ELF Communications System Ecological Monitoring Program: Summary of 1987 Progress

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A long-term Ecological Monitoring Program is being conducted to monitor for possible effects from the operation of the U.S. Navy's ELF Communications System to resident biota and their ecological relationships. Monitoring studies were selected through a peer reviewed, competitive bidding process in mid-1982, and work on most studies began in late summer of that year. Preliminary activities of the Program consisted of site selection, characterization of critical study aspects, and validation of assumptions made in original proposals. Subsequently, increasing emphasis has been placed on the collection of preoperational and operational data bases at the Michigan and Wisconsin Transmitting Facilities. The data bases are being used to make proposed spatial and/or temporal comparisons of biological and ecological variables. This report summarizes the progress of the Ecological Monitoring Program during 1987.

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FOREWORD

The U.S. Department of the Navy is conducting a long-term program to monitor for possible effects to resident biota and their ecological relationships from the operation of the Navy's Extremely Low Frequency (ELF) Communications System. The program is funded by the Space and Naval Warfare Systems Command through a contract to IIT Research Institute (IITRI). IITRI provides engineering support to the program and coordinates the efforts of ecological investigators. Monitoring projects are being conducted under subcontract arrangements between IITRI and study teams.

This report summarizes the activities of the Extremely Low Frequency (ELF) Communications System Ecological Monitoring Program during 1987. This is one of a series of reports[1-5] that document the yearly progress of the Program. IITRI also compiles annual reports of subcontractor progress[6-11] as well as reports on engineering support activities.[12-16] All of these reports are unclassified and are submitted to the National Technical Information Service for unlimited distribution. Project personnel are also encouraged to report their findings in presentations and as publications (see Appendix A).

Since 1983, summary reports[1-5] have chronicled the results of wildlife surveys performed at the Wisconsin Transmitting Facility by the U.S. Forest Service. Surveys of ruffed grouse, eagle, and deer populations were performed annually commencing in 1974, 1975, and 1982, respectively, and continued through 1986. There were no effects to these populations from the operation of the ELF Communications System, and the studies were concluded after the 1986 surveys.

Respectfully submitted,
IIT RESEARCH INSTITUTE

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1. INTRODUCTION

1.1 PURPOSE

The purpose of the Ecological Monitoring Program is to determine whether electromagnetic (EM) fields produced by the Navy's ELF Communications System will affect resident biota or their ecological relationships.

1.2 ELF COMMUNICATIONS SYSTEM

The complete ELF Communications System consists of two transmitting facilities, one located in the Chequamegon National Forest in Wisconsin and the other located in the Copper Country and Escanaba River State Forests in Michigan (see Figure 1). Each facility consists primarily of a transmitter building connected to long overhead wires (antennas) with buried ground terminals at each end. Both the antenna and grounding elements are located in cleared rights-of-way (ROW). The transmitters broadcast messages using ELF EM fields; these fields are the operational components to be monitored.

EM exposure from the ELF Communications System can be conveniently divided into preoperational, transitional, and operational phases. During the pre-operational phase, biota received no EM exposure from the ELF Communications System. The transitional phase began with the initiation of system testing; exposures during this phase are intermittent and at lower intensities than intended for an operational system. When the system achieves full operational capability, EM exposure will be nearly continuous and at full intensity. The achievement of a fully operational capability at the Wisconsin Transmitting Facility (WTF) occurred during the last quarter of 1985; intermittent operation of the Michigan Transmitting Facility (MTF) was initiated in the second quarter of 1986 and continued through 1987.

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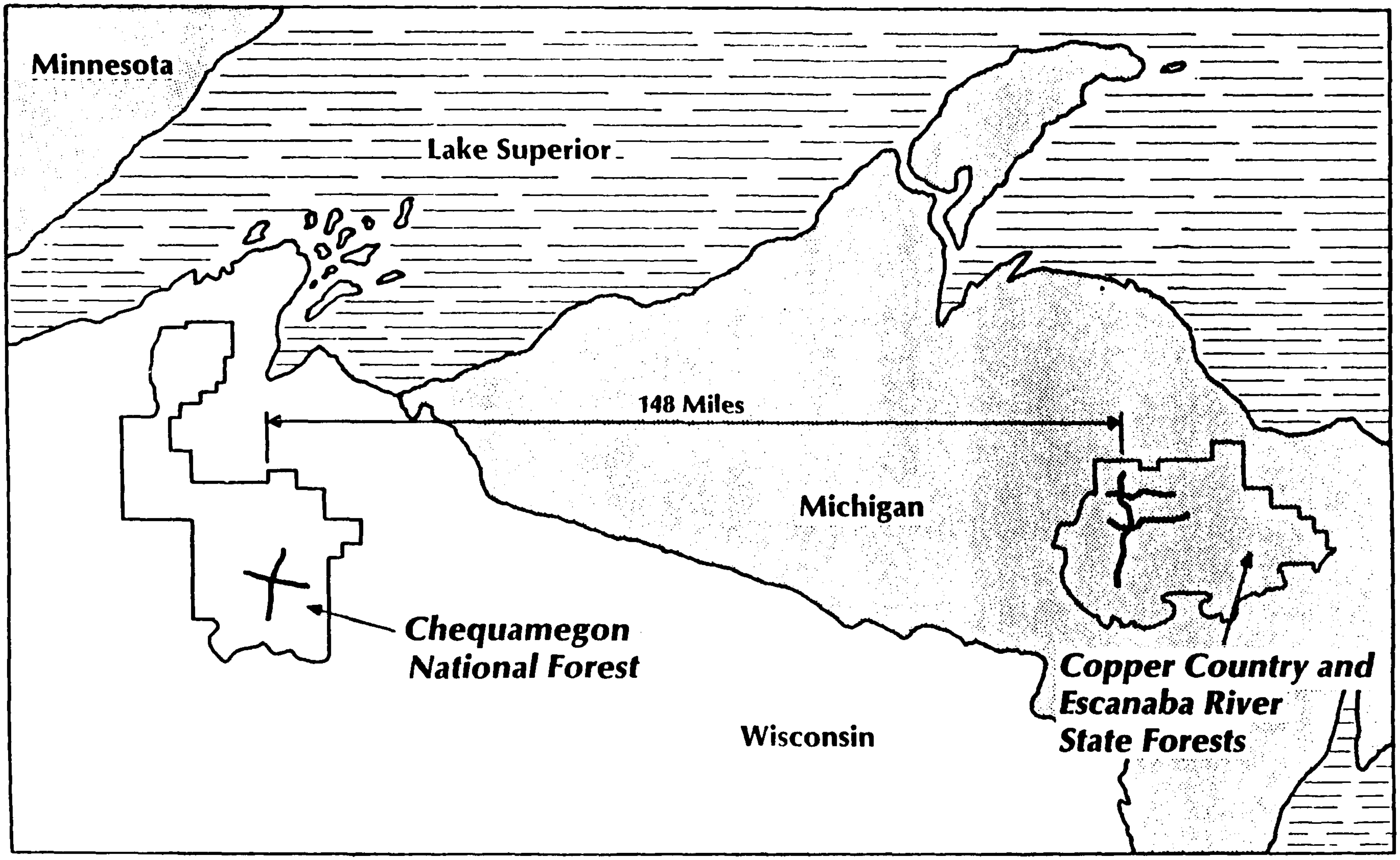

FIGURE 1. ELF COMMUNICATIONS FACILITIES IN WISCONSIN AND MICHIGAN.

1.3 ELF EVALUATIONS AND RECOMMENDATIONS

Research on possible EM effects to biota from the operation of an ELF Communications System began in 1969. Although some ecological and wildlife studies were performed in the ensuing years, the major emphasis was on laboratory research. In 1977, the Navy and the National Academy of Sciences (NAS) examined the information produced by these studies as well as studies performed at other ELF frequencies. Specific research at planned operating EM conditions of the ELF system, as well as research at other ELF frequencies, showed no acute bioeffects. Those effects reported were small and/or were controversial among researchers. The Navy and the NAS concluded that adverse effects to biota from the operation of the ELF system were unlikely. After reviewing similar types of information reported during the period 1977-1984, the American Institute of Biological Sciences reached the same conclusion as the Navy and the NAS. Nevertheless, all reviewing organizations recommended that a program be conducted in the ELF Communications System area to monitor for possible changes to resident biota.

A monitoring program was recommended partly because of the limitations of research performed in the laboratory. Although laboratory research is an important approach, it emphasizes study of select attributes of individuals; examines relatively homozygous individuals of a limited number of species over short periods of time; and does not simulate all of the potential synergistic factors present in the natural environment. In addition, the impact of environmental perturbations such as EM fields can be expressed at one or more levels of biological organization, i.e., organisms, populations, communities, and ecosystems. Our current state of understanding does not allow an accurate prediction of ecological effects from laboratory studies of individuals, particularly if the effects are small or subtle.

1.4 MONITORING PROGRAM DESIGN

In its 1977 environmental impact statement, the Navy outlined a preliminary plan for conducting an ecological monitoring program at those sites approved for operation of the ELF Communications System. The initial plan was developed from the results of laboratory research, input from state agencies, and recommendations made by the NAS for long-term ecological monitoring. The elements of the initial plan were refined based on comments submitted in response to the Navy's draft environmental impact statement. In 1981, concurrently with approval to complete construction of an ELF Communications System, the Department of the Navy funded an Ecological Monitoring Program.

Ecological studies are of fundamental importance because they integrate the responses of many biota. The purpose of these kinds of studies is to examine group characteristics such as productivity, abundance, and decomposition processes. This approach is the best method for detecting possible, marked effects to the disparate species such as are resident in the ELF Communications System area. One limitation to this approach, however, is that a sizable effect must be manifested in ecological variables in order for researchers to detect it. Generally, the numbers of replicates taken for ecological variables in this Program are adequate to ensure detection of differences of greater than 40 percent.

The Program also includes research for possible *in situ* effects at the organismal level. These studies focus on specific attributes of abundant and ecologically significant organisms. Although narrower in scope, organismal studies generally are more sensitive than ecological studies. Large data sets can easily be collected for organismal studies, thus enabling the detection of small differences when making comparisons. Except for the slime mold project, every project in the Program couples organismal level studies with monitoring at the population and/or community levels.

The general types of organisms for study were selected on the basis of ecological importance and the likelihood of their being perturbed by EM fields, irrespective of the field's intensity or frequency. Sixteen general types of organisms from three major ecosystems in the ELF Communications System area are being examined. The principal criterion for selecting specific biota was their presence in sufficient numbers to ensure meaningful statistical comparisons.

Both spatial and temporal comparisons of biological and ecological end points are being examined.

Spatial comparisons are being made by obtaining data relatively close to the overhead wires and grounds of the ELF Communications System (treatment or test sites) and at greater distances from these antenna elements (control sites), as shown in Figures 2 and 3. As in classical experimental design, the

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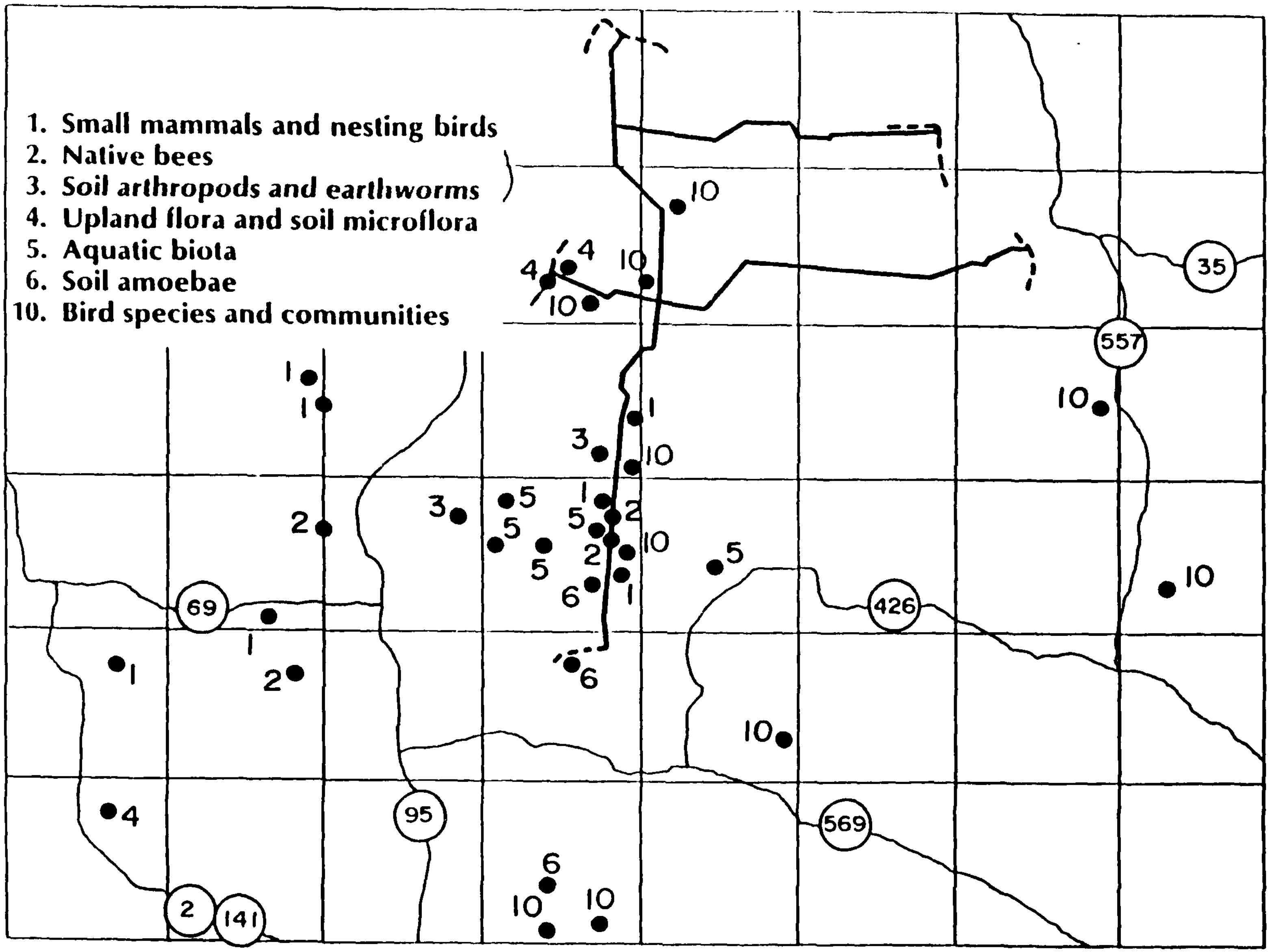

FIGURE 2. FIELD SITES FOR MICHIGAN ECOLOGY STUDIES.

