

# ELF Communications System Ecological Monitoring Program: A Summary Report for 1982-1992

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#### FOREWORD

This report has been prepared by IIT Research Institute (IITRI) on behalf of the Space and Naval Warfare Systems Command (SPAWAR) to summarize the results of SPAWAR's program monitoring for possible electromagnetic effects to biota from operation of the U.S. Navy's Extremely Low Frequency (ELF) Communications System.

Monitoring studies have been performed by research teams from Michigan Technological University, Michigan State University, the University of Minnesota-Duluth, the University of Wisconsin-Milwaukee, and the University of Wisconsin-Parkside under subcontract agreements with IITRI. SPAWAR funded these studies through contracts N00039-81-C-0357, N00039-84-C-0070, N00039-88-C-0065, and N00039-93-C-0001 to IITRI. IIT Research Institute, a not-for-profit organization, managed the program and provided engineering support to university research teams.

The Michigan Department of Natural Resources has supported the monitoring program by providing information for site selection and permits for conducting research on state lands. It also has reviewed program results and progress. The U.S. Forest Service provided similar services when studies were conducted in the Chequamegon National Forest, and it continues to support the Navy's oversight of the program.

The information in this report was largely derived from other, more detailed, technical reports on ecological investigations<sup>1-11</sup> and electromagnetic measurements<sup>12-21</sup> made since 1982 at the Naval Radio Transmitting Facility at Republic, Michigan. Overall results and conclusions for studies performed near the Naval Radio Transmitting Facility at Clam Lake, Wisconsin, can be found in other documents.<sup>22-24</sup> All reports are available from the National Technical Information Service. Study designs and preliminary findings have also been described in presentations to scientific societies and articles in peer-reviewed journals (Appendix A).

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Respectfully submitted,

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Raiph D. Carlson, Director ELF Electromagnetic Compatibility Assurance

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#### GLOSSARY AND ACRONYMS

AIBS American Institute of Biological Sciences.

- ANCOVA analysis of covariance, a statistical method for determining whether the functional relationships described by two or more regression equations are the same; it is used when treatments are compared in the presence of accompanying variables that cannot be eliminated or regulated.
- ANOVA analysis of variance, a statistical technique for partitioning the total variability affecting a set of observations between the possible and statistically independent causes of the variability.
- BACI before and after, control and impact, a statistical technique that compares the mean of the "before" differences between the control and treatment (impact) sites to the mean of the site differences "after" the treatment (i.e., full transmitter operation).
- biomass a quantitative estimate of the total mass of living organisms comprising all or part of a specified unit such as a population.
- CATMOD categorical data modeling, a statistical procedure used to compare proportions.
- chi-square a statistical method for testing the degree to which observed frequencies or values differ from frequencies or values expected from the specific hypothesis being tested.
- chlorophyll a photosynthetic pigment reflecting green light; in the aquatic studies it is used as a measure of plant productivity or standing crop.
- clone an assemblage of genetically identical organisms derived by a sexual or vegetative multiplication from a single sexually derived individual.
- clutch the number of eggs laid at any one time.
- control a location where parallel observations or experiments are carried out; they provide a site standard against which a result can be compared. As used in this report, control site is a location remote from the ELF Communications System, having 76 or 44 Hz electromagnetic intensities at least one order of magnitude less than those at its matched treatment site.

dendrometer a method of measuring plant growth by examining increases in trunk or stem diameter. band

diatoms a group of algae that often dominate aquatic communities in unpolluted rivers.

diel pertaining to a 24-hour period.

edge effect the impact exerted by adjoining communities on the population structure within the marginal zone, which often contains a greater number of species and higher population density of some species than either adjoining community.

ELF	extremely low frequency, in general use, refers to frequencies ranging from 0 to 300 Hz; as used in this report, it refers to operating frequencies of the Navy's ELF Communications System (i.e., 76 $\pm$ 4 Hz).
EM	electromagnetic.
EW	refers to antennal elements oriented in an east/west direction.
fecundity	the potential reproductive capacity of an organism or population, measured by the number of gametes.
genetic diversity	a measure of the genotypic disparity within a population, the different forms of a gene occupying the same locus or loci.
GLM	general linear modeling, a statistical procedure used when values are weighted by their rank in a sequence.
guild	a group of species having similar ecological resource requirements and foraging strategies, and therefore having similar roles in the community.
herbaceous plants	plants that have stems not secondarily thickened or lignified and that die down annually.
litter	recently fallen plant material that is only partially decomposed and in which the organs of the plant are still discernible, forming the surface layer on some soils.
MDD	minimum dectable difference, magnitude of the smallest change that can be perceived for a given sample size and parameter variance.
MSU	Michigan State University.
MTU	Michigan Technological University.
mycelium	the mass of filamentous hyphae that comprises the vegetative stage of many fungi.
mycorrhiza	the close physical association between a fungus and the root system of a plant.
NAS	National Academy of Sciences.
NRTF	Naval Radio Transmitting Facility.
NS	refers to antennal elements oriented in a north/south direction.
periphyton	a community of plants, animals, and associated detritus adhering to and forming a surface coating on submerged objects.
phaeophytin	breakdown product of chlorophyll, sometimes used in conjunction with chlorophyll as a crude estimate of algal health.
phenology	the study of temporal aspects of recurrent natural phenomena, e.g., flowering.
pupa	the life cycle stage of an insect during which the larval form is reorganized to produce the final, adult form; commonly an inactive stage enclosed within a hard shell.

provenance	place of origin.
regression analysis	in statistics, the estimation of the relationship between one variable and one or more other variables, by expressing one in terms of a linear or more complex function of the others.
RIA	randomized intervention analysis, a nonparametric statistical procedure in which the test statistic is compared to a random distribution of the data set, equivalent to BACI.
ROW	right-of-way, a cleared corridor for the location of antenna elements.
sham ROW	a cleared corridor located on control sites that is used to duplicate possible effects from the antenna ROW on study variables.
significance	the probability that experimental results have not occurred by chance alone.
species diversity	a measure of the number of species and their relative abundance in a community.
species eveness	the apportionment of individuals among those species found in a given community.
species richness	the absolute number of species in an assemblage or community.
t-test	a statistical method used for determining the significance of the difference between two means when the samples are small and drawn from a normally distributed population.
treatment site	a location where primary observations or experiments are carried out and where biota are site-exposed to 76 or 44 Hz EM intensities at least one order of magnitude greater than those at its matched control site.
UMD	University of Minnesota-Duluth.

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# ELF COMMUNICATIONS SYSTEM ECOLOGICAL MONITORING PROGRAM: A SUMMARY REPORT FOR 1982-1992

# 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 Department of the Nary has been interested in using ELF EM signals to communicate with submerged submarines since the late 1950s. Subsequent to successfully testing this concept, an experimental facility was built near Clam Lake, Wisconsin, in 1968-1969. This facility was then intermittently operated at less than full power to conduct transmission tests, to develop methods for mitigating EM interference, and to make engineering evaluations of System hardware.

In 1981, President Reagan directed the Navy to proceed with its program for constructing a fully functional ELF Communications System. The Wisconsin test facility was upgraded and reached a full operational capability in 1985. Construction of a second transmitter near Republic, Michigan, was initiated in 1984. Intermittent operation of the Michigan transmitter began in 1986, and it became a fully operation facility in 1989. As a Naval Radio Transmitting Facility (NRTF) the Wisconsin facility is now designated as the NRTF-Clam Lake, and the Michigan facility as the NRTF-Republic (Figure 1).

#### 1.3 Ecological Monitoring Program

Research on possible EM effects to biota from exposure to EM fields produced by an ELF Communications System began in 1969. Although some ecological and wildlife studies were performed at the Wisconsin test facility, most of the early, ELF System-related research was simulated in laboratories. 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. Upon their separate reviews, the Navy and NAS each concluded that adverse effects to biota from the operation of the ELF Communications System were unlikely.

Despite the unlikelihood of EM effects, the Navy and NAS recommended *in situ* monitoring of biota if an ELF Communications System was built. After their review of bioelectromagnetic research (1977-1984), the American Institute of Biological Sciences (AIBS) accordingly advised verification of low-risk predictions through continued ecological research. The AIBS noted that the results reported in the literature could not be directly extrapolated to the greater complexity found in ecological systems.

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FIGURE 1. ELF COMMUNICATIONS FACILITIES IN WISCONSIN AND MICHIGAN.

The Navy had outlined a plan for conducting a monitoring program at those sites approved for operation of the ELF Communications System in its final (1977) environmental impact statement. The plan was based on previous research and input from state agencies, U.S. Forest Service, NAS, and comments on the Navy's draft environmental impact statement. Following approval to complete the ELF System, technical elements of the Navy's plan were incorporated into a statement of work and issued in early 1982 as part of a request for proposals.

Approximately 120 proposals were received and reviewed for scientific merit by panels of scientists independent of the Navy. Most research teams selected for the monitoring program began their work by m. 1982. Early efforts focused on the selection of study sites, validation of assumptions made in the proposals, and characterization of critical study aspects. As these tasks were accomplished in 1984 and 1985, increased emphasis was then placed on accumulating a data base for statistical analysis.

Data collection for studies located near the NRTF-Clam Lake, Wisconsin, was completed, as scheduled, during 1989. Investigators concluded that there were no EM effects to bird species and communities,<sup>22</sup> slime mold physiology,<sup>23</sup> or wetland flora<sup>24</sup> from intermittent or full operation of the transmitter in Wisconsin.

Wildlife surveys near the NRTF-Clam Lake were also performed by the U.S. Forest Service. Although not an integral part of the Ecological Monitoring Program, the results of these surveys are noted

here since they are relevant to addressing EM effect issues. Annual surveys of ruffed grouse, eagle, and deer populations were initiated in 1974, 1975, and 1982, respectively. No effects on these populations from the operation of the ELF Communications System were detected, and the studies were concluded after the 1986 surveys (protocols and data are summarized in Reference 25).

Data collection for studies located near the NRTF-Republic, Michigan, progressed during 1992 and continued during 1993. Final results and conclusions for Michigan studies are expected after all data have been analyzed and peer-reviewed in 1994 and 1995. This report summarizes EM exposures and research performed at study sites near the NRTF-Republic.

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#### 2. <u>ELF COMMUNICATIONS SYSTEM</u>

# 2.1 System Description

Each facility includes a transmitter compound, overhead wire antennas, and buried ground terminals. An aerial photograph of the NRTF-Republic transmitter compound is shown in Figure 2. Both the antenna and grounding elements are located in cleared rights-of-way (ROWs). A photograph of a typical pole-mounted antenna wire, which resembles an ordinary power distribution line, is shown in Figure 3. The NRTF-Republic has a 28-mile-long north/south (NS) antenna and an east/west (EW) antenna comprised of two parallel 14-mile-long elements. Under full operational conditions, the NRTF-Republic antennas each carry 150 A of current. The end of each antenna or element is terminated with one to three miles of buried horizontal ground wire and typically one or more arrays of well-type electrodes extending to depths of 100 to 300 feet.

The ELF Communications System operates in a frequency band centered at 76 hertz (Hz). The information to be transmitted is converted to binary code (a sequence of 0s and 1s) and is used to modulate the frequency of the ELF antenna current (Figure 4). Thus, when a "zero" is transmitted, the frequency of the antenna current is 72 Hz ( $f_{LO}$ ); for a "one," the frequency is 80 Hz ( $f_{HI}$ ). The power spectral peak for this modulation scheme occurs at the center frequency of 76 Hz (Figure 4). The transmitters can operate at other center and shift frequencies, but have not done so operationally.

