the environment such as light, water, oxygen and nutrients, are available in adequate supply, virtually no herbaceous flora is present in mature Adenostoma stands (McPherson and Muller 1969).

Abandoned cropland in Oklahoma goes through a sequence of plant succession that can be shown as follows (Booth 1941): weeds--Annual grass (Aristida oligantha)--bunch grass (Andropogon scoparius)--tall-grass prairie. The order in which grasses invade abandoned cropland is apparently controlled both by dispersal rate of seeds and by nutrient requirements. The usual explanation for successional changes in old fields is that each stage increases the organic matter of the soil and improves soil structure and water capacity so that a new set of species can take hold. This has not been an adequate explanation for the early stages of oldfield succession in Oklahoma (Rice 1974). The weed stage in the field is replaced very rapidly by the grass Aristida oligantha, but this grass can live in worse soil and water conditions than the pioneer species it replaces. Rice postulated that the weed species produces chemical inhibitors which affect themselves but not A. oligantha, and this explains the rapid succession. A. oligantha, in turn improves the soil slowly, but it secretes chemicals that retard nitrogen-fixing bacteria, thereby slowing the rate of succession to the bunch grass, Andropogon scoparius. This bunchgrass invades very slowly because of a low rate of seed dispersal and because it requires more soil nitrogen and phosphorus. Finally, the tall-grass prairie species become established as the soil becomes improved, but this may require 50 years or more (Kreb 1972).

Diaz-Colon, Baur and Bovey (1970) observed that yaupon seed grown in Texas appears to possess an extremely long dormancy period and germination and growth has been observed only when seeds were planted in soils under natural conditions for one year or more. Consequently, these plants may contain inhibitors which influence plant growth and development in itself. Experiments with aqueous extracts of yaupon fruit showed that the fruit inhibited root growth of germinating cucumber seedlings. Extraction of fruit of varying degrees of maturity indicated that the concentration of the inhibitors in mature fruit was greater. The bare zones around the edge and beneath the yaupon canopy, and its rapid spread into the post oak savannah, suggests yaupon may have allelopathic effects on associated herbaceous species.

### STUDY AREA

The study area is located on the Vernon A. Young Natural Area near Easterwood Airport, 2 miles west of Texas A&M University, College Station, Texas. The study area is located within Brazos County which lies in the heart of the East Texas Post Oak Belt (Gould 1975).

The most prevalent soil type of the area is Lufkin fine sand loam with 1 to 3 percent slopes and is characterized by a hard clay pan layer.

The experimental plots are situated in an upland post oak area in which yaupon and other shrubby hardwoods create a dense thicket-like vegetation. Overstory species include post oak, winged elm, blackjack oak and Eastern red cedar<sup>1</sup>. The herbaceous plants in the openings are predominantly brownseed paspalum, broomsedge bluestem, little bluestem, silver bluestem, Florida paspalum, <u>Aristida</u> spp., and tall dropseed.

Although the vegetation and diversity of herbaceous growth is fairly lush in the open areas, the vegetation thins out near the edge of the yaupon canopy, and is essentially absent from areas beneath the yaupon canopy.

Recent climatological data has been recorded at Easterwood Airport, which is adjacent to the Vernon A. Young Natural Area. The long-term annual average precipitation is 39.20 inches (99.6 cm). The rainfall for the months during which this research project was in progress (August 1978 through April 1979) was high in comparison to long-term averages. The temperature extremes reached during the period of study were a high of  $38^{\circ}C$  (8/24/78) and a low of  $-10^{\circ}C(1/2/79)$ . The long-term average for

<sup>&</sup>lt;sup>1</sup>See appendix for scientific names.

January is 50.2<sup>0</sup>F (10<sup>0</sup>C) and for August is 84.5<sup>0</sup>F (39<sup>0</sup>C). The growing period has a length of approximately 259 days.

### Survey of Herbaceous Vegetation

In March 1979, a field survey was carried out to determine what herbaceous species live in the area and their spacial relationship to yaupon. Areas representing the following three quadrat classes were sampled: 1) grassland--quadrats in the open areas 2) edge--quadrats on the edge of the yaupon canopy (drip-line) 3) inside--quadrats beneath the yaupon canopy.