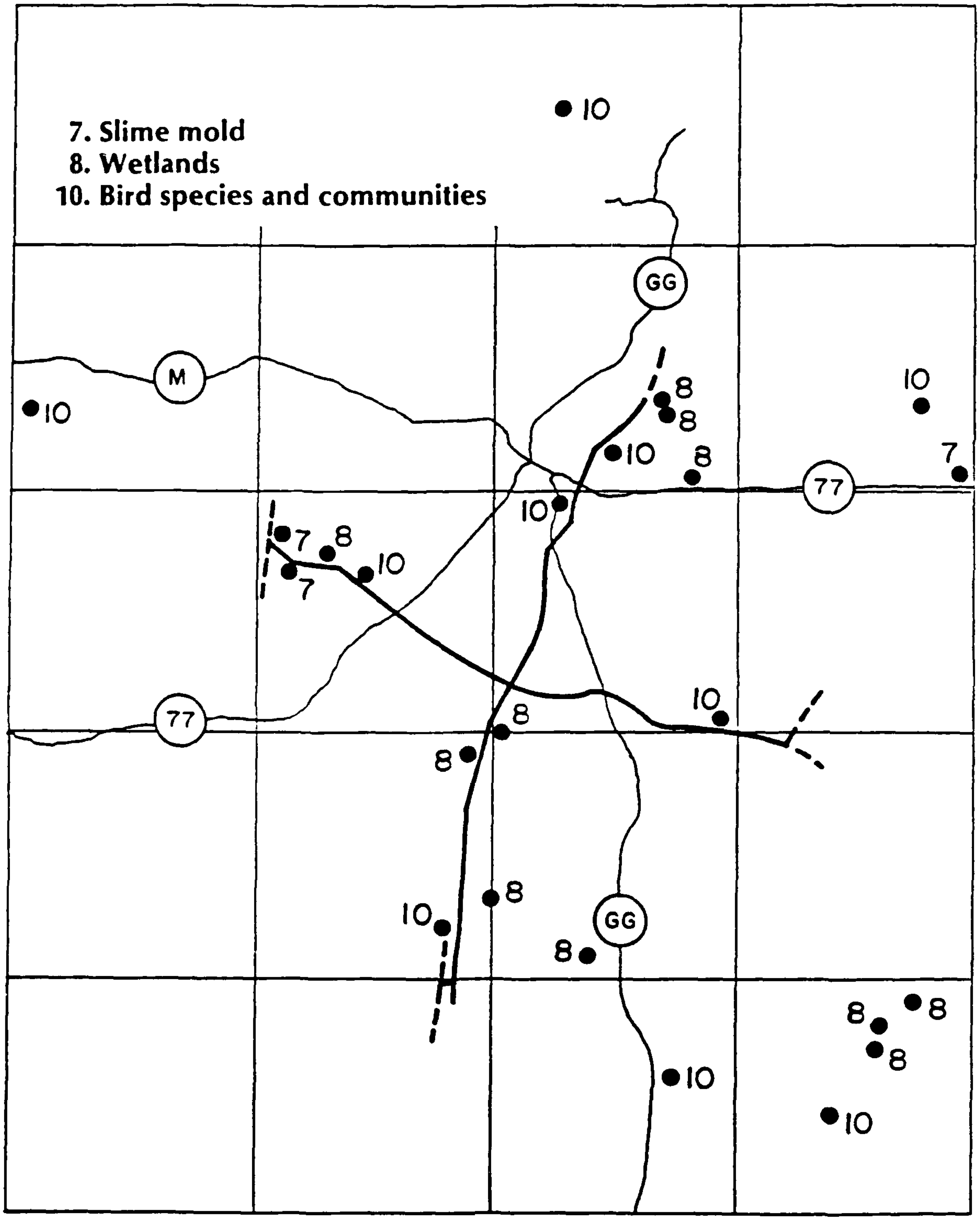

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FIGURE 3. FIELD SITES FOR WISCONSIN ECOLOGY STUDIES.

control is used to determine the effect of environmental (ambient) conditions on a variable, while the treatment site is used to measure the effects of ambient conditions plus those of the higher EM field intensities produced by the ELF system.

Temporal comparisons of biological and ecological variables are being made between the preoperational and operational phases of development of the ELF system. Multiyear studies are necessary in order to evaluate a fully operational ELF system at both transmitting facilities (see Figures 4 and 5); these studies have been initiated in both Michigan and Wisconsin. Both temporal and spatial comparisons are being made in Michigan. Comparisons in Wisconsin have been spatial only, because the transmitter there has been operating in a transitional mode since 1969 and a preoperational data base does not exist.

In summary, the Program emphasizes ecological studies but also includes investigation for possible *in situ* organismal effects. A program of long-term, site-specific monitoring is under way.

1.5 PROGRAM DEVELOPMENT

The major objectives of each study during the early years of the Program were the selection of study sites, the validation of assumptions made in the proposals, and the characterization of critical study aspects. These objectives encompassed such activities as:

- identification of biota
- assessment of data collection protocols
- quantification of spatial and temporal patterns for each variable
- assessment of biological and ecological variability

As these tasks were accomplished, increasing emphasis was placed on the collection of data and the refinement of statistical protocols.

The technical progress of each study is reviewed annually by at least four scientific peers. Critiques are provided to the principal investigators for their consideration. In addition to this annual peer review, an overall program review by a statistician was initiated in 1986 and continued in 1987.

In Wisconsin, sample collection for wetland flora and slime mold studies was completed during 1987. Both studies will complete their analytical efforts in 1988 and provide a report covering the term of their performance. In Michigan, eight studies continued to collect data and further develop their analytical protocols.

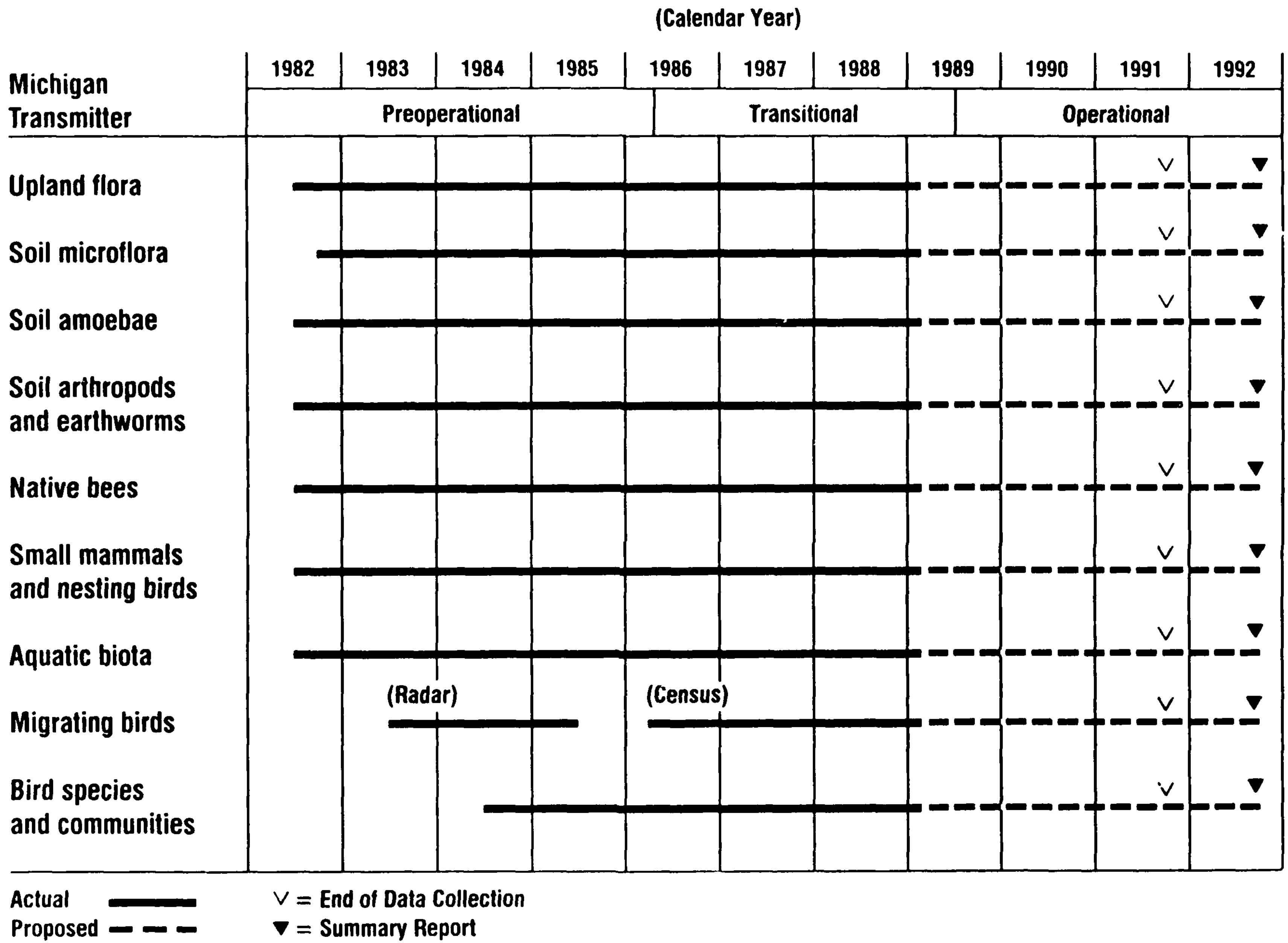

FIGURE 4. ACTUAL AND PROPOSED SCHEDULE FOR BIOLOGICAL AND ECOLOGICAL STUDIES IN MICHIGAN.

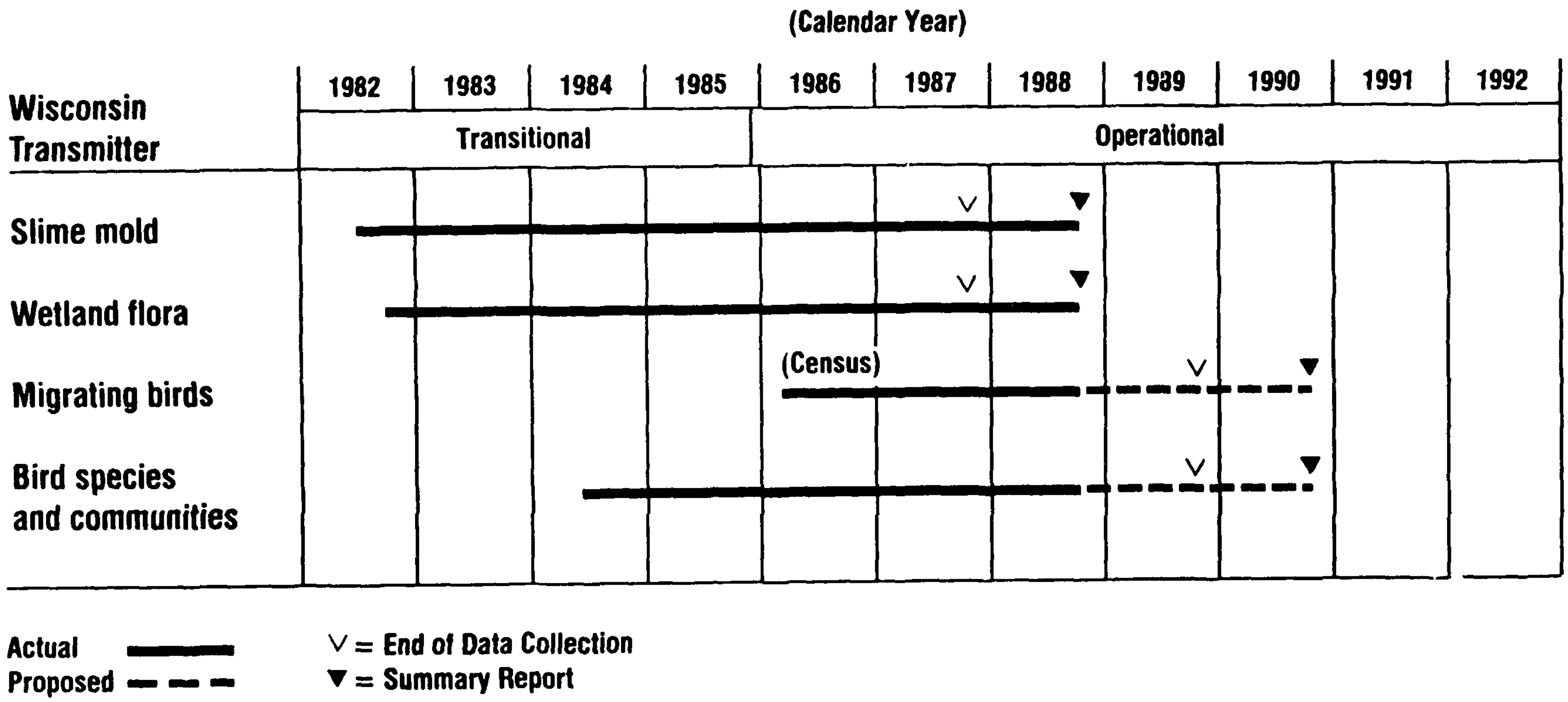

FIGURE 5. ACTUAL AND PROPOSED SCHEDULE FOR BIOLOGICAL AND ECOLOGICAL STUDIES IN WISCONSIN.

2. STUDY DEVELOPMENT: 1987 PROGRESS

This section summarizes the progress for each of the 10 studies that constituted the Program during 1987. A more detailed presentation of study protocols, methodology, and progress is given in individual project reports.[11]

The general types of biota being examined are used as subsection titles, while specific study elements are presented as underlined titles in each subsection. In order to simplify presentation of statistical results, any difference described as "significant" had a significance level of 5 percent ($P < 0.05$).

2.1 UPLAND FLORA

Forest vegetation (trees and herbs) is the dominant biota in the ELF Communications System area. The production of organic compounds by vegetation and the subsequent degradation of these compounds comprise the main method of transfer of energy and nutrients to other organisms. Organic matter turnover and distribution are regarded as major determinants of the forest ecosystem's structure. Because the production and distribution of organic matter have been shown to be measurably affected by anthropogenic factors, these processes and associated organisms are being monitored for possible effects from the ELF Communications System. The progress of the upland flora studies is presented here; that of the soil microflora studies is presented in Section 2.2.