#### 2.2 EM Fields

An operating ELF antenna generates three types of electromagnetic fields: a magnetic field, an electric field in air, and an electric field in the earth. The r agnetic field is produced by the current on the antenna wire, while the air electric field results from the difference in potential or voltage between the antenna wire and earth surface. The antenna's magnetic field, in turn, induces an electric field in the earth. A second and more localized source of earth electric field is the current conducted from the buried antenna ground terminals.

The orientations of the generated EM fields with respect to the antenna and ground wires are diagrammed in Figure 5. The magnetic field and air electric field near an antenna wire are shown separately for clarity. The magnitudes of the fields are quantified in Equations 1 through 4. These equations are valid for points whose distances to the antenna or ground wire are small with respect to the wire's length, which includes all of the monitoring program's treatment sites. EM field magnitudes at control sites require more complex equations for prediction, but are dependent on essentially the same parameters.



FIGURE 2. TRANSMITTING FACILITY AT THE NRTF-REPUBLIC.



FIGURE 3. OVERHEAD ANTENNA AT THE NRTF-REPUBLIC.



FIGURE 4. ELF MODULATED WAVEFORM AND POWER SPECTRAL DISTRIBUTION.

$$|B| = \frac{\mu_{\rm r}\mu_{\rm o}I}{2\pi\sqrt{x^2 + h^2}}$$
(1)

$$|\mathbf{E}_{\mathbf{a}}| = \left(\frac{2\mathbf{V}}{\ln\left(\frac{2\mathbf{h}}{\mathbf{a}}\right)}\right) \left(\frac{\mathbf{h}}{\mathbf{h}^2 + \mathbf{x}^2}\right)$$
(2)

$$|\mathbf{E}_{e1}| = -\mathbf{j}\mathbf{f} \, \mathbf{I} \mu_0 \mu_r \ln\left(\frac{1.85}{x\sqrt{2\pi f \mu_0 \sigma_b}}\right) - \frac{\pi f \, \mathbf{I} \mu_0 \mu_r}{4} \tag{3}$$

$$|\mathsf{E}_{e2}| = \left(\frac{\mathrm{I}}{\pi \mathrm{I}\sigma_{e}}\right) \left(\frac{\mathrm{X}}{\mathrm{X}^{2} + \mathrm{d}^{2}}\right) \tag{4}$$

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where:	в	=	magnetic flux density	x	=	horizontal distance to antenna wire
	E,	=	air electric field	V	=	voltage on antenna wire
	E <sub>e1</sub>	=	induced earth electric field			radius of antenna wire
	E.2	=	conducted earth electric field	I	=	length of ground wire
	Ι	=	antenna or ground wire current	d		
	f	=	frequency of antenna current			
	$\mu_0$	=	magnetic permeability in free space			
	$\mu_{\rm r}$	=	material relative magnetic permeability	j		√-1
	Ι f μ <sub>0</sub>	8 8 8	antenna or ground wire current frequency of antenna current magnetic permeability in free space	$\sigma_{b}$		length of ground wire depth of buried ground wire bulk earth conductivity surface earth conductivity $\sqrt{-1}$

μ, h = height of antenna wire

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Inspection of Equation 1 and Figure 5a shows that the level of magnetic field generated by an ELF antenna is dependent only on the antenna current, the distance from the antenna, and the relative magnetic permeability ( $\mu_{i}$ ) of the medium the field is in. The relative permeability of most nonferrous objects and materials, especially those found in rural forested areas, is unity. As a result, the ELF magnetic field is unchanged at the boundaries between air and material such as soil, nonferrous rocks, water, plants, trees, and other living organisms. Similarly, it exhibits no dependence on weather or seasonal changes. Spatially, the magnetic field is highly predictable, and measurements at a given location are highly repeatable over time.

The antenna-generated air electric field, as depicted in Figure 5b and quantified by Equation 2, is a function only of the antenna's operating voltage and distance from the antenna wire. Unlike the magnetic field, the air electric field is oriented vertically at the earth's surface and terminates on it. Within ROWs and cleared areas near an antenna, the air electric field is grossly distorted by Equation 2, and measurement repeatability is good. However, the air electric field is grossly distorted by the presence of virtually all objects, including trees and other vegetation. Near such perturbations it can vary greatly over small distances. The amount of field perturbation will vary daily with precipitation, seasonally as plants leaf out and defoliate, and annually as trees and shrubs grow. Under the forest canopy the vertical air field from the antenna is nearly completely shielded. At these locations as well as at those far from the antenna, the air electric field is primarily horizontal and exists only as a by-product of the electric field in the earth.

The electric field in the earth is defined and measured as the voltage between two points on the earth's surface spaced one meter apart (Figure 5c). Both the antenna and ground wire sources of earth electric field described by Equations 3 and 4 show dependence on earth conductivity, in addition to previously described parameters such as current and distance. Because of this, the uniformity of earth electric fields is tied directly to the uniformity of soil conductivity and is further affected by the presence of anomalies such as rocks, tree roots, pools of water, holes, ridges, and outcrops. Earth electric fields can thus change quickly and unpredictably over short distances, but tend to be spatially consistent when anomalies are avoided. Measurements made over time at the same location can also vary significantly because soil conductivity in turn is affected by soil moisture content and temperature, factors that change by day, season, and year.

EM fields from sources other than the ELF transmitting facilities are also of interest and importance to the Ecological Monitoring Program. The principal sources of such fields are power utility transmission and distribution lines. These lines generate the same types of EM fields as those produced by an ELF antenna, but at an unmodulated frequency of 60 Hz. The power line fields also couple to and induce 60 Hz currents on the ELF antenna wires, thereby making the antennas themselves secondary sources of 60 Hz EM fields. The 60 Hz EM fields are subject to the same sources of spatial and temporal variations as their

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ELF counterparts, but exhibit more drastic intensity changes over time because of their dependence on transmission and distribution system loading factors.

The earth's dc (static) magnetic field is also of interest to the monitoring program because of its reported use as a navigational cue by animals and because of its hypothesized synergistic interaction with ac fields in producing biological response. Variations in the dc field are generally quite small and/or occur at very low frequencies. There are no radio, television, microwave, or other communications transmitting antennas within several miles of any monitoring program study site.

#### 2.3 Operational History

In order for investigators to calculate the EM exposure regimes for their studies, they need data on the operating times of the ELF transmitting facilities in combination with measurements of EM field levels. Transmitter operations data are provided to IITRI by the Navy's Submarine Communications Project Office, and are entered and stored in a computer-based spreadsheet for ease of use. A graphical and tabular summary of the operating data with monthly breakdowns is generated for each year and is included as an appendix to IITRI's annual engineering report.<sup>12-21</sup> More detailed summaries for specific periods of interest are also generated for the study investigators on request.

The NRTF-Republic antennas were first energized in 1986 for low-power testing. Operation was for short, sporadic periods using one antenna at a time. Power levels were increased to 15 percent in 1987, and to 50 percent in 1988. During these years a cyclic pattern was employed where each antenna was on individually for five minutes and then off for ten. Energizations usually occurred during the standard workweek, with the antennas off at other times. Half-power testing continued through April 1989. Up to this point, NRTF-Republic operations consisted of about two hours or less of intermittent operation per operational day at a maximum of half power. The changing between antennas resulted in a large number of on/off cycles.

Full-power testing with the simultaneous operation of both antennas began in May 1989. The antennas were now left on when not required for testing, including nights and weekends, raising the average daily operating time to nearly 18 hours. The NRTF-Republic became an on-line Naval Communications Facility in October 1989. Since that time, it has operated essentially 24 hours per day at full power and modulated signal transmission except for twice-weekly periods of scheduled maintenance. The number of on/off cycles per year has also decreased to less than one-tenth the number that occurred during preoperational testing.

Summaries of NRTF-Republic operations are shown in Tables 1 and 2 for the NS and EW antennas for the years 1986 through 1992. Hours of operation per month and the number of on/off cycles are shown in the bar graphs of Figures 6 and 7. Virtually all study treatment sites are adjacent to one antenna, and their EM exposures are dominated by that antenna, while they are remote from the other antenna. Thus,

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			Average Hours On per On Day	Total On Time		
Period	Status/Conditions	Days On/ Total Days		h	%	On/Off Cycles
1984-02/86	Construction					
07/86-10/86	Low-power testing, 4 A, random cycle	37/123	1.7	64	2.2	259
11/86-03/87	Off	0/151	<del></del>			
04/87-06/88	10%-power testing, 15 A, 5 min on, 10 min off	215/456	1.9	398	3.6	4772
07/88-04/89	50%-power testing, 75 A, 5 min on, 10 min off	177/304	2.4	430	5.9	4966
05/89-09/89	Full-power testing, 150 A, 5 min on, 10 min off, or continuous	145/153	17.7	2556	69.6	774
10/89-12/89	Operational, 150 A	91/ 92	23.9	2180	98.7	211
01/90-12/90		365/365	21.8	7956	90.8	414
01/91-12/91		355/365	22.0	7812	89.2	451
01/92-12/92		352/365	21.8	7686	87.7	205

### TABLE 1. SUMMARY OF NRTF-REPUBLIC OPERATIONS, NS ANTENNA

summaries have been done separately for each antenna so that time exposures for individual treatment sites can be calculated appropriately. These tables and figures clearly delineate the transition of the NRTF-Republic from its testing phase to operational status, both by the marked increase in operating time and by the corresponding decrease in on/off cycling. The EW antenna data also show several months of reduced operating time during 1991 and 1992. These months correspond to periods when the EW antenna was undergoing repairs.

#### 2.4 Study Site Exposures

Annual surveys were performed in order to characterize the ELF EM environment at study sites (Figure 8). This included 76 Hz fields produced by the ELF System, the earth's dc geomagnetic field, and 60 Hz fields from electric power distribution. Since the earth's geomagnetic field is relatively unchanging over the term of these studies and since the amperage and voltage on the ELF antennas were constant over a given operational period (see Tables 1 and 2), yearly surveys are considered adequate for reliable estimates of EM exposure from these sources. Although 60 Hz fields are more variable than the others,

			Average Hours		al On me	_
Period	Status/Conditions	Days On/ Total Days	On per On Day	h	%	On/Off Cycles
1984-02/86	Construction					-
03/86-10/86	Low-power testing, 6 A, 10 A, random cycle	33/245	1.5	51	0.9	187
11/86-03/87	Off	C/151				
04/87-06/88	10% power testing, 15 A, 5 min on, 10 min off	223/456	1.8	408	3.7	4886
07/88-04/89	50%-power testing, 75 A, 5 min on, 10 min off	181/304	2.5	447	6.1	5170
05/ <b>89-09/89</b>	Full-power testing, 150 A, 5 min on, 10 min off, or continuous	145/153	17.6	2555	69.6	787
10/89-12/89	Operational, 150 A	92/ 92	23.8	2187	99	219
01/90-12/90	•	362/365	22.2	8031	91.7	651
01/91-12/91		282/365	22.0	6197	70.7	308
01/92-12/92		267/365	21.7	5791	66.1	178

### TABLE 2. SUMMARY OF NRTF-REPUBLIC OPERATIONS, EW ANTENNA

they were found to be generally similar between paired study sites and were markedly lower than 76 Hz intensities at treatment sites. Because of this they were not considered a confounder to these studies for possible effects from 76 Hz fields.