Twelve 50 x 25 cm quadrats were surveyed for each yaupon clump: three quadrats each for the north, south, west and east side of the clump. This eliminated the possibility of species variability resulting from orientation of the clump. In each quadrat the presence or absence of each species was noted. The frequencies of species represented in each quadrat class was averaged. Two yaupon clumps, a total of 24 quadrats, were sampled.

### Field Experiment

A field experiment was designed to test the effect of removing the aerial portions of yaupon shrubs under controlled conditions. The results bear upon the allelopathy hypothesis, as well as the possibility of environmental factors (light, temperature, soil moisture, wind, etc.) that might cause the change of herbaceous vegetation. It was hoped that some of the variables inherent in a natural field situation could be eliminated.

Accordingly, in November 1978, four plots, approximately 2' x 2', were established on the study site: two plots within yaupon stands, two plots in open, grassy areas. Plot A was established in a yaupon clump and the vegetation canopy was left intact; Plot B was established in a yaupon clump and the shrubs were cut 2 to 4 inches above the soil. Cut-off shrub tops were removed from the area. Plot C was established in the open, grassy area and the grass was clipped to as near soil surface as possible. The cut grass was removed from the area. Within each 2' x 2' plot, four sections of equal size were set-up. Two of the sections were cleared of litter and two were left unaltered. In each of the 16 sections, 8 cucumber seeds (<u>Cucumis sativus</u> L.) and 8 rye grass seeds (<u>Lolium</u> sp.) were planted in rows.

Three days after the plots were established and seeds planted, a hard freeze hit, killing many of the seedlings. Therefore, the results of the experiment were rendered quantitatively invalid.

### Laboratory Experiments

Yaupon plant materials and soils were collected from a mature yaupon stand on the study site near College Station, Texas, to be used in preparing stock solutions for the experiments described below. Four laboratory experiments were performed during the months August 1978 through March 1979.

The following plant materials and soils were collected:

- 1. Fresh leaves from the outer branches of a yaupon shrub.
- 2. Leaf litter from beneath the yaupon canopy.
- 3. Yaupon roots from the top few centimeters of soil beneath the yaupon canopy (These are very fine, hair-like roots).
- Soil from the top few centimeters of soil beneath the yaupon canopy (soil-in).
- Soil from the top few centimeters of soil in the open, grassy areas (soil-out).

6. Ripe yaupon berries from the outer branches (These were only collected for the one experiment in November).

To test the allelopathy hypothesis and its implications, several laboratory experiments were designed. All of these involved a bioassay technique, using cucumbers(<u>Cucumis sativus</u>) as test subjects. Cucumber seeds were chosen because they are large and easy to work with, have a high percentage of germination, and the results could easily be compared with tests by other experimenters who used cucumbers as test subjects.

The seeds were irrigated with an extract solution prepared from plant material or soil; controls were irrigated with distilled water. Three replications were made for each extract test. The cucumber plants were allowed to grow for 6 to 11 days. Results of the number of plants germinated and measurements of stem and leaf length were recorded; results were expressed as average percent germination and mean lengths within replications.

Experiment 1. The first experiment was designed to test for inhibiting effects of prepared aqueous extracts upon germination of cucumber seeds. The plant material used in extract preparation was collected from the south side of a well-established yaupon clump in early September, 1978. Five extract solutions were prepared from the material collected: fresh leaves, leaf litter, roots, soil-in, and soil-out. Ten grams, fresh weight, of the various plant materials and soils were extracted with 90 ml of distilled water for 45 minutes with mechanical stirring. The mixture was vacuum filtered and the resulting solution was diluted to 100 ml with distilled water. The solutions were then refrigerated in plastic bottles.

The bioassay was conducted using 9 cm glass petri plates with filter paper seed beds (Whatman #1 filter paper). Ten cucumber seeds were placed

in each dish, covered with a second piece of filter paper and irrigated with the appropriate extract. The petri dishes were covered with glass covers. Three replications of each of the 5 extracts and the control were prepared. The eighteen petri dishes were placed in a germinator and incubated in the dark at  $30^{\circ}$ C for 14 days. Some plants had germinated in each of the dishes at the end of the third day, but the dishes were badly infected with fungus, so as to make any quantitative results invalid.