In order to examine for possible changes in forest productivity and health, the following objectives are being examined:

- growth rates of established tree stands and pine seedlings
- phenological events of trees, herbs, and mycorrhizal fungi
- numbers and kinds of mycorrhizae on red pine seedlings
- nutrient levels of hardwood and pine foliage
- foliage production in hardwoods
- insect damage, disease, and ambient environmental factors

Treatment sites are located adjacent to the antenna and grounding elements of the MTF. A single control site is located more than 28 miles from the nearest antenna element. The antenna and control sites each consist of overstory tree plots (existing pole-size stands), plots planted with red pine seedlings, and plots for the study of herbaceous plants. The grounding treatment site consists of plots planted with red pine only. No tree stands or herbaceous plots were established at the ELF system grounds because the buffer strips required to eliminate "edge effects" would have placed the study plots at too great a distance from the grounding elements for meaningful EM exposure.

Tree Growth. The purpose of this element is to examine tree growth on both hardwood and red pine tree stands.

Diameter, height, ingrowth, and mortality are being monitored in order to gain a complete picture of tree growth and stand production at pole-size plots on study sites. Tree species being monitored on pole-size stands are oak, birch, aspen, and maple. Changes in diameter increment is the primary response variable used for monitoring stand growth. Because diameter is strongly correlated with total tree biomass, it is also used for estimates of stand production. Permanently installed dendrometer bands allow continual measurement of incremental growth on each tree in the stand. Bands are read weekly from 1 May until annual growth is nearly complete.

At present, four years (1984-1987) of diameter change data has been collected at pole-size study sites. Analysis of tree diameter increment data was accomplished using two analytical methods: (1) split plot analysis of covariance was used to determine if there were changes in the level of average yearly diameter growth due to exposure to ELF fields, and (2) the development of regression models continued so as to more accurately quantify the relationships between tree growth, site factors, and climatic variables.

An intensive screening was performed using correlations to select independent covariates prior to analyses of covariance. The covariates selected were soil temperature degree days, air temperature degree days, water holding capacity, and potassium concentration. Analysis of covariance showed no significant differences between sites in diameter growth rates. Except for paper birch, there were no significant differences between years in diameter growth rates. Finally, there were no significant statistical interactions between sites and years in the growth rates of the species examined. This analysis indicates that low current testing in 1987 did not affect the previous relationships of growth rates between treatment and control sites.

Many of the relationships between diameter growth and tree, site, and climatic variables are nonlinear. Because nonlinear relationships between covariates and response variables may result in inaccurate results, the development of growth models continued for each of the four species of trees.

At each of the three study sites, red pine seedlings are permanently marked for monitoring of total height, basal diameter, terminal bud length, and physical condition. These variables are recorded at the end of each growing season. For evaluation of height growth patterns, a subsample of the marked seedlings is measured weekly from April until shoot elongation is complete.

Annual height and diameter growth in 1987 were analyzed using split plot analysis of covariance. Covariates used in the 1987 height analysis were height in 1985 and temperatures during 1986. Covariates used in 1987 diameter growth were diameter in 1984, 1987 root volume, 1986 soil moisture, and 1986 temperatures. The use of the previous year's climatic data explained more site and yearly variation than the current year's data. There were no significant differences between sites or between years (1986 and 1987) for height or diameter growth of red pine. The results indicate no detectable effect on annual growth from exposure to ELF fields during 1987.

The rate or timing of growth during 1985, 1986, and 1987 was evaluated by models that predict total growth at a given time during the growing season. There were significant differences between years for the model's growth coefficients; however, there were few significant differences between sites in a given year. Researchers feel that the model must be further refined by quantifying the effects of additional climatic and site factors on growth coefficients.

Mortality of pine due to a root disease, first documented in 1986, increased during 1987. Although the percentage mortality is still small, it appears that conditions at the study sites are conducive to the spread of the disease. During 1987, multiple regression models were developed and tested that relate site and ambient factors to frequency of root disease. Birch stump frequency is the most important factor in explaining pine mortality frequency. Additional regression analysis during 1988 will evaluate select soil factors and pine mortality.

Phenological Events. Previous studies have indicated that exposure to EM fields may alter the timing of some physiological events, e.g., mitosis. The purpose of this element is to monitor select phenological events and morphological characteristics of plants in the ELF Communications System area. The timing of annual events and morphological characteristics of short-lived plants (herbs) are addressed here; those of long-lived plants (trees) are presented under Tree Growth, above.

The starflower, *Tridentalis borealis*, flowers frequently and is a common, abundant herb in the ELF Communications System area. The flowering and leaf expansion of at least 200 naturally growing starflower plants are followed each year on plots reserved at the antenna and control sites. When tagged plants are removed from the sample, usually due to herbivory, other plants are added to the sample so as to maintain the sample size at 200 plants. Observations are made every three days in the early part of the growing season and weekly thereafter.

Phenological events (e.g., time of leaf expansion) generally occur earlier on the control site than on the antenna site. However, using analysis of covariance techniques, there were no significant differences between sites and few differences between years for the time of stem expansion, leaf expansion, or leaf area expansion. Covariates used in the statistical analyses were soil temperature, soil moisture, and relative humidity. Additional analyses will be performed in 1988 to examine other covariates as well as other phenological events such as flowering, fruiting, and leaf senescence.

Morphological characteristics of the starflower (i.e., number of buds, flowers, fruit, and leaves) are also being monitored. Leaf area has been statistically examined as an overall morphological characteristic, and no significant differences between sites or between years were found. Preliminary nonstatistical examinations indicate that some characteristics may differ between sites.

Herbaceous Plants. Herbaceous plants have been found to be a more sensitive, short-term indicator of environmental perturbations than trees. Accordingly, the relative frequency of occurrence and percent cover of herbaceous plants on all study sites are being monitored for possible EM effects from the operation of the ELF Communications System.

Herbaceous plants and shrubs on subplots at pole stands and plantations were identified and enumerated and their cover estimated. These surveys were conducted in August, when species diversity and plant biomass were at their maximum.

Relative frequency, relative cover, and importance (percent ground cover of a species multiplied by its frequency of occurrence) values for each species were calculated. During 1987, analytical efforts focused on evaluating changes in species importance values on each site for the years 1985, 1986, and 1987 and on using discriminant analysis to determine whether ELF fields affect the importance of various herbaceous species present on study sites.

There have been significant changes between years in the importance of various herbs and shrubs present. The source of these changes cannot be ascertained, but could be natural succession or changes in climatic conditions as well as increased testing at the MTF. Researchers will concentrate their analytical efforts on evaluating different statistical techniques. In addition, permanent herbaceous plots will be established in order to make a more consistent interyear evaluation of species composition.

Mycorrhizal Populations and Root Growth. Mycorrhizal fungi form a symbiotic relationship with the roots of higher plants such as trees. The fungi utilize organic compounds synthesized by the tree for their growth and to "forage" for minerals and water in the soil. In turn, the fungi provide the tree with minerals and water more efficiently that the tree's roots alone. This relationship is considered essential to the satisfactory growth of nearly all tree species. Because the growth of fungal mycelia are dependent on physiologically produced intracellular currents, other sources of electrical currents, such as the ELF Communications System, may have an effect on the fungi and, indirectly, on trees. The population dynamics of mycorrhizae occurring on hardwood and pine trees are being examined.

The population dynamics of mycorrhizae on hardwoods are being evaluated through nondestructive monitoring of sporocarp (mushroom) production at plots located at the antenna and control sites. Surveys are performed after reports of fruiting are first received from field crews, and continue until fruiting has stopped.

Sporocarps from at least 32 species have been identified from surveys made since 1984. During 1987, the highly variable distribution of sporocarps was examined by the development of multiple linear regressions using seven of the more common fungus species. In 1988, differences between years will be examined by incorporating these results into covariate analyses.

At pine plantation sites, the population dynamics of mycorrhizae are being characterized as the frequency of occurrence of mycorrhizal types and the number of mycorrhizal root tips per red pine seedling. Due to the increase in the size of seedlings and increased numbers of mycorrhizae, a subsampling method was tested in 1986 and implemented in 1987. Mycorrhizae per gram of root weight continues to be the parameter examined, but its basis is lateral root weight rather than total root weight.

When 1985-1987 data were collectively examined, there were no significant differences between sites or between years by site for total mycorrhizae per gram of lateral root weight. There were some significant differences between months when comparing years; however, this can be attributed to the effects of seedling establishment in 1985. In 1985, seedlings had significantly lower numbers of total mycorrhizae early in the season.

In 1987 as in previous years, type 3 was the most common and type 5 the second most common mycorrhizae on the roots of the pine seedlings. Type 5 mycorrhizae have generally increased in frequency of occurrence on all three sites since the start of the study. Examining 1985-1987 data for frequency of occurrence showed the same results as obtained for total mycorrhizae.

With or without covariates, a difference of 10 to 15 percent in the total number of mycorrhizae is necessary to distinguish a significant difference between sites. Similarly, a difference of 15 to 25 percent is necessary to distinguish a significant difference between years.

Litter Production and Foliar Nutrients. The purpose of this element is to examine total litter weight, total leaf litter nutrient content, and the nutrient content of oak foliage during the growing season. Total litter weight and litter nutrients provide estimates of seasonal canopy production as well as an estimate of input to the decomposition system (see Section 2.2). A determination of foliar nutrient content makes possible the detection of changes in physiological processes of trees such as nutrient translocation.

Litter is collected in traps on existing hardwood stands at the antenna and control sites. The litter is dried, sorted, and weighed according to the following categories: foliage, wood, and miscellaneous. A subsample is taken to determine the nutrient content of the foliage. Foliage samples are also taken from the crowns of red oak trees periodically during the growing season for nutrient determinations.

In 1987, major litter fall began on 24 September and was complete by 1 October. This litter fall period was similar to that of 1986 but was earlier than in either 1984 or 1985. An analysis of variance showed no significant differences between antenna and control sites for any of the three litter components (foliage, wood, miscellaneous) examined. When litter weights on both sites were combined, a significantly greater foliage and miscellaneous litter fall occurred in 1986 than in other years. There were no significant differences between years in litter fall when the covariates of soil and air temperature were used in the analysis of variance. These results indicate that all three litter components can be used in statistical comparisons; however, the detection limit for foliage alone (12 to 17 percent) is a more sensitive indicator than total litter. Future comparisons of litter production will be limited to foliage litter.

The lack of significant differences between sites or between years indicated that the 1987 testing of the antenna in Michigan had no detectable effects on tree litter production.

Analysis of nutrient concentrations in actively photosynthesizing red oak foliage (1984-1987) showed no significant differences between sites for most nutrients. Chemical analyses of litter samples taken in 1987 have not been completed; however, statistical analyses were performed on data collected over the period 1984-1986. Average nutrient content of mixed litter showed few significant differences between sites. When the foliage was separated by species and analyzed, significant differences between sites remained. However, the results indicate that nitrogen, calcium, and magnesium concentrations of foliage litter and active red oak foliage will be suitable for monitoring possible effects to tree nutrient translocation and cycling.

2.2 SOIL MICROFLORA

Soil microflora (bacteria and fungi) play a key role in the maintenance of upland forest ecosystems such as those in the ELF Communications System area. Microflora transform (decompose) organic matter produced by forest vegetation (litter) and fix elements present in the atmosphere into a form available for plant uptake. Anthropogenic factors that disrupt these processes may directly alter the flow of nutrients to vegetation and thus indirectly affect the forest community.

The objective of this element is to monitor for possible effects of EM fields produced by the ELF Communications System on populations of streptomycetes associated with plant roots and on rates of decomposition and nutrient flux of litter. These objectives are closely related to the upland flora studies (particularly, the mycorrhizal and litter production objectives) described in Section 2.1.

Upland flora (producers) and soil microflora (decomposers) form a natural assemblage; however, these groups are being examined by separate subcontractors. Both subcontractors are with the Department of Forestry, Michigan Technological University, and both share common study sites and ambient monitoring systems.

Rhizoplane Streptomycetes. The purpose of this element is to characterize and enumerate streptomycete bacteria associated with red pine mycorrhizae (see Section 2.1). Streptomycetes have been reported to be involved in the nutrition of mycorrhizae and may influence mycorrhizae through their production of antibiotics or growth factors.

Sample sizes and protocols in 1987 were the same as in 1986. Samples were taken monthly from May through October from pine plantations at the antenna, ground, and control sites. Plate count data were transformed prior to analysis of variance in order to detect differences between years, sampling dates, and sites. Differences detected by analysis of variance were reexamined by analysis of covariance.

In 1987 as in 1985 and 1986, the streptomycete morphotypes B and F were the most commonly isolated at all three pine plantations (antenna, ground, and control). Seven other morphotypes were routinely detected each year in addition to B and F. Since 1985, the seasonal pattern has been for relatively high numbers of morphotypes in May and June, declining to seasonal lows in October.

There were no significant differences between sites in the numbers of morphotypes present in 1987. There were, however, significant differences between years in the numbers of morphotypes identified at study sites. Preliminary analysis of covariance with several environmental covariates explained some, but not all, of the interyear differences.

As in 1986, there were no significant differences between sites in the numbers of streptomycetes present during 1987. However, the numbers of streptomycetes were significantly higher in 1987 than in 1985 or 1986. As it had for the numbers of morphotypes, preliminary analysis of covariance explained some of the interyear differences.

Relatively stable streptomycete populations have become established at all study sites. Based on data collected since 1985, intersite comparisons appear warranted. Continued examination of covariates using analysis of covariance will help to identify those environmental factors responsible for interyear differences.

Litter Decomposition and Nutrient Flux. The purpose of this element is to determine the rates of decomposition and nutrient flux for the foliar litter of three species of trees (oak, maple, and pine) that are abundant in the ELF Communications System area. These variables are being used as indicators of the overall functioning of the litter community.

Litter is collected each autumn from a single location, weighed, and then either analyzed for nutrient content or enclosed in nylon mesh envelopes for emplacement at study sites. Envelopes contain either individual leaves or bulk foliage samples of a single species. Samples are emplaced at study sites in December and are retrieved monthly from April through November of the following year. Data are expressed as the percentage of original dry matter mass or nutrient mass remaining at the time of retrieval. Dry matter mass loss data from 1985 through 1987 are complete. Nutrient mass loss data for 1985 and 1986 are complete, while 1987 nutrient mass samples await chemical analysis.

Transformed data were examined by analysis of variance and analysis of covariance for differences between sites and between years. Correlation analysis showed strong relationships between mass loss and the following independent variables:

- weather (temperature and rainfall)
- initial leaf density
- foliar nutrient content

These independent variables were used alone or in combination for the analysis of covariance. Generally, there were no significant differences between sites for mass loss in bulk, and individual samples emplaced on hardwood stands and the foliage of all three species appeared to decompose faster in 1985 than in 1986 or 1987. There was no clear pattern between sites or between years for the decomposition of bulk and individual samples emplaced on pine plantations.