The EM fields at 76 and 60 Hz were measured with special field probes developed and fabricated by IITRI. The probes were used in conjunction with frequency-selective meters and were necessary custom filters. These instruments provided the sensitivity necessary to measure the very low field levels found at control sites and the selectivity to discriminate between the 76 and 60 Hz fields. The field probes were calibrated prior to and following EM field site surveys. All measurements and calibrations were conducted in accordance with IEEE standard 644-1987 for the measurement of power frequency electric and magnetic fields.<sup>26</sup> Meters were calibrated annually against NIST traceable standards. The earth's static geomagnetic field was measured with a commercially available flux-gate magnetometer, which has been calibrated annually by its manufacturer.



FIGURE 6. NRTF-REPUBLIC HOURS OF OPERATION PER MONTH.

Temporal changes in the antenna fields are generally small in comparison with spatial field variations across study sites. The number and locations of historic measurement points within study sites have therefore been selected primarily to define the sites' spatial field gradients. Control sites, which by design are located far from the ELF antennas, have small spatial field variations across them, and a single measurement location is usually sufficient to characterize the 76 Hz EM exposures. Multiple historic measurement locations, however, have been established on large control sites, such as the bird population transects and nesting bird plots, and also on control sites that have 60 Hz field gradients. Multiple measurement points are the rule on treatment sites because field exposures change rapidly over short distances. Exposure assessment strategies at treatment sites have included individual measurement points to characterize each of several isolated experiment locations, multiple periphery measurement points to



FIGURE 7. NRTF-REPUBLIC ON/OFF CYCLES PER MONTH.

bound exposures on moderate-sized plots, and measurement points spaced along a transect perpendicular to the antenna to profile the field gradients across large or irregular plots.

Paired study plots were initially sited to maximize the difference in 76 Hz EM exposures between treatments and controls while at the same time matching ambient 60 Hz EM exposures at both. EM exposure criteria were established to quantify the paired site exposure relationships and ensure that all study sites met a minimum set of EM standards. The exposure ratio equations are as follows:



FIGURE 8. SITES OF MICHIGAN ECOLOGICAL STUDIES.

	(
$T_{76 \mu_{7}}/C_{76 \mu_{7}} > 10$	(5)
	(-)

$$T_{76 Hz}/T_{60 Hz} > 10$$
 (6)

$$T_{76 Hz}/C_{60 Hz} > 10$$
 (7)

$$10 > T_{e_0 Hz} / C_{e_0 Hz} > 0.1$$
 (8)

where:  $T_{76 Hz}$  = treatment site EM exposure from ELF antenna fields

 $C_{78 Hz}$  = control site EM exposure from ELF antenna fields

 $T_{eo Hz}$  = treatment site EM exposure from power line fields

 $C_{60 Hz}$  = control site EM exposure from power line fields

The above exposure ratios require that ELF antenna field exposures at a treatment site be at least 10 times larger than those at its matched control site and 10 times larger than the 60 Hz field exposures at both sites. Further, 60 Hz field exposures at paired sites should not differ by more than a factor of 10.

F es 9 to 11 summarize graphically the measured 76 Hz EM exposures for the NRTF-Republic study sites. They illustrate the total range of exposures for full-power antenna operation over the five-year period from 1989 through 1993 for all treatment and all control sites of each study. The fact that most of the gaps between the treatment and control exposure ranges span at least a decade clearly shows that the antenna field exposure ratios are generally greater than 10 for any site pairing within a study. In fact, all actual site pairings meet cr exceed the exposure ratio criteria, with one exception.

Additional EM exposure information labeled "NS antenna operation only" is provided in Figures 9 to 11 for the upland flora (trees) treatment sites. These sites are located adjacent to the EW antenna, and their EM exposures were significantly reduced during the months when the EW antenna was off for



FIGURE 9. MAGNETIC FIELD EXPOSURES AT STUDY SITES, 1989-1993.

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FIGURE 10. EARTH ELECTRIC FIELD EXPOSURES AT STUDY SITES, 1989-1993.

repairs (Figure 6). Because of mutual EM coupling between the antennas, exposures at the upland flora sites with the EW antenna off were about one-third those experienced with both antennas on (Figures 9-11). EM exposure ranges for the antenna testing periods from 1986 through early 1989 can be found in Reference 12.

Figures 9 through 11 present study site EM exposure ranges for both ELF antennas or solo operation of the antenna nearest the treatment sites. As such, they can be used directly with the monthly and annual hours-of-operation data presented in the previous section to calculate time-intensity exposures. Interestingly, because the exposure times are the same for both treatment and control sites, paired site ratios for time-intensity exposures will have the same value as the ratios for intensity exposures.



FIGURE 11. AIR ELECTRIC FIELD EXPOSURES AT STUDY SITES, 1989-1993,

Time-intensity exposures for the antenna testing periods from 1986 through early 1989 can also be computed using the operational data and estimates or measurements of the EM exposures for these periods. It should be noted that study site time-intensity exposures show a more dramatic increase at the transition from antenna testing to operational phases than do the intensity exposures alone because both intensity levels and operating times increased significantly at that point.

Seasonal changes in antenna-generated EM fields, as previously stated, are generally smaller than the spatial changes in field intensity across treatment sites and certainly much smaller than the incremental changes experienced during antenna testing. The earth electric field, with its dependence on soil conductivity, is the most likely to exhibit seasonal change. Data loggers have been used to monitor the earth electric field for studies of soil organisms. The results of this monitoring indicate that the overall range of change in the earth electric field exposures caused by seasonal factors is 10 to 30 percent. Shorter term changes in exposures such as those associated with precipitation events typically range from

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10 to 20 percent. Some sites have also been observed to exhibit distinct diurnal patterns of earth electric field variation, but these changes are typically less than 5 percent.

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#### 3. MONITORING STUDIES

Physiological, developmental, behavioral, and ecological variables for dominant biota in upland, riverine, and wetland habitats near the Navy's ELF Communications System are being monitored for possible EM effects. The biota and ecological relationships selected for study are important to the ecosystem that they inhabit and are of interest to local residents.

Upland forest is the dominant habitat in the ELF System area. The main methods for the transfer of energy within this habitat are the production and breakdown of organic plant structures such as leaves. Organic matter turnover and distribution are regarded as major determinants of the number and types of biota that occur within the forest. Because the production and decomposition of organic matter are important and possibly affected by exposure to non-ionizing EM fields, these ecological processes and associated organisms are being monitored for possible effects from operation of the ELF System.

Aspects important to the production of organic matter by forest vegetation are presented in Section 3.1. Organisms and processes important to organic matter decomposition are presented in Sections 3.2 (bacteria and fungi), 3.3 (soil amoebae), and 3.4 (soil and litter arthropods and earthworms). In addition, native bees are studied because they are prominent pollinators of resident flowering plants (see Section 3.5).

Small mammals and birds inhabiting upland forests are examined as organisms of intrinsic interest to the public and because they serve as indicators of possible adverse effects to all vertebrates, including humans. Organismal and populational characteristics of small vertebrates are addressed in Sections 3.6 and 3.7.

The ELF System area encompasses numerous headwater streams flowing into the Mississippi and Great Lakes drainage systems. Energy is supplied to this habitat by organic materials produced by microscopic aquatic plants and riparian vegetation. Some aquatic insects feed on the plant materials, making organic compounds and energy available to higher trophic levels, such as fish, as their own biomass. Populational aspects, as well as the functional and structural components of these three trophic communities (i.e., periphyton, aquatic insects, and fish), are being monitored for possible EM effects. They are addressed in Section 3.8.

The following sections summarize the design and results of each of the eight studies that were performed near the NRTF-Republic from 1982 through 1992. The general types of biota being examined are used as numbered, underlined subsection titles, while specific study elements are presented as unnumbered, underlined titles within each numbered subsection. In order to simplify the presentation of statistical results, any difference described as "significant" had a significance level of 5 percent (P < 0.05).
### 3.1 Upland Flora

Since 1982, researchers from Michigan Technological University have examined the following study elements for possible changes in forest productivity and health:

- phenology and morphological characteristics of herbs
- · growth rates of hardwood and pine trees
- physiology of hardwood and pine trees
- litter production
- numbers and kinds of mycorrhizae on red pine seedlings

Productivity and health have been monitored at three upland sites near the NRTF-Republic. Two treatment sites are situated so that one site is adjacent to an overhead antenna element and the other is adjacent to a grounding element. A single control site is located more than 28 miles from any NRTF element. The antenna and control sites each consist of hardwood tree plots (existing pole-size trees), plots planted with red pine trees, and plots of herbaceous plants. The grounding treatment site consists of plots planted with red pine only.

<u>Herbaceous Plants</u>. The starflower, an abundant herb in the ELF System area, has been monitored since 1985. This short-lived plant is sensitive to subtle changes, and in some respects show quicker responses to environmental perturbations than other vegetation such as trees.

Phenological events and morphological characteristics of naturally growing starflower plants have been monitored at the antenna and control sites. The phenological events examined include the timing and rate of select developmental events such as flowering, fruiting, and rates of stem expansion. Morphological characteristics such as the numbers of plant structures were also observed. Analysis of variance (ANOVA), analysis of covariance (ANCOVA), and regression analysis were used to examine these data. Data collection on herbaceous plants was concluded in 1992; however, analysis of data will continue during 1993.

Although the time of onset for most developmental events differs between treatment and control sites, the pattern of differences has generally remained the same throughout the study. In 1990 and 1992, but not in 1991, there were significant differences between sites in the timing of flowering and fruiting, as well as in the number of plants flowering. Because the overall pattern across years did not match that of EM exposures, researchers concluded that there were no EM effects (Table 3).

No differences were evident between the morphological characteristics of starflowers growing on treatment sites and those growing on control sites (Table 4).

<u>Hardwood Tree Growth</u>. Studies have shown that tree growth is a good indicator of productivity, and that tree growth can be affected by anthropogenic factors. Accordingly, tree growth in existing hardwood stands has been monitored in order to detect possible changes in forest health due to the operation of the ELF System.

Parameter	ELF EM Effect
Stem expansion	None
Leaf expansion	None
Bud formation	None
Flowering	None
Fruiting	None
Leaf senescence	None

#### TABLE 3. PHENOLOGICAL CHARACTERISTICS OF STARFLOWERS

## TABLE 4. MORPHOLOGICAL CHARACTERISTICS OF STARFLOWERS

Parameter	ELF EM Effect
Leaf area	None
Stem length	None
Number of buds	None
Number of leaves	None
Number of flowers	None
Number of fruit	None

Since 1984, changes in trunk diameter have been measured throughout the growing season for hardwoods abundant in the ELF System area (maple, oak, birch, and aspen). These measurements provide data on total yearly growth as well as on rates and patterns of growth throughout each season. Parameters related to hardwood tree productivity (e.g., stand structure, ingrowth, and mortality) were also recorded for assistance in interpreting these data.

The hardwood growth data were examined using ANOVA and ANCOVA. The latter technique accounts for site and year-to-year differences in covariates (soil and climatic factors) that are known to affect tree growth. ANCOVA is constrained when there is a nonlinear relationship between growth and covariates, or when there is a significant correlation between covariates and EM exposures. Although no physical basis exists for connecting the latter pair, significant correlations have been found between them. Because of this, both hardwoods and pines were also examined using nonlinear models of tree development that involved comparison of model-predicted growth to that actually observed (Figure 12).



FIGURE 12. OBSERVED AND MODEL-PREDICTED RED PINE HEIGHT GROWTH, 1990-1991.