Experiment II. Due to the poor results obtained from the germinator test, another approach to laboratory experimentation was followed for the remaining four experiments.

Plant materials and soils were collected on October 6, 1978, from the same yaupon stand as in Experiment I. The extracts were prepared blending 20 g of fresh material with 150 ml of distilled water for 10 minutes (using a Waring Blender B-1711). The resulting solution was left to stand for 30 minutes, and then vacuum filtered. The extracts were diluted 100 percent with distilled water, bottled, and refrigerated.

The seed beds were prepared by using 6 inch plastic potting dishes and a 1:2:2 mixture of Perolite:sand:potting soil. Approximately 1.5 cm of soil was put in each dish, 10 cucumber seed planted, and another very thin layer of soil added to each dish. The soil was saturated with 20 ml of distilled water and 10 ml of the appropriate extract. The dishes were irrigated with the extracts (or distilled water for the controls) every 2 or 3 days. On the eleventh day, the experiment was terminated and the measurements of growth and germination were recorded.

In addition to the standard five extracts of Experiment I (fresh

leaves, leaf litter, roots, soil-in, and soil-out), two additional extracts were prepared with oven dry soil from within the yaupon canopy (soil-in dry) and from the adjacent open area (soil-out dry). Three replications of each extract test and the control were prepared, resulting in 24 dishes. The soil in the dishes dryed out and plants wilted before the final measurements were recorded.

Experiment III. The procedures followed in Experiment II were repeated since the measurements resulting from the previous experiment were not reliable. Plant materials and soils were collected in early October. The same techniques used for seed bed and extract solution preparation were followed as in Experiment II, except the solutions resulting after blending 20 g of material with 150 ml distilled water were not filtered, but rather simply diluted 100 percent with distilled water, bottled and refrigerated. Ten seeds were planted in each of the 24 dishes, and the soil saturated with 70 ml distilled water and 10 ml of appropriate extract (or distilled water for the controls).

The dishes were covered with plastic wrap to avoid drying. After seven days the plastic wrap was removed and the plants were irrigated with the appropriate extract solution or distilled water every 2 days thereafter. The experiment was terminated on the eleventh day; the plants were counted and stem and leaf lengths were measured and recorded.

Experiment IV. On November 3, 1978, another trip was made to the study site to collect plant material and soils for extract preparation. The yaupon berries were abundant and ripe, so along with the standard five extracts an extract of yaupon berry was prepared. Extract solutions and seeds beds were prepared as in Experiment III. The seedlings were

allowed to grow for twelve days; The number of plants germinated was counted and measurements of stem and leaf length recorded.

Experiment V. In early spring (March 3, 1979) another laboratory experiment was set-up. Plant materials and soils were collected from the study site (the berries were no longer attainable). Extracts were prepared by mixing 10 g of material with 150 ml of distilled water in an electric milk-shaker for 15 minutes. The solution was allowed to stand, refrigerated overnight, and then vacuum filtered. The filtrate was stored in plastic bottles in the refrigerator.

Seed beds were prepared in 6 inch plastic planting dishes, using only sterile sand as the substrate. Four cucumber seeds (<u>Cucumis sativus</u>) and six wheat (<u>Triticum aestium</u>) seeds were planted in each dish. The soil was saturated with the appropriate extract (or distilled water for the controls) and the dishes were covered with plastic wrap. The plants were allowed to grow for eight days. The number of plants germinated and stem and leaf lengths were recorded.

### RESULTS

### Field Survey

Results of the field herbaceous vegetation survey (Table 1) show a decrease in the number of herbaceous species represented in quadrats beneath the yaupon canopy in comparison to quadrats in open areas: a decrease from 28 species in the open to 5 species beneath the canopy. There is also a slight decrease in species diversity in quadrats established on the edge of yaupon canopy in comparison to open grassland quadrats (25 species and 28 species, respectively). Two of the valuable grass species exhibit a sharp decrease in frequency in areas beneath yaupon canopy: Brownseed paspalum exhibits a frequency of 100 percent in open areas and only a 25 percent frequency beneath the yaupon canopy; bluestem decreases in frequency from 12.5 percent in open areas to 0 percent beneath yaupon canopy. It is also interesting to note that yaupon seedlings were found only along the edge and beneath the yaupon canopy, suggesting a stimulation of growth/germination.