Efforts during 1988 will be to test the independence of the covariates from effects by EM fields and to identify other pertinent covariates for use in analysis of covariance. In the past, seasonal patterns of dry matter loss were analyzed by fitting each year's data to a single exponential decay model. In addition to the covariate analyses, comparisons of decomposition constants are planned for 1988.

Analysis of variance showed many significant differences between years in the nutrient content of parent litter collections used for dry matter and nutrient studies. The relationship between nutrient content of bulk litter samples and seasonal progress in mass loss were then evaluated by correlation analysis. Nitrogen, phosphorus, potassium, calcium, and magnesium were significantly correlated with the progress of mass loss. During 1988, nutrient levels will be tested for possible use as covariates to examine differences in rates of dry matter mass loss.

2.3 SLIME MOLD

Previous to this study, researchers reported that continuous exposure of the slime mold, *Physarum polycephalum*, to ELF EM fields in the laboratory depressed the mold's rate of respiration and lengthened its mitotic cycle. They are now seeking to determine whether similar effects occur when the mold is exposed *in situ* to EM fields and environmental conditions present at an ELF Communications System facility.

The project consisted of an *in situ* component performed at the WTF and a laboratory component performed at the University of Wisconsin-Parkside (UWP). *In situ* studies were performed at three WTF sites: two treatment sites located adjacent to a grounding and an aerial antenna element, and one control site located about seven miles from the nearest antenna element. The *in situ* studies allow assessment of possible effects from the operation of the ELF system, while the laboratory component of the project provides for a comparison of results obtained *in situ* and those obtained in previous laboratory experiments.

The *in situ* studies employed culture chambers that isolated the mold from other soil organisms. Ambient electrical fields and currents in the soil were collected by buried electrodes and were supplied to the chambers in conjunction with control and monitoring circuitry. At each site, two cultures were matched to ambient electric fields and one culture to ambient current densities. In 1987, data loggers were installed at each study site in order to more closely monitor the chambers for electrical variations. EM exposure regimes in the laboratory were simulated to match those at the WTF ground sites.

The variables examined during 1987 were respiration rate (QO_2) and adenosine triphosphate content (ATP). Although examined in 1985, mitotic cycle length was not measured in 1986 or 1987 because researchers felt that QO_2 and ATP were more sensitive variables. Data collection protocols in 1987 were essentially the same as used in 1986. The 1987 field season was the last for collection of data; during 1988, researchers will be analyzing data collected over the term of the monitoring project.

In Situ Studies. In addition to annual measurements by IITRI, EM field intensities at the study sites were measured by the principal investigators before removing cultures (weekly) and readjusted following subculture to match those existing in the soil. In order to ensure an adequate characterization of the electric fields and current densities in the culture chambers, EM data logging units were installed at all three study sites (see Section 3.3).

There were no significant differences between sites in the respiration rate or ATP content of slime molds grown at the WTF. Due to contamination, the use of backup cultures was required during weeks 10 and 13. If the data obtained from backup cultures are removed, a significant difference between sites is obtained for the ATP content, but not for the respiration rate, of the cultures. After adjusting for temperature differences, researchers also noted that the variables are changing with time. Statistical analyses suggest that the changes are not the same at all sites. A close examination of these changes is planned for 1988.

Laboratory Studies. Laboratory experiments for the QO_2 and ATP variables were performed in a manner similar to that used for the *in situ* studies. Molds were grown and subcultured on agar and then transferred to suspension culture for analysis. Treatment cultures were continuously exposed for 155 days to EM field intensities similar to those at the WTF ground site.

In 1987 as in 1985 and 1986, there were no significant differences between treatment and control cultures for either variable. However, the laboratory cultures exhibited significant changes over time in both respiration rate and ATP content. These changes did not seem to follow any orderly pattern, however, and the finding argues against the presence of a cumulative effect for duration of exposure.

Using analysis of variance techniques, investigators discovered that the length of time a culture was out of the EM field affected both the QO_2 and ATP variables. These analyses showed that the respiration rate was affected more than the ATP content. Steps were taken to reduce the length of time out of the EM field for those cultures transported from the WTF.

2.4 SOIL AMOEBAE

Soil amoebae are common soil organisms that are predators on bacteria. Bacteria, in turn, are important to the soil ecosystem because of their ability to mobilize nutrients needed for plant growth. To the extent that protozoa affect the numbers and types of bacteria in the soil, protozoa also become a potentially important factor in soil fertility. Studies on protozoa and other related organisms have suggested possible EM effects on characteristics such as orientation, growth, and physiology.

In order to examine for possible effects from the operation of the ELF Communications System, the following aspects of soil amoebae are being studied:

- species and strain characteristics
- population size and activity
- growth and feeding

In addition, select elements indicative of soil fertility are being monitored.

Studies on soil amoebae are being performed at three study sites in Michigan. Two treatment sites are located at aerial and grounded portions of the ELF Communications System. The third site, the control site, is located about nine miles from the nearest ELF system element. The aerial treatment site and the control site were the same as used in 1985; however, the ground treatment site had not been used prior to the 1986 field season. The ground site had to be moved because of a reconfiguration of the ELF grounding system.

Species and Strain Characterization. During the 1987 field season, eight types of amoebae (various generic and species levels) were isolated using soil enrichment techniques. To date, no differences between years or between sites have been reported in the types of amoebae present.

In 1987 as in previous years, the genetic diversity within a single species of soil amoeba, *Acanthamoeba polyphaga*, was determined by isoenzyme analysis. The number of genetic loci examined in 1987 was increased to 14 from the four examined in 1985 and 1986. The number of clones examined was increased from five at each site to 10 at each site. No differences were found between sites or between 1986 and 1987 for the genetic diversity of *A. polyphaga*.

Population Size and Activity. The size of the amoeba population is an ecological variable considered likely to influence the functioning of the soil system.

Soil samples for population studies are taken with a coring device. Coring locations within study sites are determined randomly using a numbered grid system and a random number generator. The soil profile at study sites is typical of northern hardwood soils, i.e., with a sharp difference between the upper, organic horizon and lower, mineral horizon. In a typical core the 1- to 2-in. organic horizon is taken as one sample, while the top 2 in. of the underlying mineral horizon is taken as a second sample. A soil-dilution counting technique is used to determine the population size of each sample.

Studies to date have shown that the total amoeba population at any given moment consists of both vegetative (actively reproducing) and encysted forms. During the growing season there are cyclic changes in the total number of amoebae present, often increasing or decreasing by two orders of magnitude over short periods of time. Using analysis of variance techniques, no significant differences between sites were found in the numbers of cysts present in the organic or mineral soil horizons. Although there were significant differences between sites in the total numbers of amoebae present during surveys in June and July, there were no significant differences between sites in total numbers during the latter portion of the 1987 season. No statistical comparisons between years were reported for cysts or total numbers.

The cyclic changes in the total amoeba population indicated to the researchers that both the vegetative and encysted forms were being actively destroyed. In an attempt to identify those organisms possibly affecting amoeba population dynamics, researchers isolated bacteria and fungi from study site soils. In 1985, they found that of 300 isolates, about a dozen were toxic, and two were lethal to soil amoebae. To date, no results on the findings of this study have been reported.

Growth and Feeding Activity. The purpose of this element is to determine the *in situ* growth and feeding activity (i.e., predation on bacteria) of soil amoebae in buried culture chambers.

The study involves suspending a known species of amoeba (*A. polyphaga*) with a food bacterium in a physiological saline, all contained in a culture chamber. In order to produce electric fields and electric currents similar to those in the surrounding soil, the chambers are connected to buried collecting electrodes. Periodic counts of amoebae and bacteria are made to determine changes in the numbers of organisms over time. Cropping activity will be determined by varying the number of bacteria for amoeba growth and then following the growth rate and maximum yield. This approach will allow examination of an important ecological function of amoebae (i.e., bacteria predation) by eliminating some of the variability inherent in the soil environment.

In 1984, preliminary studies of amoeba growth in culture chambers at Michigan study sites indicated no significant differences between sites for amoeba growth rates without EM fields. Brief studies using EM fields were performed in 1986 at the WTF. There were no significant difference between exposed and unexposed chambers in the growth rate of the amoebae.

During 1987, growth studies were performed both in the laboratory and at the MTF. In the field, several problems in protocol and equipment were identified; however, results on growth rates were not reported. In the laboratory, experiments examining amoeba growth and consumption of bacteria showed that the slopes of these two variables were the same at two different densities of amoebae. Bacterial suspensions incubated without amoebae did not change over the course of the experiment.

2.5 SOIL AND LITTER ARTHROPODS AND EARTHWORMS

Arthropods and earthworms play a major role in the decomposition of vegetation. These invertebrates shred plant material such as leaves and redistribute the remains in the soil habitat. The vegetative remains are then further degraded by soil microflora (see Section 2.2). For the purpose of detecting possible effects of the ELF Communications System on major agents of litter decomposition, this project is monitoring both the structural and functional aspects of the litter and soil invertebrate community.

The project employs one treatment site located adjacent to the antenna ROW at the MTF and one control site located at a distance west of the antenna. Both sites are situated in a maple-dominated deciduous forest. Although there are faunal differences between the sites, the sites have similar soils, vegetation, and microclimate.

In order to address faunal differences between sites, community indices and the characteristics of major populations common to both sites are emphasized. In addition to dominant groups, populations representing various roles in the soil habitat, such as predators and detritivores, are examined. To accommodate the various roles of the soil fauna may require intersite comparisons of ecological equivalents and/or preoperational and operational comparisons of populations unique to the treatment site. Litter decomposition rates will provide an overall indication of the functional aspects of the soil community.

__Arthropods__. This element examines the major arthropod fauna inhabiting the soil, litter, and surface layers at each site.

Diel and seasonal activity patterns of surface-active arthropods were assessed by consecutive, day and night, pit-trap samples taken once a week. In order to increase catches of surface-active arthropods, pit traps were provided with barriers that increase the effective area sampled by diverting moving arthropods toward the pit. Major groups trapped were springtails, mites, and ground beetles. Pit trap data for 1987, as well as some data for 1986, were incomplete at the time of reporting. The following paragraphs present information on springtails and ground beetles collected during 1985 and 1986.

There were significant differences between 1985 and 1986 in the total number of springtails trapped. Based on the available data, sites differed in the relative dominance of major species and in the occurrence of a few rare species. There were significant differences between sites and between years in the diversity of the springtail community. However, activity patterns of the major families of springtails were the same at both sites. During 1987, researchers used regression procedures to examine climatic variables as possible covariates in the activity pattern of one abundant species. Linear regression using temperature was the best approach for comparing activity patterns between sites. Future analyses will also examine density levels of springtails present in the litter as another covariate.

The total number of ground beetles trapped showed about a 14 percent increase from 1985 to 1986. Although the number of species remained about the same at both sites in both years, there were significant differences between sites and between years in the diversity of the trappable ground beetle community. Preliminary examination of activity patterns using linear regression statistical techniques showed no significant differences between sites within a given year. Activity patterns of beetles at a given site differed between years, apparently due to differing environmental factors. In addition to examining diversity and activity patterns, researchers are examining fecundity as another possible variable to be monitored.

The population and community dynamics of soil and litter arthropods are being determined from samples taken biweekly during the growing season. Litter and soil are sampled separately. The arthropods are then extracted by heat and sugar flotation techniques. Springtails and mites are the most abundant taxa in the litter and soil of both sites and are the major groups of interest. At the time of reporting, data for springtails were available for the period 1984-1986. Due to the large number of individuals and the difficulty in identifying species, information on the mite community is available only for the years 1984 and 1985.

The diversity and yearly mean density of the springtail community are comparable to those of other deciduous forest communities with similar temperatures and soil characteristics. Nevertheless, there were significant differences between sites and between years in both the diversity and density of the springtail community. Analyses indicated that differences between sites were marginal for only one representative soil species. Density estimates for this species may be used as a variable if appropriate covariates can be found. Researchers will continue to study these variables to determine if intersite differences persist. They also will continue to examine ambient conditions as possible factors influencing density and diversity.

There were significant differences between sites and between years in the density of select species of mites common to both study sites. Despite the discrepant density fluctuations, the developmental stage structure (frequency of occurrence) of each population examined was similar between sites and between years. Researchers are evaluating several variables related to mite abundance as possible covariates. They plan to use an analysis of variance method that will allow multiple-year testing of density over time.

Earthworms. The purpose of this element is to examine the major earthworm fauna inhabiting the soil and litter of the study sites.

Earthworms were extracted from litter using weak formalin, while those in soil were obtained by hand sorting followed by wet sieving. In 1987, earthworm samples were taken at biweekly intervals through July and at monthly intervals through October. Data from 1987 were not available at the time of reporting; the following summaries and analyses are based on data taken over the period 1984-1986.

Eight species of earthworms have been identified, of which three are common to both sites. As expected, species diversity indices are low but are comparable to Canadian worm communities. There were no significant differences between sites for community diversity when total numbers of worms per species were averaged over all three years. Preoperational and operational comparisons of community diversity are planned.

Population variables being examined are distribution, density, reproductive activity, and recruitment.

The vertical distribution of litter- and soil-dwelling earthworms was not significantly different between sites, and the vertical distribution of worms was governed mainly by moisture conditions. The horizontal distribution of earthworms over study sites was uneven and was independent of the presence of other worm species. The utility of horizontal distribution variables is questionable, and their continued use will be critically examined during 1988.

Average yearly densities and biomass of four abundant worm species were examined for preoperational and operational comparisons within the treatment site and/or for between-site comparisons. Estimates of changes that are detectable range from 15 to 28 percent in density or mass of adults. Although researchers did not consider these variables particularly sensitive, they will continue to monitor density and biomass as overall indicators of population trends. Other population characteristics such as cocoon weights and recruitment will be used as covariates to improve future analyses of worm density and biomass.

Cocoon weights are closely related to adult body mass and generally can be used as indicators of the physiological state of adults. During 1987, cocoons obtained in 1984 through 1986 were examined, recounted, and weighed. The weights of cocoons were characteristic for each species and varied little between years. Preliminary t-tests showed no significant differences between years for the average cocoon weight for four of the five species examined. Based on variances observed to date, estimates of detectable differences in cocoon mass between sites or years range between 3 and 8 percent at the 0.01 significance level.