ANCOVA showed no significant differences between sites in total annual growth for any of the hardwood species. However, significant correlations were found between two of three covariates and magnetic field exposures at the antenna site. In order to supplement the ANCCVA, growth models were used to examine changes in diameter. Diameter growth patterns for all four species, and growth residuals for oak and birch, did not indicate any ELF EM effect. Annual growth residuals for red maple and aspen (Figure 13) showed a pattern consistent with growth stimulation at magnetic flux density exposures between 1 mG and 7 mG. Overall conclusions on hardwood growth are summarized in Table 5.

TABLE 5.	<b>GROWTH CHAP</b>	IACTERISTICS OF	HARDWOOD	TREES	(1984-1992)
----------	--------------------	-----------------	----------	-------	-------------

	ELF EM Effect				
Parameter	Maple	Oak	Birch	Aspen	
Total annual growth	None	None	None	None	
Annual growth residuals	Possible	None	None	Possible	
Growth Pattern	None	None	None	None	



FIGURE 13. ASPEN DIAMETER GROWTH MODEL RESIDUALS FOR THE ANTENNA TREATMENT SITE, 1986-1991.

<u>Pine Tree Growth</u>. Because young trees experience more rapid growth than older trees, red pine seedlings were planted and monitored for possible growth effects due to their exposure to ELF EM fields.

After their planting in 1984, 300 pine seedlings at each of three sites were randomly selected and marked for long-term monitoring of pine growth. The basal diameter and total height of each marked seedling was measured periodically through the growing season. Growth changes were examined using ANOVA, ANCOVA, and a height growth model.

Analyses showed significant differences between sites and years (1985-1991), as well as significant site-by-year interactions, in the height and diameter growth of red pine. Generally, diameter growth has been consistently greater at the antenna site, and height growth cenerally greater at the control site. Significant correlations were found between the covariates and magnetic field exposure; therefore, height growth was also examined using a model. Model residuals support the possibility of stimulating growth at about the same magnetic flux density levels as for the hardwoods. There were no indications of a significant difference between observed and predicted seasonal growth patterns (Table 6).

<u>Pine Tree Physiology</u>. Optimum tree growth depends on many factors, including the normal functions that allow adequate uptake and translocation of both water and nutrients. Therefore, leaf water potential (LWP) and foliar nutrients of pine have been monitored since 1986. If exposure to ELF EM fields

Parameter	ELF EM Effect
Total annual diameter	Possible
Total annual height	Possible
Seasonal height pattern	None

# TABLE 6. DIAMETER AND HEIGHT GROWTHOF RED PINE TREES (1985-1991)

affects physiological processes, changes in water or nutrient variables could be more quickly perceived than changes in overall growth. Water and nutrient variables are also important because they are indicators of plant stress.

Average annual LWP for 1986 through 1992 were examined using ANCOVA. No significant differences were found among sites; however, the investigators noted significant differences both among years and a significant site-by-year interaction. The relationship of LWP between sites and preoperational/ operational conditions was quite variable, and did not correspond to EM exposures.

Samples of one-year-old foliage were collected annually and were then analyzed for nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), and magnesium (Mg). Foliar concentrations of N decreased, while concentrations of Ca and Mg have increased at all three study sites over the study period. These changes appear to be related to the initial increase in soil nitrogen after plantation establishment and/or increased maturity of trees, rather than an increase in EM field exposure. Differences in foliar concentrations of Ca, Mg, and P among sites were not significant either during years prior to or during full antenna operation.

Researchers conclude that there are neither EM effects to the uptake and movement of water and nutrients within trees (Table 7), nor stress to EM exposed trees. The collection of data on LWP was completed at the end of 1992; the collection of data on foliar nutrients will proceed through 1993. Investigators continue to search for covariates that could account for the variability in the data.

<u>Hardwood Tree Physiology</u>. Actively photosynthesizing red oak foliage was collected throughout the growing season and was examined for macronutrient content (N, P, K, Mg, and Ca). This analysis monitors nutrient accumulation as well as the timing of translocations from the foliage to the branches prior to leaf fall.

Nutrient concentrations in red oak foliage showed considerable variability between the antenna and control sites both before and after full-power operation of the antenna. Results from covariate analyses using soil and climatic data showed no significant site-year interactions for any foliar nutrient. Researchers conclude that the differences in red oak nutrient concentrations between the antenna and control sites were not related to operation of the ELF System (Table 8).

Parameter	ELF EM Effect
Water potential	None
Macronutrients	
N	None
Р	None
к	None
Mg	None
Ca	None

#### TABLE 7. LEAF WATER POTENTIAL AND FOLIAR NUTRIENT CONTENT OF RED PINE

TABLE 8.	FOLIAR NU	TRIENTS OF	RED OAK

Parameter	ELF EM Effect
N	None
Ρ	None
К	None
Mg	None
Ca	None

Litter Production. Litter fall is an important component in the transfer of nutrients and energy within the upland ecosystem. The sensitivity of foliage production to overall physiology and ambient conditions (including EM exposure) also make this parameter an important indicator of possible ELF EM effects on trees. The total weight and nutrient content of litter produced by trees abundant in the ELF System area were therefore monitored to provide estimates of seasonal production and nutrient inputs to the decomposition system.

Litter was collected in traps on existing hardwood stands at the antenna and control sites from 1984 through 1992. The litter was dried, sorted, and weighed according to the following components: foliage, wood, and miscellaneous (e.g., seeds). A subsample was taken to determine the nutrient content of the litter.

Climatic variables, soil nutrients, and magnetic flux densities were used in testing of differences in leaf litterfall weight and nutrient concentration between sites and years. No pattern of significant site differences in yearly litterfall weight relative to operation of the ELF System were found. Significant site differences in nutrient concentration were found only for total phosphorus in one preoperational year (1985)

and in one full-power year (1992), indicating that ELF magnetic flux densities had no detectable effect on nutrients (Table 9).

	ELF EM Effect					
Parameter	Weight	N	Р	К	Mg	Ca
Foliage (composite)	None	None	None	None	None	None
Oak	None	None	None	None	None	None
Hazelnut/birch	None	None	None	None	None	None
Aspen	None	None	None	None	None	None
Maple	None	None	None	None	None	None
Wood (composite)	None	None	None	None	None	None
Miscellaneous (composite)	None	None	None	None	None	None

TABLE 9.	LITTERFALL	CHARACTERISTICS	OF	HARDWOOD TR	EES
			_		

<u>Mycorrhizal Populations</u>. Mycorrhizal fungi form a symbiotic relationship with the roots of trees. The fungi use organic compounds synthesized by the tree for their growth and to "forage" for minerals and water in the soil using mycelia. In turn, the fungi provide the tree with minerals and water more efficiently than the tree roots alone. This relationship is considered essential to the satisfactory growth of nearly all tree species. Because the growth of fungal mycelia is dependent on physiologically produced intracellular electrical currents, other sources of electrical current, such as the ELF Communications System, may also affect the fungi.

Possible effects to mycorhizzal populations associated with pine roots were characterized by their types and numbers. In 1992 as in previous years, Type 3 mycorrhizae were the most common, Type 5 the second most common, and Type 6 the least common encountered at the study sites. The total number of mycorrhizae declined over the period 1985-1988, but increased slightly over the period 1989-1991 (Figure 14).

ANOVA and ANCOVA showed no significant differences between sites or significant site-by-year interactions. Because there was no change in the types or number of mycorrhizae on treatment sites (i.e., a significiant site-by-year interaction) after the antenna became operational, researchers conclude that there were no EM effects (Table 10).



FIGURE 14. TOTAL NUMBER OF MYCORRHIZAL ROOT TIPS PER GRAM OF RED PINE ROOT.

TABLE 10.	MYCORF	RHIZAL	PÒPUL	<b>_ATIONS</b>
ASSC	CIATED	WITH R	ED PIN	٩E

Parameter	ELF EM Effect
Total number	None
Туре 3	None
Type 5	None
Туре 6	None

# 3.2 Soil Microflora

Researchers at MTU have also been monitoring microflora inhabiting the soils near the NRTF-Republic. Soil microflora (bacteria and fungi) are being studied because they are important in the processing of plant litter and inorganic gases into compounds usable by vegetation, and because they are infective agents that cause disease in trees.

In order to monitor the range of microbial functions for possible bioelectromagnetic effects, researchers have examined the following study elements:

- bacterial population ecology
- litter decomposition
- root disease epidemiology

Bacterial population ecology and litter decomposition elements are closely related to the mycorrhizal and litter production elements examined in the upland floral studies (Section 3.1). The incidence and progress of root disease has been monitored since such disease first occurred among the pine trees planted on study sites.

Although examined by different research teams, the upland flora and soil microflora projects share common study sites and the ambient monitoring data taken there. Eight years of decomposition data have been collected for leaf litter emplaced in hardwood stands and pine plantations. Streptomycete bacteria have been monitored on pine plantations since 1985, and the incidence of *Armillaria* root disease since 1986.

<u>Streptomycete Bacteria</u>. Microfloral activities are important in the orderly transfer of nutrients to vegetation, and changes in their population dynamics have been shown to influence the rate of organic matter turnover. It has also been reported that EM fields can affect microorganisms (e.g., slime mold physiology and bacterial orientation). EM fields produced by the ELF System are therefore being examined as a human-induced factor possibly affecting soil bacteria.

Streptomycete bacteria degrade organic molecules, nourish mycorrhizae, and influence neighboring microbial populations through their production of compounds such as antibiotics and vitamins. Streptomycetes are part of the indigenous soil and root-related microflora, and their populations do not normally undergo large changes in habitats such as used in these studies. For these reasons, the types and numbers of streptomycetes associated with red pine mycorrhizae were selected for monitoring.

Seven years (1985-1991) of mycorrhizoplane streptomycete population data have been collected. Samples were taken monthly from May through October from pine trees located on plantations at the antenna, ground, and control sites. Plate count data for morphotypes and population levels associated with predominant (Type 3) mycorrhizal fine roots were expressed as numbers per gram of root. Samples were also analyzed for bacterial-isolate ability to degrade complex organic molecules.

Data for streptomycete morphotypes and numbers were logarithmically transformed prior to statistical analyses. The data were then examined by ANCOVA, using weather-related variables. Whenever significant differences were detected, pairwise comparisons of means were examined using other statistical options such as least-square means procedures. The power of the experimental design was calculated as detection limits, i.e., the percentage difference between two sample means that would be detected 50 percent of the time with alpha of 0.05.

Over the period of study, the number of morphotypes initially decreased and then stabilized at all three study sites. This pattern probably reflects the establishment of those types most capable of survival within the pine plantations. ANCOVA showed significant differences between years, but no significant site differences or site-by-year interactions. Significant shifts in morphotype numbers greater than 14 to 26 percent among years, or of 8 to 9 percent among sites, should have been detectable more than 50 percent of the time. That is to say, two to four of the 20 morphotypes identified could have been lost before such a change was detected by investigators.

Representatives of each streptomycete type were also tested for their ability to degrade calcium oxalate, cellulose, and lignocellulose. Each morphotype could degrade one or more of these organic compounds, indicating no detectable change in either morphotypes or their ability to degrade organic compounds over the period 1988-1991.

The total number of streptomycetes present in October samples has been lower than the number isolated from samples taken from May through September. Total numbers have not shown a recognizable pattern across years. ANCOVA showed significant differences between years, but none between sites, The lack of significant site-year interaction indicates a constant temporal relationship between sites. Shifts in streptomycete levels of 21 to 37 percent among years, or of 12 to 13 percent between sites, should have been detectable 50 percent of the time.