Although it is not demonstrated in the data, the density of herbaceous vegetation sharply decreases from grassland area to the area beneath the yaupon canopy. The vegetation in the open was lush and many individual plants of each species were present; the vegetation thinned out along the edge of the yaupon canopy and much more soil surface was exposed. Beneath the yaupon canopy, only a few sprouts were found of the species recorded. These were usually young seedlings which most likely would not survive the season. Results show definite inhibition of herbaceous growth beneath the yaupon canopy and along the edge of the canopy, whether by allelopathic effects or some other cause.

### Chart 1.

### FREQUENCY OF HERBACEOUS SPECIES WITH REFERENCE TO RELATIVE LOCATION OF YAUPON

All values represent averages of specie frequency in 8 25x50 cm quadrats

3/79

brownseed paspalum100Sesbania sp.100plantain87.5bluet87.5oldfield threeawn87.5camphorweed87.5yaupon0pinweed62.5our-clover75forb 150Dxalis sp.25oluestem12.5Sedge62.5Scribner's panicium37.5dewberry50geranium37.5western ragweed25call dropseed0elm seedling12.5Duncus sp.25	100 75 100 87.5 75 75 75 75 75 75 50 25 50 62.5 12.5 37.5 12.5	25 12.5 0 0 0 0 0 0 87.5 0 0 0 0 0 0 0 0 0 0 0 0 0
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hervil 37.5	0	0
coadflax 12.5	12.5	0
orb 2 12.5	12.5	0
ed lovegrass 12.5	12.5	0
roton 12.5	0	0
ainlily 12.5	0	0
enuslookingglass 0	12.5	0
ignalgrass 12.5	0	0
row-poison 12.5	0	0
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tal number of		

### Field Experiment

The November 1978 field experiment yielded no quantitative results due to the hard freeze, yet some conclusions can be made from observation of the 4 plots. Cucumber plants and rye grass were present in all 4 plots--in yaupon stands, as well as in the open. This suggests that, if germination is inhibited by yaupon, it is not inhibited 100 percent under these experimental conditions.

#### Laboratory Experiments

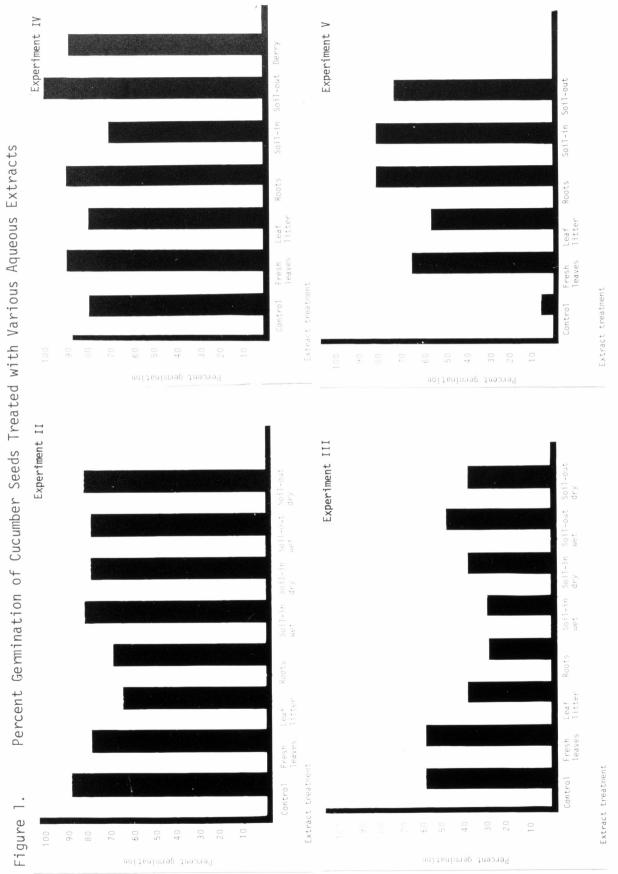
Laboratory experimental results indicate that the aqueous extracts of yaupon plant materials and associated soils do not significantly inhibit growth nor germination of cucumber plants and wheat grass.