Cocoon characteristics are also being used to examine the long-term consequences of reproduction and recruitment on overall population levels. Seasonal and year-to-year densities of worms vary as functions of new cocoons produced, old cocoons hatched, and the effects of ambient conditions (possibly including EM fields) on reproductive adults. Although there were significant differences between years in cocoon densities and numbers of immature worms, recruitment and growth patterns were similar at both treatment and control sites. These variables will be examined for continued use in intersite comparisons by analyses to be performed during 1988.

Litter Decomposition. This element examines the input, decomposition, and standing crop of leaf litter at each study site. These variables provide an estimate of the overall functioning of the soil and litter community.

Litter inputs were determined by collection of leaves in litter traps located at each site. Traps were emptied weekly during the time of greatest leaf fall and monthly at other times. Samples were sorted by category, then oven-dried, cooled, and weighed. Maple leaves constituted the bulk of the litter inputs. Differences between sites or years for total litter inputs were not statistically significant for data collected from 1984 through 1987. Input values are consistent with data reported for similar forests and latitudes.

Exponential decay rates of forest floor litter were calculated based on total annual litter inputs and maximum standing crop after leaf fall. Turnover times of 1.2 to 1.3 years were determined from the 1987 decay rates; these values were higher than those of previous years. There were no significant differences between treatment and control sites.

Sampling forest floor litter is inherently imprecise; therefore, less variable estimates of decay rates were also obtained by examination of mass loss from leaves of known initial weight. Samples of dried maple litter were weighed and placed in mesh netting on the soil surface at both study sites. At monthly intervals, 10 samples per site were retrieved, dried, and weighed. Correction for soil contamination was determined by combustion of ground samples and weighing the residue. At the time of reporting, 1987 samples had not been processed.

Values of 1986 decay rates determined from leaf packs yield turnover times of 1.3 to 1.4 years at the treatment and control sites, respectively. Forest floor turnover rates were estimated at about one year. Actual rates probably lie between the forest floor and leaf mass loss estimates. There were no significant differences between sites for 1986 turnover times.

2.6 NATIVE BEES

Enervated cells containing iron granules have been found in the abdominal segments of foraging honeybees. It has been speculated that these iron structures may be used in orientation and may provide a basis for the sensing of EM fields by bees. Behavioral changes such as increased dispersal, increased levels of activity, lowered overwintering survival, and modification of nest structure have been described as effects from fluctuations in the earth's magnetic field and from exposure to the EM environment associated with transmission lines.

Honeybees are rare in the forested areas in which the ELF Communications System is located. However, native bees are abundant and are of particular importance to ecological communities in the area as pollinators of the resident flowering plants. Native bees have coevolved with resident plants and are able to overwinter in the study area. Therefore, native bees, rather than honeybees, are being studied. Aspects of nesting activity, nest architecture, and the emergence of native bees have been monitored for possible EM effects from the operation of the ELF Communications System.

Observations on native bees have been made at two treatment sites and two control sites since 1983. Data on nesting activity were collected by direct observation as bees were constructing their nests. Information on nest architecture and emergence was collected using techniques that involve setting predrilled blocks of wood on shelved hutches at study sites ("trap nesting"). The biological aspects of the occupying bee species are then studied as the bees make use of the holes to construct their nests.

Each nest consists of a series of reproductive (cell) and nonreproductive (basal and vestibular) spaces within the bore of a hole. Each cell is lined with elongate leaves and is provisioned with pollen. After an egg is deposited, the open end of the cell is closed by a partition consisting of rounded leaves. The ends of the nonreproductive spaces are also closed with a series of plugs using rounded leaves and other material. Generally, the egg hatches and the larva molts through a series of stages to overwinter as a prepupa.

Over 40 species of native bees are known to occur in the ELF Communications System area, 20 of which will use trap nests. This study focuses on two abundant species, *Megachile inermis* and *M. relativa*.

<u>Nesting Activity</u>. Disorientation or agitation of bees while foraging or building nests may be reflected in the time taken to construct a nest and/or in the number or duration of foraging trips made by the bees.

From 1983 through 1986, an extensive effort was put forth in observing, recording, and determining the different activity patterns of various species of native bees. Statistical analyses performed during 1986 showed that efforts to record the time periods for construction and provisioning of cells with pollen required large amounts of observation time and yielded little and variable data. However, the timing of foraging trips for nest materials and the time spent manipulating the materials in constructing the nest were considered to be of particular relevance.

Based on these results, the following new protocols were implemented during 1987:

- emphasis on foraging behaviors
- more systematic sampling (i.e., equally over locations, season, and time of day)

- examination of a few trips for many different individuals (previously many trips for a few individuals were recorded)
- correlation of behaviors with ambient environmental factors

In 1987, the duration of trips to retrieve round leaves was found to be shorter at the control sites than at the treatment sites. Nevertheless, there were no significant differences between sites in the duration of the trips. There were significant differences within sites, between observers, between dates, and trip order. Steps will be taken during 1988 to reduce or statistically address these latter sources of variability.

Nest Architecture and Orientation. When honeybees were exposed to EM fields produced by a high voltage transmission line, their reproductive output was lowered, and they increased the amount of propolis at their nest entrance. If native bees respond to the EM fields produced by the ELF Communications System in a similar manner, they may alter architectural aspects of their nests such as reproductive spaces (cells), or produce thicker cell caps.

The available nest architecture data have been compiled, and most of the data have been entered into the computer. Researchers have not detected significant differences between sites or between years in cell lengths, cell volumes, or number of cells per nest for each of the two species being examined. Sources of significant variance have been identified as including the individual taking the measurement, order of the cell in the nest, and the sex of the offspring. In the latter case, cells with male offspring were significantly smaller on average than cells with female offspring. Protocols will be changed during 1988 to reduce and/or appropriately treat these sources of variation.

During 1987, researchers continued to collect information on nest plugs and number of leaves used to line cells. At the time of reporting, they were examining various methods for analyzing these data. We anticipate that analytical results will appear in the 1988 Annual Report.

Since honeybees may use the earth's magnetic field to orient their comb, it is possible that fluctuating ELF magnetic fields could disturb any preference that native bees have in orienting their nests. Previous analyses showed no preference of native bees for any cardinal compass point in orienting their nests. Analysis of data has continued, and the results of statistical analyses will be presented in the 1988 Annual Report.

Emergence and Mortality. High voltage transmission lines have been reported to lower the overwintering survival of honeybee colonies. In order to monitor for a possible similar effect in native bees, researchers are examining the proportion of nest cells that produce adults and the sources of mortality at test and treatment sites.

Completed nests were allowed to overwinter at study sites. During the spring, the nests were removed from the sites and taken to a laboratory, where they were split open and data on nest architecture were recorded. Cells were placed in individual plastic culture tubes and labeled with nest and cell identification numbers. Tubes were kept indoors at room temperature until emergence. Date of emergence, species, and sex of offspring were then recorded. Adults were released at the sites where their nest had been constructed the previous summer. Cells that showed no signs of emergence were opened and the contents were recorded to determined the condition of the bee.

At the time of reporting, data from 1986 had not been analyzed.

2.7 SMALL MAMMALS AND NESTING BIRDS

Some laboratory studies performed at EM intensities and frequencies similar to those produced by the ELF Communications System have indicated effects to small vertebrates. Although these reports are controversial, many species of small mammals and birds reside in the ELF Communications System area, and in principle, any could be affected by the operation of the system.

As in the case of other studies in the Ecological Monitoring Program, this study examines several levels of biological organization for possible effects from the ELF system. Community characteristics are being used to assay for possible effects to many different species of resident mammals. In addition, two abundant species of mammals are being monitored for possible changes in populational characteristics. Community and population characteristics of birds are being examined by other researchers (see Section 2.8).

Population and community studies are inherently variable; therefore, only pronounced effects are detectable when monitoring these levels of organization. Laboratory research indicates that if operation of the ELF Communications System has any effect on small vertebrates, the effect will be small. Therefore, to complement the population and community approach, specific attributes of individuals are also studied. The purpose of examining individual characteristics is to gather a sufficiently large set of data to detect small differences in exposure comparisons. The individual aspects being examined are based on previous research and include reproductive, developmental, behavioral, and physiological characteristics of select species.

Those species selected for studies of population and most individual attributes are the deermouse, chipmunk, and tree swallow. The black-capped chickadee is also being examined but solely for physiological variables. The project uses five treatment sites in, or immediately adjacent to, the antenna ROW and four control sites with habitats similar to the treatment sites. Areas on the control sites (sham ROWs) have been cleared and are being treated the same as the antenna ROW.

Population and Community Studies. The purpose of this element is to monitor the small mammal community and select mammalian populations for possible effects from the operation of the ELF Communications System.

Live trapping was the primary method used to characterize the community and select populations of small mammals at study sites. In order to detect species that were not likely to be trapped, sign surveys and pitfall traps were also employed. Species diversity (an index of species evenness and richness) was determined and was used for intersite and interyear comparisons of the mammalian community. Densities of deermice and chipmunks (trappable population number) were used to monitor for possible effects at the population level.

The diversity of the small mammal community was not significantly different between Michigan study sites in either 1985 or 1986; however, the control site had a significantly higher diversity than the treatment site in 1987.

During 1985 and 1986, there was no significant difference between sites in the density of deermice; however, the density of deermice was significantly higher on the treatment site in 1987. The density of chipmunks during the period 1985-1987 was always significantly higher on the control site. The overall density of both deermice and chipmunks on all study sites has declined steadily since 1985. Researchers speculate that the decline may be due to a similar decline in rainfall over the period 1985-1987.

Researchers anticipate that interyear comparisons will be of little value in assessing possible ELF Communications System effects on population variables, although intersite comparisons within a year will be useful. They estimate that a 20 percent change in diversity and a 5 percent difference in population densities at study sites will be detectable using current protocols.

Embryonic Development. Prenatal developmental stages are especially sensitive to environmental perturbations. At present, there is conflicting evidence of direct EM effects on embryonic or fetal development. In addition, possible effects of the ELF system on parental behavior could also have an indirect effect on development. The purpose of this element is to determine the incidence of abnormalities in embryonic development in tree swallows at treatment and control sites and to test for possible effects of the ELF Communications System on the incidence of these abnormalities.

Embryos of tree swallows are collected at treatment and control study sites after four days of incubation. Embryos are dissected from the egg, preserved, and initially scored for normality. The preserved specimens are later cleared, stained, mounted whole on glass slides, and examined in detail for normality. The final determination of normality is carried out according to a "blind" procedure.

Chi-square analysis of the frequency of developmental abnormalities for embryos collected at treatment and control sites during the period 1985-1987 showed that the frequency of abnormalities was not uniform among the sites. One control site and one treatment site had a markedly higher frequency of developmental abnormalities than the other sites. Reanalysis of the data without the elevated site showed no significant difference between or among the treatment and control sites. The frequency of abnormalities for the pooled treatment and control sites (without the high-incidence sites) was 9.7 percent.

Researchers do not consider the high frequency of abnormalities at the one treatment site to be associated with the operation of the MTF. They will continue to monitor all sites, including those demonstrating a high incidence of abnormalities. In order to examine for the cause of the elevated abnormalities at some sites, ambient and nest temperatures at each site will be recorded during the egg-laying period.

Parental and Nestling Behavior, Fecundity, Growth and Maturation: Tree Swallows. The purpose of this element is to monitor important aspects of the reproductive and postnatal growth processes in the tree swallow. Variables are parental attentiveness to eggs and young, numbers of eggs per clutch, hatching success within clutches, rates of postnatal growth, development of hatchlings, and nestling mortality.

Studies are carried out in clearings where researchers have erected arrays of nest boxes. The boxes can be opened to permit inspection and weighing of the young. Active nests are checked daily or every other day to determine the dates that eggs are laid, the number of eggs, hatching dates, and overall hatching success. Monitoring of the nests for nestling growth and mortality then continues until all young fledge. Parental attentiveness to eggs is monitored using temperature probes, and their attentiveness to nestlings is monitored with video recording devices.

Clutch size (maximum number of eggs laid in a nest) has been used as an indicator of fecundity. In 1985 and 1986, treatment sites had significantly higher clutch sizes than control sites. However, in 1987 there was no significant difference between sites. Mean clutch size was 5.0 eggs per nest at both sites in 1987. Researchers are examining available food supply (insect biomass) as one possible factor influencing clutch size.

There were no significant differences between study sites in mean hatch frequency (percent of available eggs that hatch) in 1987. The mean hatch frequency for both sites was 4.2 young per nest. When 1985 through 1987 data were pooled, the likelihood of hatching was shown to be independent of both plot and year.

There was no significant difference between sites in the postnatal landmarks of mean number of days to eye opening or feather eruption during 1987. Results for eye opening were similar to those obtained in 1986; however, the mean number of days to feather eruption was significantly higher at the control site.

In order to examine growth rates, periodically measured values were fit to models. Data on body weight and on tarsus and ulna length taken in 1986 and 1987 best fit logistic models, while wing growth best fits an exponential model. Models were then used to produce values for an analysis of variance. In general, the rates for each growth variable showed significant differences among nests, but pooled rates did not differ between treatment and control sites.

Fledging success (percent of hatchlings that survive to fledge) was not significantly different between sites in 1987. The mean frequency of success for both sites was 3.1 fledgings per nest. Data pooled over the period 1985-1986 indicate that the likelihood of fledging is highly dependent on year and plot. When each year was tested separately, researchers found no significant departure from independence except in 1985, when hatchlings on the control sites were less likely to fledge than hatchlings on the treatment site. As noted in previous annual reports, 1985 was the first year that nest boxes were available on control sites, and a large number of inexperienced breeders occupied the nests.

Values obtained in these studies of fledging success, hatching success, and clutch size are similar to those reported in the literature for other studies of tree swallows.

Using all active nests on both treatment and control sites, data were pooled to create time units for analysis of overall nesting success. For example, one nest with five eggs observed for four days would represent 20 egg days and four nest days. Using this concept along with mortality, it was found that egg mortality, nest mortality, and incubation phase nest mortality were higher at treatment sites than at control sites in 1987. During this same period, there were no significant differences between sites for nestling mortality or nestling phase nest mortality. In 1986, control sites had significantly higher egg mortality, while treatment sites had a significantly higher mortality of nestlings. All other intersite comparisons using 1986 data were not significantly different.

In 1987, average egg temperature during incubation was used as an indicator of parental attentiveness to eggs. Although ambient air temperatures were significantly different between sites, there were no significant differences in average egg temperatures.