These studies were completed at the end of the 1991 growing season, although data analysis may continue. Based on the work performed to date, investigators conclude that there were no ELF EM effects on streptomycete populations (Table 11).

TABLE TI. SUIL STREPTUMYCETE POPULATIONS		
Parameter	ELF EM Effect	
Morphotype	None	
Total number	None	
Ability to degrade organic compounds	None	

Litter Decomposition. MTU has also been monitoring the overall functioning of the decomposer community for possible EM effects by examining the loss of mass from leaf litter.

The decomposition of maple, oak, and pine foliage has been monitored on plantations and hardwood stands since 1985. Preweighed, bulk samples from a single source were emplaced at study sites in December of each year. Subsets of foliage samples were then retrieved from May through

November of the following year. Retrieved samples were reweighed, and decomposition was expressed as the percentage of original dry matter mass remaining. Mass loss data are complete for samples retrieved from 1985 through 1992.

Data were transformed to the arcsin square root to homogenize variances prior to ANCOVA. Two types of ANCOVA were used to statistically examine the data. An 'effects model' examined for differences between sites and years as well as for site-by-year interactions, whereas a 'means model' was used to identify changes in the relationship between sites over the study period. Covariates were based on seasonal inputs of energy and precipitation to the decomposition system. A covariate to address sampling date differences between years was also used.

ANCOVA (effects model) of data collected for samples emplaced in plantation plots showed no statistically significant differences; however, decomposition at hardwood stands indicated many significant differences between sites and years, as well as interactions suggesting possible EM effects. Other data collected at hardwood sites also suggested that EM fields may accelerate the rate of decomposition. Prior to 1988, decomposition progressed significantly faster at control than at antenna hardwood stands, while decomposition proceeded significantly faster at the antenna site from 1988 through 1992 (Figure 15).



FIGURE 15. ANNUAL PROPORTION OF OAK LEAF LITTER DECOMPOSING IN HARDWOOD STANDS.

Not all site differences were significant during the period 1988-1992. Those differences that were significant were not large. Although there was a significant difference between sites in the decomposition of oak during 1992 (Figure 16), there was no such difference for maple or pine foliage. Possible confounding of covariates and EM exposure has not yet been examined for. Detection limits between sites were below 2 percent, between years 3 to 8 percent (pine-oak), and between site-years (means model) 3 to



FIGURE 16. PROPORTION OF INITIAL MASS REMAINING FOR BULK OAK LEAVES DECOMPOSING AT HARDWOOD STANDS.

10 percent. Researchers conclude that the results to date indicate possible EM effects to the decomposition of all litter species (Table 12).

Parameter		ELF EM Effec	zt
	Pine	Oak	Maple
Decomposition	Possible	Possible	Possible

TABLE 12. DECOMPOSITION RATES OF LEAF LITTER (M	(MTU)	TTER	LIT	EAF	U	OF	RATES	OSITION	DECOMP	12	TABLE
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Data collection is to continue during 1992-1993. The biological relevance of spatial and temporal differences remains an important issue to be resolved.

<u>Red Pine Root Disease</u>. The occurrence of a lethal root disease caused by the fungus Armillaria was first documented at MTU pine plantations in 1986. Because the spread of this disease is often enhanced by stressors, investigators are examining the rates of disease progress in relation to EM exposures.

The pathogen responsible for the root disease was obtained from dead seedlings and mushrooms collected at the plantations (Figure 17). Isolates were cultured and clones were identified. All clones responsible for pine mortality belonged to a single species, *Armillaria ostoyae*. Other *Armillaria* are present at the study sites, but they are not pathogenic toward red pine.



FIGURE 17. ARMILLARIA ROOT DISEASE ON A RED PINE SEEDLING.

Environmental factors may influence the spread of root disease. Therefore, estimates of disease progress were made by deriving rate constants based on the overall pine mortality at each plantation. Preliminary comparison of the rate constants using ANOVA showed a statistically significant difference between sites (antenna > control > ground). These results will next be compared to results from ANCOVA using precipitation, pine height, and number of hardwood stumps as covariates.

A ostoyae clones occupy a smaller area of the ground plantation than similar clones present on the overhead antenna or control plantations. Because of this, mortality counts among plantations may not be an appropriate test for ELF EM effects on root disease. Accordingly, investigators plan to also compare disease progress between sites based on the land area occupied by individual clones.

Investigators have not reached a conclusion concerning EM influence on the spread of pine root disease. They were to continue testing data on the occurrence of the disease during 1993, and were to

use ANCOVA to examine differences between sites using rate constants derived from both clonal areas and overall plantation mortality.

### 3.3 Soli Amoebae

Soil amoebae are common soil organisms that feed on bacteria. Bacteria, in turn, are important to the soil ecosystem because of their ability to mobilize nutrients needed for plant growth. Protozoa, to the extent that they affect the number and types of bacteria in the soil, also become a potentially important factor in soil fertility. Studies on protozoa and similar organisms have suggested that their orientation, growth, and physiology may be affected by EM fields. Indeed, when in the vegetative state, amoebae exist as "naked cells" closely coupled to electric fields and currents flowing in the soil.

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

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

Parameters indicative of soil fertility have also been monitored.

Studies on soil amoebae were being performed at three study sites in Michigan. One treatment site was located adjacent to an overhead antenna of the NRFT-Republic; the other was located adjacent to a buried grounding element. A third site, the control site, was located about nine miles from the nearest ELF System element.

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

Soil samples were taken with a coring device for taxon identification and population studies. The coring locations within study sites were 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. A soil-dilution counting technique was then used to determine the population size of each sample. Prior to 1992, samples were taken from both horizons on a monthly regime from June through October. In 1992, sampling was limited to the organic horizon during July, August, and September.

The total amoeba population consists of both vegetative (actively reproducing) and encysted forms. The size of the total amoeba population appears to be related to soil moisture. Accordingly, population sizes vary from year to year (Figure 18), and greater numbers of amoebae are found in the organic horizon. ANOVA generally has not shown significant differences between sites in the total number of amoebae. Significant differences between sites have been more frequent for numbers of cysts; however, no clear pattern has emerged that can be related to EM exposure.



FIGURE 18. AVERAGE NUMBER OF AMOEBAE PER GRAM OF SOIL (ALL STUDY SITES).

<u>Species and Strain Characterization</u>. Since the beginning of these studies in 1983, eight types of amoebae (various generic and species levels) have been consistently isolated using sample enrichment techniques. To date, no differences between years or between sites have been reported in the types of amoebae present.

The genetic diversity within a single species of soil amoeba, *Acanthamoeba polyphaga*, was determined by isoenzyme analysis. This parameter was monitored from 1985 through 1988, when its scope was reduced. Limited study continued at all sites from 1989 through 1991 by monitoring the genetic heterogeneity of the same species reared in *in situ* culture chambers. Soil populations of *A. polyphaga* were again monitored in 1991 to determine if any changes had taken place.

No ELF EM effects were indicated in either cultured or soil populations until 1991, when natural populations in the soil at the antenna site had a significantly lower diversity than in previous years and at other sites. Issues raised by peer reviewers concerning sample sizes, culturing techniques, and data interpretation were to be addressed by continued study during 1993.

<u>Growth and Feeding Activity</u>. The purpose of this element is to determine the *in situ* growth and feeding activity (i.e., predation on bacteria) of soil amoebae in culture chambers located at all three study sites.

The study protocol involved suspending a known species of amoeba (A. *polyphaga*) and a food bacterium in a physiological saline, all contained in a culture chamber. In order to provide electric fields and currents, the chambers were connected to buried collecting electrodes. Culture media with bacteria were replaced on a two- to three-week cycle, using EM-exposed amoebae from old cultures to inoculate the new media. Periodic counts of amoebae were made to determine changes in the number of organisms. Over the period 1989-1991, the genetic heterogeneity of the cultured amoebae was determined at the onset and termination of the experimental period.

A logarithmic transform of the growth data provided a straight-line plot (numbers over time), which was then quantified by regression analysis. Using a modified t-test, the resulting slopes of the lines were compared to examine for statistically significant differences between sites. There were no significant differences between sites in amoebal growth, and the generation time for all cultures ranged from 16 to 18 hours. There were no changes in the genetic heterogeneity over the experimental period.

Table 13 presents overall conclusions concerning possible EM effects on soil amoebae.

Parameter	ELF EM Effect
Total number	None
Number encysted	None
Species diversity	None
Genetic diversity	Possible
Growth	None

TABLE 13. THE SOIL AMOEBA COMMUNITY

#### 3.4 Soil and Litter Arthropods and Earthworms

Arthropods and earthworms play a major role in the initial decomposition of vegetation by shredding plant material such as leaves and redistributing the remains in the soil. These and other soil organisms are also closely coupled to electric fields and currents generated in the soil by the ELF Communications System. Therefore, this project has been monitoring various aspects of the soil and litter invertebrate community for possible EM effects.

The project uses one treatment site located adjacent to the antenna ROW at the NRTF-Republic and one control site located at a distance west of the antenna. Both sites are situated in a mapledominated deciduous forest. Although the sites display faunal differences, they have similar soils, vegetation, and microclimate. Indices of community structure as well as population characteristics of major fauna common to both sites were emphasized in these studies. In order to address faunal differences between sites, intersite comparisons of either ecological equivalents or major populations common to both sites were examined. Temporal changes in community and population characteristics were used for preoperational and operational comparisons. Possible changes in the overall functioning of the soil community were monitored by determining rates of litter decomposition.

<u>Surface-Active Arthropode</u>. This project element examined the major arthropod fauna inhabiting the surface layers of the soil.

Diel and seasonal activity patterns of surface-active arthropods were assessed by consecutive, day and night, pit-trap samples taken once a week. Major groups trapped were springtails, mites, and ground beetles. Data were collected from 1985 through 1991, when this project element was completed. Community characteristics and seasonal activity of these groups were analyzed using before and after, control and impact (BACI) analysis. Investigators considered the "before" or preoperational period to be 1985-1988, and the "after" or operational period to be 1989-1991.

There were no significant differences in the overall temporal/spatial relationships of springtail diversity or evenness. The data showed a higher diversity at both sites during the fully operational period of the antenna than during the preoperational period, while evenness indices changed little between those two periods. Seasonal activity patterns of springtails (as evidenced by the number trapped) were highly variable and correlated between sites. BACI analysis showed no significant differences in springtail community characteristics or seasonal activity patterns.

The few species of mites that were trapped did not well represent the overall community, and therefore were not examined from this aspect. However, three species were caught in numbers sufficient for examination of their activity patterns. No significant temporal differences in the intersite/operational relationship of this parameter were revealed by BACI analysis, and graphs of seasonal frequency of developmental stages of two species did not show any EM effects on their life cycle. Catches of the remaining species consisted entirely of adults.

BACI analysis indicated a significant change in the overall diversity relationships of the ground beetle community between sites after the antenna became fully operational. These findings are due to a significant reduction in diversity indices during the operational period. The reduction occurred at the control site but not the treatment site, and therefore was not considered an ELF EM effect. No significant differences in evenness were revealed. Seasonal activity patterns of abundant beetle species during operational years were consistent with preoperational observations.

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At the time of reporting, variations in sex ratios and diel activity preferences of ground beetles had not been analyzed in detail. Yearly summaries show that the ranges of preoperational and operational values overlap.

Investigator conclusions on possible EM effects on surface-active arthropods are given in Table 14.