Figure 1 illustrates germination success of cucumber plants after treatment with various extracts in the 4 successful experiments. There was little significant variation between extract treatments within experiments, and no significant variation from the controls. Where variation is exhibited, there is no consistency between experiments. For example, germination of seeds treated with "leaf litter" extract in Experiments II and III were less than the respective controls; in Experiments IV and V, germination of seeds treated with this extract were better than the controls. Also, "soil-out" extracts should have functioned as controls, since no allelopathic substance should be present in the grassy areas. Yet, germination of seeds treated with "soil-out" extracts was erratic and results are inconclusive.

Results obtained from measuring stem and leaf length of cucumber plants show similarly inconsistent results (Figure 2). There is little significant variation in growth between extract tests and no significant variation from the controls. Where variation is exhibited, there is no

consistency between experiments.

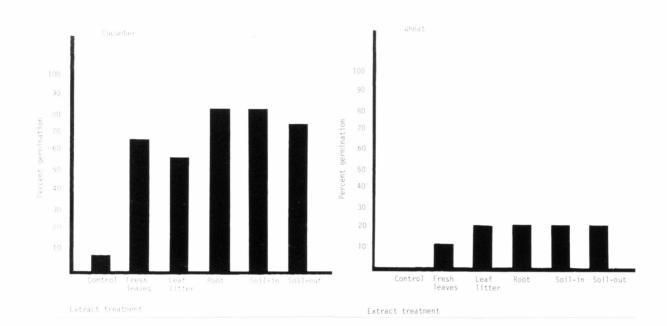
Figure 3 illustrates results obtained in Experiment V, where both cucumber seeds and wheat seeds were planted in sterile sand and treated with test extracts. Both cucumber and wheat showed poorer germination and growth under the control conditions than after treatment with any of the extract solutions. The wheat seedlings, as a whole, showed poorer germination than the cucumber seedling; but this is probably due to the nature of the plant species rather than a greater response to an allelopathic effect of wheat in comparison to cucumber.

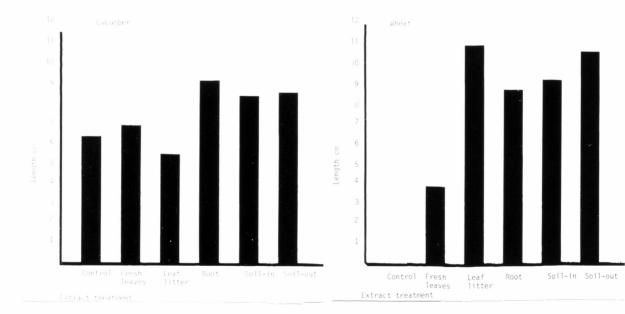


Stem and Leaf Length of Cucumber Seedlings Treated with Various Aqueous Extracts Figure 2.



Figure 3. Percent Germination and Stem Length of Cucumber and Wheat Seedlings Treated with Various Aqueous Extracts. Experiment V.





#### DISCUSSION

The experimental results obtained from bioassays of aqueous extracts of yaupon plant material and associated soils did not indicate the presence of a water soluble inhibitory factor in extract solution which affected germination/growth of cucumber or wheat grass. However, field surveys of herbaceous vegetation exhibit a sharp decrease in species diversity and density in areas beneath yaupon canopy in comparison to open, grassy areas.

It is possible that the inhibition of herbaceous growth is caused by competition for environmental factors, such as light, water, or minerals. However, light does not appear to be a critical factor, since many of the yaupon stands are quite open and even the most dense stands have occasional openings in the crowns. This uncovered ground, exposed to full sunlight, fails to exhibit herbaceous growth comparable to open areas. Also the areas around the edge of the yaupon canopy, which receives sufficient sunlight, show a decrease in species diversity and density.

It is more likely that the inhibition of herbaceous growth is due to a shortage of soil moisture during seasonal droughts. The yaupon shrub possesses a dense mat of fine roots which occupy the first few centimeters of soil beneath the yaupon canopy--the soil layer where it would most effectively inhibit herbaceous growth. The dense mat of roots extends to the edge of the yaupon canopy and thins out at a rate which is proportional to the increase in herbaceous growth.