Results on parental attentiveness to nestlings in 1987 were not reported. During 1986, however, several variables were measured from video recordings of male and female activities at nests. Of the three variables examined, only data on visits per hour were considered statistically adequate for further study.

Parental and Nestling Behavior, Fecundity, Growth and Maturation: Deermice. The purpose of this element is to monitor important aspects of the reproductive and growth processes in deermice. Variables are maternal attentiveness to nestlings, number of young born per litter, proportion of young surviving until weaning, rate of postnatal growth, and rate of development of nestlings. (Note: the prenatal development of mammals is not being studied because reproductive females would have to be killed in order to examine the fetuses. The removal of the number of females required to meet statistical sufficiency for these studies would have adverse effects on the local population.)

Large, open enclosures are being used to restrict the movements of deermice during studies of behavior, fecundity, and growth. The deermice to be studied are captured in mixed deciduous forest near the enclosure sites. The animals are paired, and when the female is pregnant, she is transferred to the large enclosure to give birth and rear the young to weaning. The attentive behavior of the mother is monitored by using treadles attached to nest boxes and feeding stations.

In 1987 as in 1985 and 1986, age at eye opening and age at incisor eruption were not significantly different between sites. Growth studies to date have shown that growth curves of temporal change in the body mass of nestlings are different between litters. Therefore, growth rates have been estimated using linear regression analyses for growth of each individual and combined growth of all individuals in each litter. Analysis of this combined growth in 1987 showed no significant differences between sites.

No results have been reported on studies of maternal attentiveness.

Homing Studies. Animals are able to find food and escape predators more effectively in their home range or territory than in less familiar areas. Published information suggests that magnetic fields are one of several cues used in the orientation of some birds and mammals. Thus, any disturbance of the ability to return to, or use, a home range could decrease the probability of survival. The purpose of this element is to monitor the homing ability of tree swallows and deermice to assess for possible effects from the operation of the ELF Communications System. The variables being examined are the proportion of displaced individuals that return home and the amount of time taken to return home.

Adult birds from treatment and control sites were captured at nest boxes while brooding their young. Captured birds were banded, color-marked, and taken to release sites. (Release sites are located in open areas 30 km from the capture site.) The direction of the release points from the nest sites requires birds returning to their nests at treatment sites to cross both east-west antenna elements. Birds taken from control sites are displaced at angles and distances similar to those used for birds taken from the treatment sites, but do not cross or come near any of the antenna elements. Observers located near the nest boxes record the times at which the displaced birds return.

In 1986, there were no significant differences in return times between tree swallows captured at treatment or control sites; in 1987, however, birds took significantly longer to return to control sites than to treatment sites. In 1987, return times to treatment sites remained the same as in 1986, while return times to control sites were significantly longer than in 1986.

Chipmunks and deermice were captured on a trapping grid at treatment and control sites. Displacements took place during, or just prior to, the next activity period following capture; deermice were displaced at dusk and chipmunks in the morning. Individuals were displaced either to the south or west of the trapping grid, with each animal displaced 450 m from the trap at which it was captured. The displacements to the south were through relatively continuous forest, while displacements to the west required the returning animals to cross the antenna ROW or sham ROW. Once an animal was displaced, traps on the grid were checked morning and evening for the following five days.

There were no significant differences between sites in the frequency of return of displaced chipmunks or deermice in 1987. Previous studies have shown no significant differences between sites, genders, or directions of displacement.

Physiology: Peak Aerobic Metabolism. The purpose of this element is to determine the peak aerobic metabolism of chickadees and deermice during an annual period of severe stress. This variable provides a general index of an animal's health.

Black-capped chickadees and deermice are collected during the winter along the ELF Communications System's ROW and at a control site. Animals to be tested are held at an outdoor facility with food and water provided *ad libitum*. Tests for peak metabolism are performed in an ethanol-cooled chamber using a version of the helium-oxygen method. Test equipment is located at a laboratory in Crystal Falls, Michigan, while the holding facility is situated several miles south of the city. Once tested, animals are released at their collection site.

There were no significant differences between animals collected on treatment and control sites in their rate of oxygen consumption during 1987. Both weight-specific rates (mL of O_2 per gram hour) and whole-body rates (mL of O_2 per animal hour) of oxygen consumption were employed in the statistical analyses. Previous work also showed no significant differences between sites for weight-specific rates. Except for whole body rates of oxygen consumption in deermice, there were no significant differences between 1986 and 1987.

2.8 BIRD SPECIES AND COMMUNITIES

Many species of birds migrate from a nesting range to an overwintering area and back again. Successful migratory movement requires a mechanism that permits judgment of direction in order to arrive at the appropriate location. Many experiments have indicated that birds are sensitive to magnetic cues and use such cues, along with others, for orientation during migration. The magnetic environment in which a bird is raised may also be important in its development of orientation ability.

This element monitors for possible effects to resident birds, migratory birds that breed in, and birds that migrate through, the ELF Communications System area. The study concentrates on characteristics such as total species richness and abundance, abundance of common bird species, and abundances of birds within selected guilds. In order to properly qualify results, researchers are also examining habitat structure on treatment and control transects, possible "edge effects" caused by the antenna ROW, and the variability due to differences between observers.

A line transect method (variable width) was used to census bird community characteristics. Observers walked a designated transect and recorded information directly from sightings or indirectly from bird songs. The variables recorded were species, sex, behavior, perpendicular distance from transect, and distance along transect. In 1987 as in 1986, the characteristics of both the breeding and migrant bird communities were examined for each of five periods: spring migration (May), early breeding (June), late breeding (July), early fall migration (August) and late fall migration (September).

Study sites consist of 10 transects (five treatment and five control) in Wisconsin and 10 transects in Michigan. Treatment transects are parallel to and about 125 m from the edge of the antenna ROW. Control transects are variously oriented and generally at distances greater than 10 km from the antenna. Control transects do not have sham corridors located adjacent to them. Each transect is subdivided into eight 500-m segments.

<u>Vegetation Studies</u>. Treatment and control transects in Michigan and Wisconsin pass through a wide range of habitats, including deciduous and coniferous woods, bogs, meadows, marshes, and logged areas of differing ages. Because habitat structure has a pronounced influence on many aspects of bird communities, areas that differ in structure and species composition of vegetation will also differ in species and composition of birds. In order to determine if the ELF Communications System has affected or does affect the bird communities, it is necessary to account for habitat differences between treatment and control transects.

During 1987, a detailed habitat assessment of all Wisconsin transects was completed. Statistical analyses of vegetation on Wisconsin transects and qualitative assessments of habitat types in Michigan revealed several differences between treatment and control transects. In both states, treatment transects had more coniferous and lowland habitats than did control transects. Differences in relative abundance of deciduous and coniferous trees are likely to be the most meaningful in terms of the bird communities present.

Logging activities have affected the distribution of vegetation between treatment and control transects. In Michigan, clear cutting along several transect segments has resulted in a significant difference in the amount of early successional habitat, with more present on treatment transects than on control transects in 1987. Logging had less pronounced effects in Wisconsin, but did result in significant changes in some habitat variables. All segments affected by logging were omitted in the 1987 analyses of the bird communities.

Vegetation data will be used in future analyses of the possible influence of the ELF system on bird communities. Researchers will pair treatment and control transect segments on the basis of vegetational similarity. If a sufficient number of similar pairs can be determined, the pairs will be used to compare EM exposures using matched transect segments. This approach will allow researchers to detect smaller differences than if all data were combined into treatment and control categories.

Species Richness and Abundance of Individuals. No consistent pattern has emerged to suggest that the numbers of bird species or the numbers of individuals are larger on treatment transects than on control transects in either Michigan or Wisconsin. A few significant differences between treatment and control transects were found; however, these differences were not always found in other months or in subsequent years.

Habitat- or EM-related differences that exist between treatment and control transects may not influence all bird species in the same manner. If some species are more abundant on one type of transect and others on another, differences may be lost when all species and numbers are combined. Therefore, abundant species (mean number > 1 individual per 500-m transect segment), common species (present on at least five transect segments during a season), and a combination of both were also examined. If differences between treatment and control transect segments are primary factors (related to either habitat or EM exposure), then any given species should show similar patterns between years and seasons.

Few species were consistently and significantly more abundant on either treatment or control transects. This was true whether the comparison was made between seasons in one year or within seasons in several years. Researchers feel that differences, particularly in Michigan, are most likely due to habitat differences. Planned analyses to examine species-habitat relationships in more detail will help to resolve the influence of habitat on bird species distribution patterns.

Guild Analysis. Species that belong to the same guild share some biological characteristics. Thus, if the ELF Communications System influences the distribution of bird species, members of the same guild should be affected in the same manner.

Species were classified into guilds on the basis of migratory strategy, nest site preference, diet and foraging location, and preferred breeding habitat, with classifications based on published sources. Abundances of different guild types on treatment and control transect segments were compared for the primary breeding season (June) in 1985, 1986, and 1987. Few significant differences were found and most were not consistent between years. Differences were most consistent for habitat categories, indicating that habitat differences may be responsible for the guild distribution patterns.

Researchers will be better able to assess the relative influence of habitat versus ELF EM exposure on the distribution of guild members in Michigan after the MTF begins operation. Similarly, they will be able to examine guild patterns in Wisconsin more closely after segments are paired on the basis of habitat structure. Definitive statements cannot be made at this time regarding the cause of observed differences in bird guilds.

Edge Effect. The presence of a cleared ROW is a potential source of error when comparing treatment and control (no ROW) bird community characteristics. The variety and density of birds are known to be larger at the edges of plant communities. Vegetation changes associated with clearing of the antenna ROW are different from undisturbed habitats on control transects or in undisturbed areas adjacent to the antenna. Because clearing of sham corridors on control sites was not feasible, treatment transects were placed at a distance from the antenna ROW in an attempt to eliminate edge effect variability from the study design.

Analyses conducted on 1986 data were repeated using 1987 data. Except for one species, the indigo bunting, there were no significant differences between sides of the transects in the abundance of a given bird species. Thus, there appears to be little, if any, direct effect of the ROW and associated edge habitat on the study's results.

Observer Differences. Another potential source of error in censusing is the variability between observers in detecting and recording bird species and numbers. Several factors may contribute to the variations, including avian density, the observer's hearing acuity, and his or her ability to estimate distance to singing birds.

This potential source of error, checked annually since 1984, was again examined in 1987. In 1987, two observers (separated by 10 minutes) censused the same eight transect segments during the June breeding season, when almost all bird data are determined by sound. The same two observers censused the transect in 1987 as had done so in 1986; however, the order of their passage along the transect was reversed. In 1987, as in 1986, the first observer to pass along the transect recorded more birds than the second observer. Because the order of the observers' passage was the reverse of that used in 1986, researchers feel that the differences between observers were attributable to behavioral changes in singing and flushing of some birds caused by the passage of the first observer.

2.9 WETLAND FLORA

In addition to the upland flora described in Section 2.1, wetland ecosystems are also found in the ELF Communications System area. Wetlands play a valuable role in supporting diverse food chains, providing wildlife resources and, under some conditions, maintaining natural hydrologic systems. They are sensitive ecosystems that are easily modified by environmental perturbations.

Laboratory studies at ELF frequencies and intensities higher than those produced by the ELF Communications System have affected plants. It has been hypothesized that EM fields may affect biota by altering the transport of materials across their cell membranes. Therefore, variables important to the stability and functioning of wetland systems and which could be affected at the membrane level were monitored. The three types of variables examined were foliar nutrient content, stomatal resistance, and litter decomposition.

A common type of wetland found in the ELF Communications System area is the peatland. Three types of peatland plants (herbs, shrubs, and trees) were examined in 11 peatland sites near the WTF. The study sites were of four EM exposure types: antenna, ground, intermediate, and background. The antenna and ground sites (treatment sites) were located adjacent to the transmitting elements they describe. The background sites (control sites) were located more than six miles from the nearest transmitting elements and had EM intensities two orders of magnitude less than the treatment sites. Intermediate sites were located so as to have EM intensities between those of the treatment and control sites.

Each site was a rectangular plot oriented with the long side parallel to the nearest transmitter element. Each plot was subdivided into six subplots, with a shallow well located at the center of each subplot. Ambient environmental factors (e.g., ground water chemistry and temperature) were taken at the well; biological samples and other measurements were taken from areas immediately adjacent to the well.

The 1987 field season was the last for collection of data. During 1988, researchers will be chemically analyzing 1987 samples and preparing a final summary report.

Foliar cations. Foliar cations are important in plant physiology as active constituents of a number of biochemical reactions. Peatlands of the type found in the ELF Communications System area are relatively cation-poor environments, and the plants that exist there tend to conserve minerals by a variety of methods. As ELF EM fields may directly affect the transport of cations across cell membranes, four species of wetland plants were monitored for their foliar cation content.

Examination of previous data using nested analysis of variance techniques showed no significant differences among the exposure types. In order to more closely examine these results it was decided to increase the sample size. Instead of collecting 36 samples per species three times a year, as had been done previously, a single collection of 120 samples per species was taken once during 1987. Based on previous data, it is estimated that the 1987 sampling regime can detect a 20 percent difference in cation means at the 0.05 level of significance with an 80 percent probability. Each species was sampled at its physiological peak during the growing season.

Results of the analyses for the 1987 cation concentrations in foliar samples of leatherleaf, labrador tea, spruce, and *Smilacina* will be included in the final summary report.

Stomatal Resistance. The possibility of EM field effects to the transport of ions across biological membranes has been noted. The transport of ions, in turn, may indirectly affect water uptake through osmotic processes or may directly restrict stomatal opening. In some plants even a mild potassium deficiency can hamper stomatal opening. Therefore, the stomatal resistance was examined to determine the physiological status of wetland plants exposed to EM fields from the ELF Communications System.

Efforts previous to 1986 were devoted to development of an appropriate protocol for measuring stomatal opening using a steady-state diffusion porometer. After examining four species of plants as possible test organisms, researchers chose leatherleaf for further studies. Only newly expanded leaves were used, and measurements were taken only when the photosynthetically active radiation was above 400 microeinsteins per square meter per second. In 1986, 30 readings were taken in each of the 11 bogs under study; in 1987, the sample size was increased to 60 leaf measurements per bog. Researchers estimated that the increase in sample size would allow detection of 20 percent differences in means at the 0.05 level of significance with an 80 percent probability. In both 1986 and 1987, the sampling periods each lasted several days and were carried out in July and August.