Parameter	ELF EM Effect		
	Mites	Beetles	Springtails
Taxon evenness		None	None
Taxon diversity	-	None	None
Activity patterns	None	None	None

TABLE 14.	COMMUNITY CHARACTERISTICS AND SEASONAL ACTIVITY	1
	OF SURFACE-ACTIVE ARTHROPODS	

Soil and Litter Arthropods. The population and community dynamics of soil and litter arthropods are being determined from samples taken during the growing season (May through October). Litter and soil are sampled separately. The arthropods are then extracted by heat and sugar flotation techniques. All sampling and extraction of soil arthropods was completed at the end of 1992 as planned. Identification and enumeration of arthropods collected in 1992 vas to proceed during 1993. Springtails and mites were the most numerous taxa in the litter and soil of both sites, and were the major groups of interest. At the time of reporting, data were available for the period 1984-1991.

Springtails were more abundant at the control site than at the treatment site (Figure 19). Temporal fluctuations in total numbers and developmental stages, however, were highly correlated between sites. Three of seven species representative of the springtail community indicated significant changes in the intersite relationship of their seasonal abundance. Based on multiyear observations, investigators suggest that non-EM ambient factors produce occasional differences between sites.

Since 1984, 73 species of springtails have been identified at the study sites, and in any given year 44 to 55 species have been collected. Most these species are recovered from both litter and soil horizons. The distribution of individuals among species was significantly different before and after full operation of the antenna. The evenness of the springtail community at the control site has experienced a significant change in this index; however, there has been no significant change in the treatment community. Community diversity (which includes the distribution of individuals among species) showed no significant temporal differences in the intersite relationships. For these reasons, investigators conclude that those



### FIGURE 19. MEAN ANNUAL NUMBER OF TOTAL SPRINGTAILS (1984-85 NOT CORRECTED FOR EXTRACTION EFFICIENCY).

differences found to be significant were not consequential, and result from reasons other than EM exposure.

Identification of all mite species found in the litter and soil is an intractable problem; therefore, only two easily identified and relatively abundant species were selected for this monitoring. Year-to-year changes in density have been similar at both sites for each of the two species. No evidence was found of EM effects on overall abundance or developmental stages. Investigator conclusions are summarized in Table 15.

	ELF EM Effect		
Parameter	Mites	Springtails	
Density (soil)	None	None	
Density (litter)	None	None	
Population structure	None	None	
Species eveness	-	None	
Species diversity		None	

#### TABLE 15. POPULATION AND COMMUNITY CHARACTERISTICS OF SOIL AND LITTER ARTHROPODS

<u>Earthworms</u>. Alternating electrical currents are known to force earthworms to the surface of the soil. Accordingly, this project element monitors the behavior and other characteristics of the earthworm community for a possible reaction to chronic exposure to the relatively low-intensity EM fields produced by the NRTF-Republic.

Soil and litter samples were taken at regular intervals from May through October. Worms and their cocoons were extracted, identified, and enumerated. Seven of the eight species identified were found at both treatment and control sites; however, the distribution of individuals among species was markedly different between the sites. At the control site, three species were particularly abundant, while at the treatment site only one species was numerically dominant. Overall, the treatment site had fewer individuals but markedly greater biomass than did the control site (Figure 20).





Reproductive characteristics of the dominant worm species at the treatment site, Aporectodea tuberculata, were compared to those of a "sister species," Aporectodea turgida, at the control site. Preoperational data on two of three reproductive parameters for these sister species were highly correlated ( $R^2 > 90$  percent). BACI analysis showed significant differences in this intersite relationship after the antenna became fully operational. There were no apparent changes in the reproductive characteristics of *A. tuberculata* at the treatment site before or after full operation of the NRTF-Republic; however, investigators contend that these characteristics should have changed as they did for *A. turgida*.

Throughout the study of A. *tuberculata*, the number and relative proportion of reproductive adults (clitellates) were significantly correlated to both soil moisture and temperature. However, correlation coefficients were lower during the operational period of the antenna. The principal investigator ascribes the reduction in correlation coefficients to EM exposure during the operational period. Investigators also found that the number of cocoons produced per clitellate worm (worm production) was higher in the period after full operation of the antenna. The implication of these results is unclear, because one method of calculation showed statistically significant temporal differences, while another method did not.

In order to reduce the variability of these data, researchers have been studying A. *tuberculata* in retrievable mesh bags. The bags contain the worms, yet allow near-natural environmental conditions and permit entrance of ambient ELF EM fields. Worms were collected from both the treatment site (Treatment provenance) as well as from a similar deciduous forest site removed from the influence of the ELF transmitter (Fire Tower provenance). Worms from each provenance were then partitioned between treatment and control sites. Treatment-provenance worms were emplaced in 1991 and overwintered for observation in 1992. Fire Tower-provenance worms studied in 1991 were replaced with new individuals during April 1992. In 1992 all bags were watered in such a manner as to prevent moisture stress.

The plot of mean proportion of adult worms of Fire Tower provenance in the clitellate state showed a sigmoid increase, starting with iowest values in May 1992, peaking in August, and then gradually declining over the next two months (Figure 21). The percent clitellate was slightly higher at the treatment site; however, there were no statistically significant differences between sites. Although more "bell-shaped," the curve of the mean number of cocoons showed the same pattern, and peaked at the same time as the proportion of adults in the clitellate state. The mean number of cocoons present was significantly greater at the treatment site in July, August, and September. Based on numbers or mass, the mean cocoon production was significantly greater at the treatment site. There were no significant site differences in the average mass of individual cocoons, indicating that there were no EM effects on cocoon physiology.

Treatment-provenance worms held at the treatment site during 1992 showed a bell-shaped curve for the proportion of adults becoming clitellate, with the peak percentages occurring in August. Worms held at the control site had an irregular seasonal pattern, showing relatively high percentages at the beginning of the season, with a precipitous drop occurring after July. ANOVA showed this change and



FIGURE 21. MEAN NUMBERS OF COCOONS AND PERCENT ADULT IN CLITELLATE STATE FOR APORECTODEA TUBERCULATA.

site differences to be statistically significant. Peak values were lower than those exhibited by Fire Towerprovenance worms at corresponding sites.

The plotted abundance of cocoons from Treatment-provenance worms was also bell-shaped. Peak values occurred in June at the control site and in July at the treatment site; both peaks occurred one month earlier than maximum proportions of clitellates. Date-specific statistical analyses showed that site abundances differed significantly on three dates: In June, worms at the control site outproduced those at the treatment site, while the reverse was true in September and October. Cocoon production rates by Treatment-provenance worms showed both significant and nonsignificant site differences, depending on the method used to calculate the rates. In all cases, time-site interactions were significant, supporting other evidence of a change in the intersite relationship during the 1992 season.

There were no significant differences between sites for the cocoon mass of individual Treatmentprovenance worms, again indicating that there were no EM effects on cocoon physiology. Indeed, average cocoon mass strongly corresponded to average clitellate body mass. Accordingly, worms of Treatment provenance produced significantly smaller cocoons (22 to 23 mg) than those of Fire Tower provenance (25 mg).

Mesh bag and field population results for *A. tuberculata* are consistent with a pattern of increased cocoon production in response to ELF EM exposure. It follows that, as a result of higher cocoon production, adults leave the reproductive pool earlier. Investigators further hypothesize that the effects

on cocoon production, if valid, are immediate and long-lasting, but that there is a time delay for a secondary effect on the proportion of adults in the reproductive state. Similar effects on other worm species have not been detected. Overall conclusions based on these results are summarized in Table 16.

Parameter		ELF EM Effect	
	A. tuberculata	D. octaedra	L. rubellus
Distribution	None	None	None
Adult mass	None	-	
Cocoon mass	None		
Clitellate percent	Possible	-	
Clitellate density	Possible	None	—
Cocoon production	Possible	-	-
Cocoon density	Possible	None	-
Reproduction	None		None

TABLE 16. POPULATION CHARACTERISTICS OF EARTHWORMS

Litter inputs and Decomposition. This project element provides an estimate of the productivity of the dominant flora and overall functioning of soil biota involved in organic matter breakdown and nutrient release. In the latter case, MSU investigators have shown that initial rates of leaf decomposition were strongly related to the biomass of several worm species present at the study sites (Figure 22). Thus, this system-level response complements the faunal parameters under investigation, and, if seen at the populational and organismal levels of organization, would provide a context to evaluate overall effects.

Litter inputs were determined by collection of leaves in litter traps located at each site. Traps were emptied weekly during the time of greatest  $k \leq 4$  fall, and monthly at other times. Samples were sorted by category, then oven-dried, and weighed. Total litter inputs (g/m<sup>2</sup>) during 1992 were similar to those of previous years at both treatment and control sites. No EM effects from a fully operational antenna were indicated by BACI analysis. Litter input values at both sites continue to be consistent with data reported for forests at similar latitudes.

In the fall, preweighed samples of dried leaves were placed in mesh netting on the soil surface at both study sites. Throughout the following year, subsamples were periodically retrieved, dried, and reweighed. Estimates of decay rates were then obtained from mass loss and the period spent at the study site. At the time of reporting, several 1992 leaf samples remained unprocessed.

No significant differences in leaf decomposition rates before (1984-1988) or after (1989-1991) the NRTF-Republic became fully operational were detected using BACI statistical techniques. Preliminary

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FIGURE 22. LEAF LITTER TURNOVER RATES RELATIVE TO LITTER-DWELLING WORM SPECIES.

examination of available 1992 data did not indicate ELF EM effects. Investigator conclusions are presented in Table 17.

DECOMPOSITION (MSU)			
ELF EM Effect			
None			

# TABLE 17. LITTER PRODUCTION AND

### 3.5 Native Bees

Over 40 species of native bees occur on the Upper Peninsula of Michigan. Having coevolved with resident plants they serve as important, and perhaps unique, pollinators of flowering plants in the ELF Communications System area. Increased dispersal of bees, increased levels of their activity, lowered overwintering survival, and modified nest structure have been reported from exposure to the EM environment associated with electric transmission lines, as well as from fluctuations in the earth's magnetic field.

Accordingly, researchers from MSU have been monitoring the foraging activities, nest architecture, and mortality of native bees residing near the NRTF-Republic for possible EM effects from the operation of this transmitter. Since their initiating studies in 1982, investigators have focused data collection on two abundant species, *Megachile inermis* and *Megachile relativa*, as representatives of the native bee community.

Information on the characteristics of both species has been collected at two treatment sites located along the NS leg of the NRTF-Republic and two control sites over 10 miles from the antenna. Nesting activities were enumerated and timed by direct observation of foraging behaviors, while data on nest architecture and mortality were collected using trap nest techniques.

Nest trapping involved setting predrilled blocks of wood on shelved hutches and allowing bees to construct nests within the bore. Completed nests were allowed to overwinter at study sites. During the spring, the nests were split open and data on nest architecture was recorded. Completed nests consist of a series of reproductive cells, nonreproductive spaces, and plugs (Figure 23).

Intact reproductive cells were removed from the nests, placed in individual plastic tubes, and then kept outdoors at ambient temperature until emergence of adults. 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 to determine the condition and stage of the bee.

<u>Foraging Activity</u>. Disorientation and agitation have been reported for honeybees foraging in the relatively high intensity EM environment found near electric transmission lines. To examine for possible similar behaviors from exposure to EM fields generated by the ELF System, researchers monitored the foraging activities of native bees residing at study sites.