Allelopathic effects of yaupon on associated herbaceous growth should not be entirely ruled out. Toxic substances may be excreted into the environment during seasons or periods which were not encompassed in this project. It is possible the yaupon excretes a toxic substance that is not

water soluble or excreted into the environment by some means in which these tests could not detect (for example: volitilization). Since many allelopathic excretions are highly selective as to their target subject, it may be possible that native species are inhibited by an toxic excretion from yaupon to which the test subjects (cucumber and wheat) are not sensitive.

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APPENDIX

### Table 1.

Extract Treatment	Mean Percent Germination	Average Stem Length (cm)	Average Leaf Length (cm)
Control (distilled water)	90	2.5	1.0
Fresh Leaves	80	2.0	1.0
Leaf Litter	70	3.5	1.0
Yaupon Root	70	5.2	1.0
Soil-in (wet)	80	2.6	1.0
Soil-in (dry)	80	3.8	1.0
Soil-out (wet)	80	4.2	1.0
Soil-out (dry)	80	4.2	1.0

Experiment II. Germination and Growth of Cucumber Seedlings Treated with Aqueous Extracts of Yaupon Plant Material and Associated Soils.

Cucumber seedlings were allowed to grow for eleven days; however, the dishes dryed out and plants wilted before final measurements were made.

### Table 2.

Experiment III. Germination and Growth of Cucumber Seedlings Treated with Aqueous Extracts of Yaupon Plant Material and Associated Soils.

Extract	Mean Percent	Average Stem	Average Leaf	_
Treatment	Germination	Length (cm)	Length (cm)	
Control (distilled water)	60	10.2	2.7	
Fresh Leaves	60	8.9	2.2	
Leaf Litter	40	9.0	1.9	
Yaupon Root	30	8.5	1.9	
Soil-in (wet)	30	11.0	3.0	
Soil-in (dry)	40	8.0	2.3	
Soil-out (wet)	50	10.1	2.4	
Soil-out (dry)	40	9.2	2.1	

Cucumber seedlings were allowed to grow for eleven days.

### Table 3.

Extract Treatment	Mean Percent Germination	Average Stem Length (cm)	Average Leaf Length (cm)
Control (distilled water)	80	10.6	3.1
Fresh Leaves	90	11.5	2.9
Leaf Litter	80	10.4	2.6
Yaupon Root	90	10.3	2.5
Soil-in	70	11.2	2.7
Soil-out	100	10.6	2.5
Yaupon Berry	90	11.0	2.1

Experiment IV. Germination and Growth of Cucumber Seedlings Treated with Aqueous Extracts of Yaupon Plant Material and Associated Soils.

Cucumber seedlings were allowed to grow for twelve days.

### Table 4.

Experiment V. Germination and Growth of Cucumber and Wheat Seedlings Treated with Aqueous Extracts of Yaupon Plant Material and Associated Soils.

Extract Treatment	Mean F Germir	Percent Nation	Average Length		Average Leaf Length (cm)
	Cucumber	Wheat	Cucumber	Wheat	Cucumber
Control (distilled water)	8.3	0	6.5	0	1.5
Fresh Leaves	66.7	11	7.1	4.0	1.7
Leaf Litter	58.3	22.2	5.6	11.2	1.6
Root	83.3	22.2	9.3	8.9	1.8
Soil-in	83.3	22.2	8.6	9.4	1.7
Soil-out	75	22.2	8.8	10.8	1.7

Seedlings were allowed to grow for 8 days.

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	tall dropseed	Sporobolus asper
venuslookingglass <u>Triodanis</u> <u>sp</u> .	toadflax	<u>Linaria</u> sp.
	venuslookingglass	<u>Triodanis</u> <u>sp</u> .

Scientific Name

Table 5. Common and Scientific Names Cited

Common Name

continued

### Table 5. continued. Common and Scientific Names Cited.

Common Name

Scientific Name

western ragweed

Hymenopappus artemisiaefolius

Shrubs and Trees

blackjack oak	Quercus marilandica
eastern redcedar	<u>Juniperus</u> virginiana
post oak	Quercus stellata
winged elm	<u>Ulmus</u> alata
yaupon	<u>Ilex</u> vomitoria

Test Subjects

cucumber	<u>Cucumis</u> <u>sativus</u>
rye grass	<u>Lolium sp</u> .
wheat grass	<u>Triticum</u> aestivum