Nested analyses of variance of 1987 data detected significant differences between exposure types for stomatal resistance of leatherleaf during July. No significant differences were detected for August measurements. In 1986, the difference detected was between the antenna and control exposure types, and there were no significant differences between other comparisons (e.g., antenna and intermediate sites or ground and intermediate sites). In 1987, significant differences were noted between intermediate and the other three exposure types, and between the ground and control exposure types. The significance of these findings is unclear, especially since they are not consistent with other analyses, interpretation of WTF operations, and the presence of significant differences between sites and plots within sites.

Decomposition. The decomposition of plant material, a major process in bog development and change, is accomplished by microorganisms. As a group, microorganisms have been shown to be affected by ELF EM fields at intensities greater than those produced by the ELF Communications System.

Senescent leaves from labrador tea plants were collected from a control bog site, air dried, and weighed. Leaves were randomly selected, enclosed in mesh litter bags, and then emplaced on the peat surface of each study bog. In 1986, the number of litter bags placed in each bog was increased from 48 to 96. The increased sample size should provide an 80 percent certainty of detecting a 20 percent difference among sample means at the 0.05 level of significance. In 1987, after one year, the litter bags were retrieved and the leaves were dried and reweighed to obtain percentage weight loss.

Statistical tests showed that leaf weight loss was significantly greater at the antenna sites than at the other exposure sites. Leaf weight loss at the antenna sites was 6 percent greater than at the control sites; this difference was less than the level that researchers could detect with an 80 percent certainty. In addition, antenna sites had the highest frequency of moss overgrowth on litter bags, indicating that leaf weight loss differences between sites may have been due to microenvironmental differences in bag placement.

2.10 AQUATIC BIOTA

In headwater streams such as exist in the ELF Communications System area, energy is supplied to the streams primarily by organic materials from periphyton and riparian vegetation. Macroinvertebrate consumers feed on the organic materials of the plants, making energy and invertebrate organic material available to higher trophic levels such as fish. The purpose of this study is to monitor a riverine ecosystem for possible effects from the long-term exposure to low level EM fields produced by the ELF Communications System in Michigan.

Two similar sections of the Ford River are used as matched study sites. One site is located adjacent to the antenna ROW (treatment); the other is located more than 10 km downstream (control). No major tributary occurs between the sites. At each site, ambient environmental factors are monitored and ecological experiments occupy adjacent stream segments. In order to determine the migration pattern of fish, four additional sites are located upstream of the control site.

2.10.1 Periphytic Algae

Periphyton are a community of microscopic plants and animals associated with the surfaces of submerged objects. Unlike the structural and functional aspects of organisms suspended in the water column, those of the periphyton community at a given location are governed by conditions at that point. Periphyton, therefore, are useful in assessing the effect of perturbations on streams because they show responses immediately at the source of the perturbation. Structural and functional aspects of the periphyton community are being monitored for possible changes due to the operation of the ELF Communications System.

Because of the periphytons' association with surfaces, quantitative determinations require the collection of periphyton from a known surface area such as that provided by an artificial substrate. Preliminary studies in the Ford River have found that the periphyton established on glass slides gave a good representation of the community found on natural substrates.

Because the periphyton are dominated by diatoms (plants), these organisms are emphasized in monitoring of the structural aspects of the community. However, functional aspects such as chlorophyll, biomass, photosynthesis, and respiration are determined for the entire community, i.e., diatoms, other plants, and animals.

Structural Aspects. The purpose of this element is to monitor select variables of the diatom community. Indices for species diversity, evenness, and abundance allow the detection of subtle shifts in the community's makeup, while total cell density and biovolume provides an indication of any overall change in the diatom community.

Glass slides emplaced at study sites for 28 days were used to identify and enumerate colonizing diatoms. The community that develops on emplaced slides most often consists of 50 to 70 species of diatoms. Because diatoms vary greatly in their size distribution, the number of individuals (total cell density) alone does not give an adequate picture of the community's makeup. Therefore, cell volume measurements for the dominant diatoms were also determined. Volume estimates were multiplied by the density of each species and summed to provide an estimate of the total biovolume for all cells present.

Since 1983, the seasonal pattern has continued to be high diversity and evenness during winter, with lower values in summer. Diatom cell density showed wintertime lows, with sustained high densities throughout the summer. For a given species, cell volume tends to be larger in the winter than in the summer. In 1987 as in the past, no statistically significant differences between study sites were found in species diversity, species evenness, or density of diatoms. There were no significant differences between sites in individual cell volumes, nor in total biovolume of 20 dominant species of diatoms. Similarly, there were no significant differences between sites in individual cell volumes or total biovolume when data were combined over the period 1983-1987 and examined using paired t-tests or three-way analysis of variance.

Functional Aspects. As indicated previously, numbers or types of diatoms alone do not provide a complete characterization of the periphyton community. The purpose of this element is to monitor such aspects as chlorophyll *a*, organic matter accumulation, photosynthesis, and respiration, which represent the functioning of the entire community.

Slides were emplaced in the Ford River for 14 days for determinations of accrual rates and 28 days for determinations of standing crop estimates of chlorophyll *a*, phaeophytin *a*, and organic matter biomass. Fluorometric methods were used for analyses of chlorophyll and phaeophytin. Organic matter biomass was determined using changes in ash-free dry weight per unit area.

Annual patterns for chlorophyll *a* standing crop and accrual were similar. The annual pattern was one of winter lows, with a peak value occurring in July or August. There was considerable year-to-year variability in both standing crop and accrual rates, due to the presence (or absence) of a secondary peak occurring from late March through June and the magnitude of the summer peak. The secondary peak occurs when spring conditions are dry, i.e., low stream flows and relatively warm temperatures. Although there were differences between sites in 1983 and 1984, no significant differences between sites were detected over the period 1985-1987 for standing crop or accrual rates.

Organic matter standing crop and accrual rates showed the same annual pattern as chlorophyll. In 1987 as in the past, there were no significant differences between sites.

In order to estimate community productivity and respiration, light/dark chambers with periphyton-covered substrates receiving recycled water were used to determine changes in dissolved oxygen concentrations. There were no significant differences between sites in net production, respiration, and gross production of the community over the period 1984-1987.

During 1987, a correlation matrix was generated using all the available data collected from each individual site over the previous five-year period. Although some water chemistry constituents appeared to influence the biological parameters at one site more than another, there was general agreement between sites in the influence of environmental factors, or water chemistry. The results of the correlation analyses agreed with previously reported analyses using multiple regression techniques.

2.10.2 Aquatic Insects

As part of the integrated studies of the aquatic ecosystem, insects are being monitored as representative of the primary and secondary consumer levels in the aquatic food chain. These studies examine the important functional insect groups, such as shredders, collectors, predators, and grazers. Both community and individual aspects of organization are being monitored. The community aspects are leaf litter processing; insect colonization patterns on leaf litter and artificial substrates; and the frequently used structural descriptors of community change such as species richness, individual abundance, and species diversity. The monitoring of individual aspects emphasizes changes in individual behavior such as alterations in movement patterns and feeding activity.

Feeding Activity of Grazers. The purpose of this element is to monitor the relationship between the producer and primary consumer trophic levels. This was accomplished by examining the periphyton community for effects from insect grazing.

The study approach uses streamside chambers to which are added tiles precolonized with periphyton and grazing insects. The chambers are subdivided so as to allow the introduction of different numbers of grazers (0 to 30) per experimental run. After a period of time the tiles are removed and the periphyton are analyzed for chlorophyll *a*, organic matter biomass, and diatom cell counts. Data collection techniques were developed during 1985, and at that time studies were conducted at one site per experimental run. In 1986 and 1987, experiments were conducted simultaneously at both treatment and control sites.

Grazing by the caddisfly, *Glossosoma nigrior*, caused a shift in dominance within the diatom community, with *Cocconeis* decreasing in abundance and *Achnanthes affinis* increasing in abundance as a function of the length of the grazing period. Such shifts in community structure occurred at both the treatment and control sites despite the absence of significant changes in such community parameters as chlorophyll *a* or organic matter (ash free dry weight) accumulation. The results were consistent enough to consider this pattern of response typical of the preoperational period. This pattern will be used as a basis of comparison for results obtained after the MTF becomes fully operational.

Preliminary studies performed during 1985 indicated that grazing by the caddisfly, *Glossosoma*, caused species and diversity shifts in the diatom community even though these shifts were not reflected in chlorophyll *a* or organic matter standing crop. In 1986 as in 1985, grazing caused no significant differences between sites for shifts in standing crop of chlorophyll *a* or organic matter. Species counts had not been completed at the time of reporting.

Benthic Insect Community. The purpose of this element is to monitor the major organisms constituting the primary and secondary consumer trophic levels, i.e., benthic insects, for changes in community structure and function.

Riverine substrates contained in sample baskets were emplaced at study sites for one-month periods at intervals throughout the season. Insects were collected from the substrates, identified, and counted. Numbers of individuals, diversity, richness, evenness, and percent numerical dominance for selected species were determined for each replicate. Total sample biomass and the biomass for functional feeding groups were determined. For those insects with high numerical abundance, mean dry weight per individual was also computed.

The structural community parameters (taxon diversity, evenness, and richness) have an annual pattern of high values during the summer and low values during the spring. In 1987 as in previous years, taxon diversity, richness, and evenness were significantly higher at the treatment site than at the control site. Sufficient samples were taken to have 95 percent confidence that the determined mean was within 40 percent of its estimated value at the 0.05 level of significance. Differences between mean values for diversity, evenness, and richness values at the study sites ranged between 13 and 28 percent. Chironomids were present in high numbers in the Ford River, and their abundance has a marked effect on these structural community parameters.

Over the five-year period 1983-1987, distinct seasonal patterns were found for total biomass of insects. The annual pattern of insect biomass showed both summer (July-August) and spring (February-April) peaks, with troughs over the October-December period. Insect biomass was significantly correlated with water temperature and diatom density until 1986-1987.

Unusually mild weather during the winter of 1986-1987 was suspected as being the principal factor for the lack of continuity in the relationships. Graphic presentations indicate higher insect biomass at the treatment site than at the control site. The biomass of functional feeding groups including collector-gatherers, collector-filter feeders, and predators were highly correlated with diatom densities until 1986-1987. Their biomass had the most influence on total insect biomass. Shredders had less influence on total biomass, and were not significantly correlated to diatom densities.

Future plans include examining total biomass values during transition periods to compare preoperational and operational periods using regression techniques. Researchers will also examine abundance for differences between sites using a "before and after at control and impact" statistical design. In this design, the differences between the means are plotted rather than the means themselves. If there is no change, one would expect a horizontal line. Changes would be reflected in the direction, amplitude, and frequency of change.

Leaf Litter Processing. In headwater streams such as exist in the ELF Communications System area, only a portion of the energy supply to the ecosystem is provided by aquatic plants and algae. The maintenance of community structure is largely dependent on the input of organic materials (i.e., leaves) from riparian vegetation. Macroinvertebrate consumers, mainly insects, process the leaves, making consumer biomass available to higher trophic levels (predators).

Leaf decomposition and insect colonization patterns using "leaf pack" bioassay techniques are being used to monitor for possible EM effects to this energy pathway. Species diversity, evenness, and richness are used to characterize colonization patterns. Leaf processing rates (mass loss) are used to quantify the feeding activities of the colonizing organisms.

Processing rates of both fresh and dried green leaves were examined by emplacement during the fall of 1986 and the summer of 1987. Leaf samples were retrieved at regular intervals after emplacement. At the time of reporting, data on leaf processing were available, but data on leaf pack colonizing insects were not ready for examination. Analysis of variance showed some differences between sites on several retrieval dates in leaf mass remaining for the samples emplaced during the fall of 1986. No differences between sites were noted for samples emplaced during the summer of 1987. Some data taken previous to 1986 indicate that, at times, leaves were processed significantly faster at the treatment site than at the control site. The researchers feel that the reported site differences may be due to scouring at the treatment site and sand deposition at the control site. An analysis of covariance for the processing rates of fresh and abscissed leaves will be presented in the 1988 Annual Report.

During 1986, there were significant differences between sites for all structural community parameters including taxon diversity, evenness, and richness. The percent dominance of chironomids generally increased over the period of emplacement of leaf packs. The steady decrease in taxon diversity and evenness during the latter part of emplacement was attributed to the steady increase in numerical dominance by chironomids.

Functional community parameters for colonizing organisms include total insect biomass (adjusted to leaf biomass), biomass of functional groups, and the mean (dry) weight per individual for representative species within each functional group. Analysis of variance showed significant differences between sites for total biomass values on two of the four dates examined. Analyzing the 1986 biomass of functional groups showed that in all cases shredder biomass was significantly greater at the treatment site than at the control site, with no significant differences between sites for collector/gatherers and predators. Three insect species were examined for changes in mean dry weight per individual. Of the three species, a collector/gatherer and a predator showed increases in weight on both fresh and senescent leaves over the period that the leaf pack was left in the stream. The third species examined failed to show a consistent pattern.

<u>Insect Movement Patterns</u>. Other riverine studies have used the behavioral drift of aquatic invertebrates as a measure of community response to stressed conditions. In order to monitor for possible effects from the operation of the ELF Communications System, mark and recapture techniques are being used to discern the movement patterns of displaced dragonfly naiads.

Naiads of *Ophiogomphus colubrinus* (dragonfly) traveled downstream for short distances after their release. Chi-square tests showed that the marked naiads moved significantly farther downstream at the control site than at the treatment site. This downstream distribution pattern was found consistently over the period 1985-1987, and was attributed to the higher mean stream velocity at the control site. Lateral movements of the naiads were related to flow patterns at each of the sites. Recapture success indicates a high probability in detecting possible alterations in the movement pattern of the dragonfly due to the operation of the ELF system.

2.10.3 Fish

An extraordinary ability to perceive EM fields has been reported for some species of fish. It is believed that fish use this perceptive ability to orient themselves and to detect prey. Fish also represent the tertiary consumers in the aquatic food chain; therefore, the characteristics of the mobile fish community, and the migratory behavior of trout in particular, are being monitored for possible effects from the operation of the ELF Communications System in Michigan. In addition, the parasitic populations of two "nonmotile" species of fish are being monitored. Changes in the parasitic populations are being used as an indicator of possible stress to the fish.

Mobile Fish Community. Fyke nets and weirs have been deployed across the width of the Ford River drainage at five sites in or near the ELF Communications System. All fish are collected, and both community characteristics and movement through the area are recorded. Community characteristics recorded are species composition, species abundance, and biomass.

In 1987, the number of species collected was higher at the upstream treatment site than at the control site--the reverse of previous years. Sixteen species were collected at the treatment site and 14 at the control site. The difference between sites in the number of species was attributed to the presence of a few rarely found species. There were no significant differences between sites in the diversity of the mobile fish community over the period 1983-1987, and the diversity at each site was generally similar from year to year.