From 1983 through 1986, an extensive effort was made to observe, record, and determine the activity patterns of various species of native bees. The duration of trips for leaves to cap reproductive cells was short and less variable than other foraging behaviors. Because *M. inermis* was easily identified and active, its leaf foraging behavior was chosen as the best indicator of possible bee disorientation or agitation.



FIGURE 23. CUTAWAY VIEW OF COMPLETED MEGACHILE INERMIS AND M. RELATIVA NESTS.

Analyses using the general linear modeling (GLM) procedure showed significant differences between sites and between years (1987-1991). The duration of collecting trips has generally been longer at treatment sites than at controls throughout the study period (Figure 24). As the GLM did not demonstrate a significant site-year interaction, no change in the intersite relationship occurred after the antenna



FIGURE 24. MEAN DURATION OF LEAF FORAGING TRIPS FOR MEGACHILE INERMIS.

became fully operational. Researchers conclude that the data do not demonstrate an ELF EM effect on bee foraging.

<u>Nest Architecture</u>. When honeybees were exposed to EM fields produced by high-voltage transmission lines, they altered the hive's architecture by producing markedly fewer cells and increasing the amount of propolis at the entrance. If bees are disoriented because of EM fields produced by the ELF System while foraging, they may gather resources more slowly and an altered nest architecture may also result.

To examine for these possibilities, researchers are monitoring the size, number, linings, plugs, and orientation preferences for nests constructed by native bees. To date, nest architecture data from *M. relativa* nests constructed in 1983 and *M. relativa* and *M. inermis* nests constructed during 1985-1991 have been statistically analyzed.

GLM of 1985-1991 data showed no significant differences between sites or site-year interactions in the length of reproductive cells produced by *M. inermis*. Similar analysis for *M. relativa*, however, showed significant differences between sites, site-year interactions, and changes at the antenna site. These results indicate an increase in mean cell length at the treatment site compared to changes at the control site. The increase appears to be related to EM exposure, but the relative magnitude of the change is quite small (0.1 mm). The power of the GLM was such that there is as much as a 39 percent chance that the interaction was declared significant when in fact it was not.

The number of reproductive cell\_ per complete nest ranged from 1 to 12 for *M. relativa*. A categorical data modeling (CATMOD) procedure found significant differences between sites and years, but no significant site-year interactions or changes between preoperational and operational periods at the treatment site. These results indicate that none of the differences can be attributed to a fully operational antenna. The number of cells in a complete nest of *M. inermis* ranged from 1 to 8. In all years, the treatment sites had more cells per nest than nests at the control sites. No significant site differences occurred before or after full antenna operation, again indicating that site differences were not related to ELF EM exposure.

GLM analysis did not demonstrate any ELF EM effects on the number of leaves per reproductive cells made by *M. relativa*. The leaves per reproductive cell of *M. inermis*, however, appear to have increased in 1990 and 1991 compared with earlier years. Control cells increased by about 0.5 leaf per cell, while treatment cells increased about 1.4 leaves per cell. The GLM results suggest that reproductive cells constructed at the control sites intrinsically had more leaves than nests at the treatment sites and that *M. inermis* padded its cells with an extra leaf in the presence of ELF EM fields.

GLM of nest plug length for *M. inermis* confirmed that the plugs were longer at control sites, and have been so since initiating study on this parameter in 1985. The analysis also showed no significant

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temporal change in plug length at the treatment sites. Therefore, there does not appear to be any influence of ELF EM fields on nest plugs for this species.

In addition, bee preferences for orienting their nests were analyzed using a log-likelihood ratio contingency test. Only data for *M. relative* were analyzed, since nest numbers for *M. inermis* at control sites were very low. Results indicate a consistent preference for nest orientations over the years but the preferred direction was different between the sets deployed at a given site. Investigators conclude that orientation preferences were probably due to shading or proximity to resources.

<u>Mortality</u>. High-voltage transmission lines have been reported to lower the overwintering survival of honeybee colonies. In order to monitor for a possible similar effect from fields produced by the ELF System, researchers are comparing the mortality of native bees at treatment and control sites.

Prior to emergence of adults from reproductive cells, native bees are subject to mortality during any of several developmental stages (egg, larva, prepupa, pupa, or adult). Failure to emerge was used as an indication of morbidity. Prewinter mortality was related to the egg and larval stages, and overwinter mortality to the prepupal and later stages. Initial studies by the MSU investigators showed that prewinter mortality was primarily related to non-EM environmental conditions. For this reason and because the prepupal stage receives the longest EM exposure, overwintering mortality was selected for monitoring.

One confounding factor in determining overwinter mortality for native bees was the inability of investigators to distinguish the prepupa of *M. relativa* and *M. inermis* from that of a parasitic native bee, *Celioxys*. Since both host and parasite receive the same EM exposure, analyses were performed on their combined mortality data. Mortality was analyzed both as a percentage of reproductive cells and as a percentage of nests with at least one mortality. Data for nests constructed during full operation of the NRTF-Republic (1989-1991) and two prior years (1987-1988) were analyzed using CATMOD procedures.

*M. relativa* (plus parasite) overwinter mortality was not significantly affected by ELF EM exposures. However, the mortality within *M. inermis* nests and cells (plus parasite) was significantly greater at treatment sites when the antenna became fully operational. To confirm this finding, some nests constructed at a treatment site during 1990 and 1991 were transferred to a control site to overwinter. Transferred nests had a mortality similar to that experienced by nests constructed and overwintered at the control site (Figure 25). CATMOD procedures indicated that the overwintering site, but not the construction site, was the significant source of mortality for *M. inermis* plus its parasite, *Celioxys*.

<u>Summary</u>. Analysis of data collected through: 1991, show possible effects to the nest architecture of both study species (Table 18). These putative effects are small and, even if existent in other species, are not likely to adversely affect the bee community in the ELF System area. However, analysis of data collected through 1992 indicates increased mortality to one of two species because of EM exposure. Unlike the foraging and architecture results, an increased overwintering mortality could indicate adverse





	ELF EM Effect		
Parameter	M. relativa	M. inermis	
Foraging	-	None	
Nest architecture			
Ceil length	Possible	None	
Cell number	None	None	
Leaves per cell	None	Possible	
Plugs		None	
Orientation	None	-	
Mortality	None	Possible	

## TABLE 18. NATIVE BEE BEHAVIOR AND MORTALITY

effects to the bee community and the plants that they pollinate. If the pattern of greater mortality at the treatment sites persists in data collected during 1993, researchers will conclude a significant ELF EM effect.

Data collection for bee foraging and nest architecture elements was completed in 1991 and 1992, respectively. Data collection on mortality will be completed in 1993 when 1992 constructed nests are opened and examined. During 1993, investigators will incorporate foraging data collected prior to 1987

using GLM. Meteorological factors will be included as covariates. Architectural data on nests constructed in 1991 (opened in 1992) and mortality data for nests opened in 1993 will be added to the existing data base. These data will then be reanalyzed by the same statistical procedures summarized in this report.

#### 3.6 Small Mammals and Nesting Birds

Mammals and birds play important ecological roles as consumers in the ELF System area. These small vertebrates are also of intrinsic interest to local residents and can serve as indicators of possible adverse effects to other vertebrates, most importantly humans. The results of some studies performed at electric power and pulsed ELF frequencies have implied EM effects to laboratory vertebrates. To determine if operation of the ELF System affects resident vertebrates, researchers from MSU have been monitoring important biological and ecological characteristics of mammals and birds since 1982.

The project uses five treatment sites in, or immediately adjacent to, the NRTF-Republic ROW (NS leg) and four control sites with habitats similar to the treatment sites. Control sites were located 12-18 miles west of the antenna and have cleared areas (sham ROWs) that were treated (plantings, brushing, etc.) the same as the antenna ROW. Investigators consider 1986 through 1989 as preoperational years, and 1990 through 1992 as operational years.

Organismal aspects—including reproductive, developmental, and physiological characteristics—were examined for representative vertebrate species. Species studied were the deermouse, chipmunk, and tree swallow. The black-capped chickadee, a permanent resident on the Upper Peninsula, was used for physiological studies. Ecological aspects of the mammalian community were monitored until 1988 when these studies were discontinued due to highly variable results. Ecological characteristics of the bird community in the ELF System area have been monitored since 1984 by ornithologists from the University of Minnesota-Duluth (see Section 3.7).

<u>Embryonic Development</u>. Prenatal developmental stages have been shown to be particularly sensitive to many types of environmental perturbations. Although different from the fields produced by the ELF System, some EM fields have been reported to have a direct effect on embryonic development. Should EM exposure affect parenting behavior, indirect effects on development may also be feasible. Based on these premises, the prenatal development of tree swallows nesting near the ELF System was monitored for possible EM effects.

Embryos of tree swallows were collected at treatment and control sites after four days of incubation. The embryos were dissected from the egg, preserved, and then examined microscopically. Each egg was coded so that the investigator who examined for abnormalities was unaware of the source site. The status of the following in each embryo was assessed: developmental stage, brain, eye, ear and branchial arches, heart, spinal cord and somites, limb buds, extraembryonic membranes, and flexion and rotation of the embryo.

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In 1992, 29 of the 151 embryos collected were abnormal. Of the embryos collected, 15 were from control sites and 14 from treatment sites. As in the past, there was no significant difference between sites. Throughout the study, the incidence of abnormalities occurring in the ELF System area has remained consistent with the proportion of swallow hatch failures reported in ornithological publications (i.e., 15-20 percent).

Since avian embryos must develop in a closed system, the resources allocated to each offspring during oogenesis could have a marked influence in determining chick survival. To determine whether operation of the ELF System adversely affects the amount of nutrient deposited, each egg was weighed and measured at the time of collection. ANOVA showed significant differences between years, but no difference between sites or between periods before and after full-power antenna operation. Researchers conclude no ELF EM effects on the weight or volume of swallow eggs.

<u>Fecundity, Growth, and Maturation: Tree Swallows</u>. Effects of EM fields at ELF frequencies have been reported elsewhere for reproductive physiology, postnatal growth, and behavior of vertebrates. Accordingly, MSU investigators have monitored the fecundity, postnatal growth, and mortality of tree swallows for possible direct (EM) or indirect (parental behavior) effects from operation of the NRTF-Republic.

Studies were carried out in clearings where arrays of nest boxes had been erected. Active nests were checked daily or every other day to determine the dates that eggs were laid, the number of eggs, hatching dates, and mortality. The growth and development of the nestlings were then monitored until all of the young fledged. To facilitate the organization of study elements, MSU researches partitioned events into an incubation phase (which would include embryological events) and a postnatal, nestling phase. The former represents the time from egg laying to hatching, the latter from hatching to fledging of the young.

All data (1985-1992) on clutch size and young hatched per nest were examined using nested ANOVA. This statistical procedure found no significant differences between treatment and control sites, differences between preoperational (1985-1989) and fully operational periods (1990-1992), or any interaction between sites and operational status. Significant differences occurred between individual years, which investigators attributed to annual variations in weather. Analysis of 1992 data using chi-square procedures showed no significant differences between sites in incubation phase mortality. Investigators are 70 percent certain that they can detect differences of more than 7 percent as significant for clutch size and hatch success.

During mid-June 1992, study sites experienced a prolonged period of inclement weather during which several nights had freezing temperatures. This occurred at a time when hatching had already

occurred for most nests and resulted in high fledgling mortality (Figure 26). As a result, no 1992 data were available for several nestling growth parameters.



FIGURE 26. FLEDGING SUCCESS FOR DISPLACED TREE SWALLOWS.