The fish community was dominated by the same five species at both sites. In 1987 as in 1986, the numbers of common shiners and creek chubs made up the two highest percentages of the catch; the other common species were the burbot, brook trout, and white sucker. Percent catch by biomass showed the same five species as most abundant; however, brook trout and burbot were the dominant species. Percent catch by biomass was more variable than percent catch by numbers.

Two to three times as many nonsalmonid fish were marked in 1987 than in any previous year. Although the recapture frequency in 1987 (about 11 percent) was similar to that of previous years, the movement of fish through the ELF Communications System area was slower than in previous years. Site-to-site movements continued to decline for creek chubs. Differences in fish movement were attributed to a significantly lower flow rate for the Ford River in 1986 and 1987 than that experienced during 1984 and 1985.

In order to assess for possible direct effects of the ELF Communications System on the mobile fish community, analyses were initiated in 1986 to examine the growth and condition of captured fish. The common shiner, creek chub, white sucker, and northern pike were selected as indicator species for the community. Growth analysis indicated that common shiners and creek chubs showed better than average growth when compared to literature values. White suckers and northern pike displayed poor growth when compared to literature values. In addition, the condition of common shiners, creek chubs, and white suckers was examined using relative weight condition factors. Condition factors for creek chubs and white suckers were 4 to 20 percent below their respective species mean, while condition factors for the common shiner were above the species mean for all years examined. A statistical analysis of differences between years is being performed.

Brook Trout Movement. Magnetic cues have been shown to be used by fish in their migratory movements. In a thermally unstable stream such as the Ford River, it is particularly important to determine any possible disorientation of fish in their search for an optimal temperature regime.

The general pattern of trout migration has been an upstream movement in the spring to early summer, with a varied intensity and timing of peak movement from year to year. Peak catch occurred in June in both 1984 and 1987, and in July in 1985, with no apparent peak during 1986. Trout migrate through the ELF Communications System area (control and treatment sites) to the confluence of the Ford River and Two Mile Creek. Virtually all trout migrate up Two Mile Creek; optimal growth temperatures appear to be responsible for the movement of trout up Two Mile Creek instead of their continuing up the Ford River. No downstream movement from Two Mile Creek was found for sampling periods lasting through November. Factors affecting timing of peak catches and distribution pattern appear to be water temperature, stream velocity, and population size.

Brook trout were found to move between the antenna ROW and Two Mile Creek at average rates of 1.4 km/day in 1984, 1.6 km/day in 1985, and 1.8 km/day in 1987. No trout were recaptured in 1986, and only one was recaptured in 1987. The movement rates were more variable between other possible station arrangements. Overall mean rates of 1.2 to 5.0 km/day have been determined for movement of trout through the ELF Communications System area.

Age and growth analysis indicated that the brook trout in the Ford River exhibit average or better growth than that reported in the literature. As for the nonsalmonid species, brook trout were examined using relative weight condition factors. A standard weight formula was calculated from 45 populations reported in the literature. Brook trout from the Ford River were in average to below average condition when compared to the calculated literature average. These condition factors declined from 1983 to 1986 and improved in 1987. Statistical analysis of this data is in progress, and will be reported in the 1988 Annual Report.

Fish Parasites. If stressed, fish may become more susceptible to parasites as their physiological condition deteriorates. The parasitic fauna of two "nonmotile" fish species were being monitored at treatment and control sites as indicators of possible stress induced by operation of the ELF Communications System. This element was suspended in 1987. However, progress was made in finalizing the data from previous years for publication and for comparison with studies to be conducted after the ELF system becomes fully operational.

The infestation frequency and prevalence of the parasitic fauna were extremely variable. Because of the variability of the data, age/cohort analyses will be performed for the three most common parasites associated with each fish species.

3. ENGINEERING SUPPORT

Accurate data are needed to evaluate cause-and-effect relationships between EM exposure and biological/ecological end points. As part of the Ecological Monitoring Program, IITRI assists university investigators by providing annual EM measurements and other EM engineering support. EM engineering support includes such activities as analysis of EM aspects of research protocols; design, fabrication, and installation of special EM exposure equipment; and review of EM aspects of investigator reports in the context of environmental protection or risk.

This section summarizes both the measurement of EM field exposures at study sites and those engineering activities carried out in support of the program during 1987. A more extensive presentation appears elsewhere.[16]

3.1 TRANSMITTER OPERATIONS

In order to evaluate the effect of the operation of the ELF Communications System, investigators may need to examine the intensity as well as other characteristics of EM exposure as covariates in their statistical analyses. This section summarizes the operational characteristics, particularly duration, of the transmitting facilities in both Wisconsin and Michigan. EM intensities experienced at study sites are discussed in Section 3.2.

Data on antenna operations have been provided to IITRI by the Navy on a minute-by-minute basis, and included all changes in operational frequency, modulation, power, and phasing for each antenna element. This information has been provided to investigators as graphical and tabular summaries and, when requested, in detailed tabular form.

The WTF has been fully operational since the last quarter of 1985. Its operating history during 1986 and prior years has been reported previously.[11-13] During 1987, the WTF transmitted modulated 76 Hz signals for approximately 8300 hours (about 95 percent of the total available time; the approximately 5 percent of nonoperational time represented scheduled weekly maintenance periods and unscheduled repairs). The north-south and east-west antenna elements were operated simultaneously at 300 amperes for more than 99 percent of the transmission time.

The MTF was intermittently operated at low antenna currents for the first time during 1986. In 1987, the MTF was operated at 3 to 6 amperes during April and May, and at 15 amperes during the months of June through November. In 1987 the two east-west antenna elements were operated in parallel, rather than independently as in 1986. The MTF was operated for about 400 hours (less than 5 percent of the total available time), evenly split between the north-south and both east-west antenna elements.

3.2 EM FIELD MEASUREMENTS

EM measurements were made at all active Wisconsin and Michigan study sites during the months of August through October 1987. These measurements have been performed annually since 1983 to document major changes and to characterize the temporal variability of the EM exposure. The number of measurement points in Michigan was increased in 1987 to better characterize the spatial variation experienced across large treatment sites.

The protocol for EM measurements at Wisconsin study sites was the same in 1987 as in 1986. EM measurements were limited to 76 Hz fields. Because the WTF is fully operational and being used for submarine communications, 76 Hz EM field intensities are much larger than, and mask, ambient 60 Hz field intensities. Only 76 Hz measurements have been made at Wisconsin study sites since 1984.

Except for one wetland study site, no unexpected changes in EM field intensities were measured in Wisconsin. At this site, a 50 percent reduction in electric field intensity, but no change in magnetic flux density, was measured. The nature of the observed EM intensities indicates an alteration in the soil conductivity at the site.

The EM measurement protocol at Michigan study sites during 1987, as in 1986, included the characterization of both 76 Hz and 60 Hz EM fields. As a result of equipment reconfigurations at the MTF, some data processing and analytical protocols were altered. The principal change at the MTF in 1987 was the simultaneous operation of the two east-west antenna elements. These antenna elements were operated separately during the 1986 test period. The changes cause no difficulty in characterizing EM exposures, but represent a modification of EM conditions that may need to be accounted for in the statistical treatment and interpretation of biological and ecological results. Other 1987 operational reconfigurations at the MTF were higher antenna currents and changes in the antenna-to-power amplifier configurations of the nonactive antenna element. These differences also influenced both 76 Hz and 60 Hz EM fields at study sites.

3.3 OTHER SUPPORT ACTIVITIES

The 60 Hz fields at the laboratory holding facility for small vertebrates were greater than site selection criteria dictate, and, as a result, the holding facility was relocated. The 60 Hz EM fields at the new facility were compatible with all site selection criteria. A similar problem was identified for maple leaf and pine needle collection sites, and new foliar collection sites are being considered by the principal investigator.

It has been recognized that the measurement of EM fields once a year may not adequately account for time variations that occur in the intensities of electric and magnetic fields. Both 76 Hz and 60 Hz electric fields in earth are influenced by changes in soil conductivity, and 60 Hz magnetic fields change in response to alterations in the current flowing in transmission and distribution lines. Microprocessor-controlled data loggers were designed, fabricated, and installed at the slime mold study sites in Wisconsin. The data loggers recorded electrical data in culture chambers hourly from 9 July through 19 October. The experience gained with the data loggers will be used at Michigan study sites in 1988.

IITRI also performed EM measurements and analyses of voltages induced onto ambient monitoring equipment used by the upland flora and soil microflora studies. EM measurements were made for safety and interference considerations as well as to develop plans for protecting the equipment from lightning damage. The design of interference mitigation and lightning protection was begun during 1987. It is anticipated that the design will be implemented during 1988.

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APPENDIX A

ECOLOGICAL MONITORING PROGRAM: LIST OF PUBLICATIONS/PRESENTATIONS, 1982-1987

ECOLOGICAL MONITORING PROGRAM: LIST OF PUBLICATIONS/PRESENTATIONS, 1982-1987

Upland Flora (Michigan Technological University)

1. Fuller, L. G.; Reed, D. D. A model of seasonal diameter growth development for four northern hardware species. (In preparation.)

2. Richter, D. L. Shifts in mycorrhizal fungus populations on red pine seedlings following outplanting on clear hardwood sites in Michigan's Upper Peninsula. (Dissertation, in preparation.)

3. Jones, E. A.; Reed, D. D.; Cattelino, P. J.; Mroz, G. D. Seasonal height growth in young red pine plantations. Forest Science. (Submitted for publication.)

4. Holmes, J. J.; Reed, D. D. Competition indices for mixed species northern hardwoods. Forest Science. (Submitted for publication.)

5. Reed, D. D.; Jones, E. A.; Liechty, H. O.; Mroz, G. D.; Gale, M. R.; Jurgensen, M. F. Microsite factors influencing northern hardwood productivity in upper Michigan. Canadian Journal of Forest Research. (Submitted for publication.)

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10. Cattelino, P. J.; Brooks, R. H.; Jurgensen, M. F.; Mroz, G. D. Determination of coarse fragment volume in northern hardwood forest soils. Presented at the American Society of Agronomy meeting, 1988.

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16. Fuller, L. G.; Holmes, M. J.; Reed, D. D. Development and testing of a seasonal diameter growth model for four northern hardwood species. Presented at the International Union of Forest Research Organizations--Forest Growth and Prediction Conference, Minneapolis, Minnesota, August 1987.

17. Fuller, L. G. Modeling northern hardwood diameter growth using weekly climatic factors in northern Michigan. M.S. thesis, School of Forestry and Wood Products, Michigan Technological University, Houghton, Michigan, 1986.

18. Becker, C. A. The effects of plant moisture stress on red pine (*Pinus resinosa*) seedling growth and establishment. M.S. thesis, School of Forestry and Wood Products, Michigan Technological University, Houghton, Michigan, 1986.

19. Becker, C. A.; Mroz, G. D.; Fuller, L. G. Effects of moisture stress on red pine (*Pinus resinosa* Ait.) seedling root and mycorrhizae development. Presented at the Conference on Roots in Forest Soils: Biology and Symbiosis, Victoria, British Columbia, 1986.

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<u>Slime Mold (University of Wisconsin-Parkside)</u>

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<u>Soil Amoebae (Michigan State University)</u>

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2. Blake, J.; Hanowski, J. M.; Niemi, G. J. Annual variation in abundance and species composition of breeding birds in northern Wisconsin and Michigan. (In preparation.)

3. Collins, P. T.; Niemi, G. J.; Blake, J.; Hanowski, J. M. Lateral distance distribution patterns for northern forest birds. (In preparation.)

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Aquatic Biota--Periphyton (Michigan State University)

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2. Cornelius, D. M.; Burton, T. M. Studies of *Ophiogomphus colubrinus* in the Ford River in Michigan. Presented to the American Benthological Society, Orono, Maine, 1987.

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APPENDIX B

ECOLOGICAL MONITORING PROGRAM: FY 1987 RESOURCES

ECOLOGICAL MONITORING PROGRAM: FY 1987 RESOURCES

The Navy has been committed to a program of long-term ecological monitoring since the ELF Communications System site selection process was initiated. The Ecological Monitoring Program is identified separately from other environmental protection work for future year budgeting purposes; therefore, continuity of the Program is anticipated, presuming continued Congressional approval and funding of the ELF Communications System.

During 1987, monitoring studies were conducted under 10 subcontracting agreements between IITRI and study teams from five universities (see Table B-1). IITRI provides engineering support and overall program management. Each study team is headed by a principal investigator with academic training to the doctoral level. Most of the staff also have advanced degrees, with expertise and publications in the areas under study. During 1987, the Ecological Monitoring Program consisted of more than 100 people expending a total of more than 100,000 staff hours.

TABLE B-1. ECOLOGICAL MONITORING PROGRAM: FY 1987

Study	Subcontractor	Principal Investigator(s) and (Total Staff)	Professional Staff Hours 1987
Upland Flora	Department of Forestry Michigan Technological University	M. F. Jurgensen, Ph.D. (18 persons)	14,598
Soil Microflora	Department of Forestry Michigan Technological University	J. N. Bruhn, Ph.D. (7 persons)	4,812
Slime Mold	Biomedical Research Institute University of Wisconsin (Parkside)	E. M. Goodman, Ph.D. (6 persons)	5,370
Soil Amoebae	Department of Zoology Michigan State University	R. N. Band, Ph.D. (6 persons)	7,372
Soil Arthropods and Earthworms	Department of Zoology Michigan State University	R. J. Snider, Ph.D. R. M. Snider, Ph.D. (15 persons)	14,519
Native Bees	Department of Entomology Michigan State University	K. Strickler, Ph.D. M. Scriber, Ph.D. (10 persons)	7,149
Small Mammals and Nesting Birds	Department of Zoology Michigan State University	D. L. Beaver, Ph.D. (10 persons)	14,221
Bird Species and Communities	Natural Resources Institute University of Minnesota (Duluth)	G. J. Niemi, Ph.D. J. M. Hanowski (9 persons)	7,134
Wetland Flora	Department of Botany University of Wisconsin (Milwaukee)	F. Stearns, Ph.D. (5 persons)	6,541
Aquatic Biota	Departments of Zoology, Entomology, Fisheries and Wildlife Michigan State University	T. M. Burton, Ph.D. R. J. Stout, Ph.D. W. W. Taylor, Ph.D. (15 persons)	15,622
Program Integration and Engineering Support	Electromagnetics and Electronics Department IIT Research Institute	J. E. Zapotosky, Ph.D. (4 persons)	4,160
		TOTAL	101,498