To examine nestling growth rates, periodically measured values were fit to models. Body weight, tarsus length, and ulna length data were fit to logistic models, whereas wing length data were fit to an exponential model. The models produced parameters (e.g., rate constants or rate at the inflection point) that were then examined along with other nonrate parameters (maximum value, age at maximum value) by nested ANOVA.

Statistical results were generally consistent among multiple parameters for a given growth characteristic. Nested ANOVA showed no significant differences either between sites or between pre- and full-operational periods for all nestling growth parameters. There were significant differences, however, between years and between nests. Some significant interactions between sites and operational periods occurred. Minimum changes in the mean that can be detected with a certainty of 70 percent ranged 8-61 percent for growth constants and 20-95 percent for inflection points. Minimum detectable differences for other parameters await confirmation of statistical procedures.

For further examining significant differences, ANCOVA on growth data was also attempted using insect biomass as a covariate. Significant interactions arose, however, between the main factors (sites, years) and the covariate, and this procedure could not be used. By another approach, randomly selected nestlings were transferred within and between sites during 1990 and 1991. Maximal weight, tarsal length, and ulnar length were then examined by ANOVA. No effects attributable to EM exposure, or transfer, were found.

Data on developmental landmarks (bird age at eye opening and feather eruption) were not available for 1992 because of the very high nesting mortality experienced at all study sites. Nested ANOVA of 1986-1991 data demonstrated significant differences between nests, between years, and site-year interaction. These results were attributed to temporal and spatial disparities in meteorological factors. No significant differences between sites or operational periods were found for these landmarks. To assure 70 percent confidence, differences in main factors would need be greater than 25 percent for eye opening and 45 percent for feather eruption.

The number of fledged swallows per nest was examined by ANOVA. No significant differences between treatment and control sites or between preoperational and operational years, nor any interactions between sites and operational status, were demonstrated. There were significant differences between years (1985-1992) which investigators attributed to annual variations in weather (Figure 26). Chi-square comparison of sites for nestling phase mortality and overall nest mortality during 1992 showed significantly greater nesting mortality on treatment sites. Investigators are 70 percent confident of detecting an 18 percent or greater difference between sites for the success of fledglings.

<u>Growth and Maturation: Deermice</u>. The growth and development of deermice were monitored for the same reasons given for the above tree swallow studies.

Large, open enclosures were used to restrict the movements of deermice during studies of postnatal growth and development. The deermice to be studied were captured in mixed deciduous forests near the enclosure sites. The animals were paired, and when the female was pregnant, she was transferred to the large enclosure to give birth and rear the young to weaning. Observations were then made while the young were located in a nest within the enclosure.

Growth studies to date have shown that growth curves of temporal change in the body mass of nestlings are different between litters. Growth rates, therefore, have been estimated using growth constants derived from linear regression analysis of each individual at the time of weaning. Nested ANOVA showed significant differences between mothers and years (1986-1991), but none between sites or operational periods. Nested ANOVA of the developmental markers, age at eye opening and age at incisor eruption, gave the same results as for growth.

Investigators are not confident that required data collection protocols for mammals (i.e., observations every other day) allow adequate detection of subtle differences, if EM effects do indeed occur. They are 70 percent confident that they can perceive the following differences in main factors as significant: >108 percent for eye opening age, >125 percent for gr, wth, and >167 percent for incisor eruption.

Homing Studies. Animals are able to find food and escape predators more effectively in their home range. Any disturbance that affects orientation could affect an animal's ability to return to, or use, a home range and thereby decrease the probability of survival. Since orientation has been reported to

be altered by ELF EM exposure, the ability return to home range has been monitored for vertebrates in the ELF System area.

The homing of tree swallows, deermice, and chipmunks after displacement was examined from 1986 to 1992. The homing parameters were the likelihood to return (number of displaced individuals that return home) and, with swallows, 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. The direction of the release points from the nest sites requires those birds returning to treatment sites to cross both EW antenna elements of the NRTF-Republic (Figure 27). Birds taken from a single control site are displaced at an angle and distance similar to that 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.



FIGURE 27. RETURN ROUTES FOR TREE SWALLOWS.

Inclement weather and the poor physical conditions of swallows prevented MSU investigators from collecting homing data on tree swallows during 1992. Overall results for other years of study (1986-1991) demonstrate that birds captured from test plots are more likely to return (95 percent) than those captured at the control site (78 percent). Chi-square tests indicate that the likelihood to return was significantly greater for birds captured on treatment sites during 1987, 1990, and 1991. There were no significant differences between sites for the other years.

Mean return speed has been consistently faster for birds captured at treatment sites than that of birds captured at control sites (Figure 28). ANOVA demonstrated significant differences between capture sites and years, but no significant interaction between capture site and full operation of the antenna. These results and other experiments (tests for release point effects) demonstrate that operation of the NRTF-Republic has had no effect on return of displaced birds. Investigators conclude that the return characteristics of swallows are related either to the capture site or to some intrinsic factor related to the return path.



FIGURE 28. RETURN SPEEDS FOR DISPLACED TREE SWALLOWS.

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, whereas 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 next five days.

Since there was no significant difference between west or south displaced animals in their likelihood to return, all data for each site were pooled for intersite comparisons. Analyses of all data collected during the 1986-1992 period showed no significant differences between sites in the likelihood of displaced chipmunks returning to their capture locale. During 1989 the likelihood for deermice to return was significantly greater at the treatment site, and during 1990 it was significantly greater at the control site. There were no significant differences between sites for other years of deermouse studies.

<u>Physiology: Peak Aerobic Metabolism</u>. It has been speculated that EM fields adversely affect organisms by acting synergistically with other environmental factors. In the ELF System area, low temperatures make winter the most physiological stressful time of year. In order to examine for possible synergistic interactions, the aerobic metabolism of EM exposed vertebrates was monitored as an indicator of their physiological health during winter.

Black-capped chickadees and deermice were collected during the winter along the NRTF-Republic's ROW and at a control site. Animals to be tested were held at an outdoor facility with food and water provided ad libitum. Tests for peak metabolism were performed in an ethanol-cooled chamber using a version of the helium-oxygen method. Test equipment was located at a laboratory in Crystal Falls, Michigan; the holding facility was situated several miles south of the city. Prelimanary studies showed that the peak metabolic rates of deermice and chickadees did not change during three weeks of holding in the outdoor cages. After testing, animals were released at their collection site.

Data on the metabolism of chickadees were collected prior to (1986, 1987) and during full operation (1990, 1991) of the NRTF-Republic. Whole body peak metabolic rates (mL  $O_2/g$ -h) tended to be lower during the latter period than rates measured during the former period. ANOVA on logarithmic transformed data showed significant differences between sites and years, but no significant site-year interaction. Thus, even though the metabolic rates of animals differed between sites, the differences were not attributable to EM exposures from a fully operational NRTF-Republic.

Data on the metabolism of deermice were also collected prior to (1986, 1987) and during full operation (1990-1992) of the NRTF-Republic. Statistical comparisons of peak metabolic rates were carried out using ANCOVA. The logarithm of the peak metabolic rate was used as the dependent variable and the logarithm of body weight as the covariate. As for the chickadees, metabolic rates were significantly lower during 1990-1992 than rates measured during 1986, 1987. There were no significant differences between site rates nor any site-year interactions.

**Summary.** Based on statistical results, investigators conclude that the EM fields produced by the ELF System have no effects on small vertebrates (Table 19).

Additional data collection for many parameters will, however, be continued during 1993. As planned, investigators will be extending the scope of their statistical analyses. They anticipate using meteorological factors to further examine significant differences between years and significant interactions between site and operational periods. Investigators will also be scrutinizing all mortality data (1986-1993) using multidimensional contingency table analysis.

#### 3.7 Bird Species and Communities

Several bird species have been shown to sense and use geomagnetic fields to orient themselves. Thus, it has been suggested that birds may also be able to sense man-made EM fields and be attracted,
	ELF EM Effect	
Parameter	Birds	Mammals
Prenatal growth & development		
Egg weight, volume	None	-
Clutch size	None	
Embryology	None	-
Hatch success	None	
Incubation phase mortality	None	-
Postnatal growth & development		
Body weight	None	-
Tarsal length	None	
Uina length	None	-
Wing length	None	
Eye opening age	None	None
Feather eruption age	None	
Incisor eruption age	-	None
Nestling phase mortality	None	-
Metabolism	None	None
Homing	None	None

#### TABLE 19. SMALL VERTEBRATE DEVELOPMENT, PHYSIOLOGY, AND BEHAVIOR

repelled, or otherwise affected by such sources. In order to determine if the EM fields produced by the ELF Communications System cause adverse effects to the bird community, this project element monitored birds living in, and migrating through, areas near the Navy transmitters in Wisconsin and Michigan.

A line-transect method has been used to census the bird community in the ELF System area. The method involves two ornithologists simultaneously walking along randomly assigned treatment and control transects to determine bird species and numbers from sightings and songs. Since 1986, the identification and enumeration of bird species has been performed during each of five periods of the year: spring migration (May), early breeding (June), late breeding (July), early fall migration (August), and fall migration (September). In Wisconsin, studies were completed after the 1989 census; however, data collection continues in Michigan.

The following parameters were derived from the census data:

- total number of individuals (abundance)
- number of species (richness)
- number of individuals per common species
- number of individuals per guild (e.g., foraging and habitat types)

Differences between parameter values at treatment and control transects for any given year were examined using one-way ANOVA or a Kruskal-Wallis test. In 1992, differences between years were examined using a repeated-measures ANOVA procedure. In previous years, these differences were examined using two-way ANOVA and t-tests of preoperational/operational means.

The following subsections report on results along Michigan transects only; however, the conclusions rely on all data collected, including data collected in Wisconsin.

<u>Abundance and Richness</u>. The seasonal pattern for abundance and richness has generally been that more numbers of birds and species were observed during the breeding season than were observed during either the spring or fall migration periods (Figure 29). This pattern has remained the same over the preoperational, intermittently operational, and fully operational periods of the NRTF-Republic.

In June 1992, the abundance and species richness of birds was significantly greater on control transects than on treatment transects; in July, bird abundance was significantly greater on treatment



FIGURE 29. MEAN NUMBER OF BIRD SPECIES AND NUMBERS ON STUDY SITES.

transects. There were no other significant differences between treatment and control transects during the 1992 season.

Both species richness and abundance were significantly different between some years (during 1986-1992) for all months throughout the migration and breeding seasons. The species richness of birds breeding (June) in the ELF System area has changed little over the course of these studies; however, the number of individuals breeding in the area declined from 1986 through 1990, and increased since then. This pattern of change in breeder abundance closely matches annual rainfall patterns in the NRTF-Republic area (i.e., less than normal 1986-1989, near normal 1990-1992).

There were no significant differences in abundance or species richness between preoperational (1986, 1987) and operational periods (1989-1992).

<u>Abundance of Common Species</u>. EM exposure may not influence all bird species in the same manner. If, as a result of such exposure, some species are more plentiful in control areas and other species are more plentiful in treatment areas, such differences may cancel, producing few significant results in total numbers of individuals or in the total number of species. Therefore, the abundance of individual species that are common along study transects was also examined.

The most prevalent species present on treatment and control transects varied among samplings. However, when all sampling periods during 1992 were considered, particularly abundant species included black-capped chickadee, red-breasted nuthatch, hermit thrush, red-eyed vireo, Nashville warbler, blackthroated green warbler, ovenbird, and white-throated sparrow. Generally, these have remained the most abundant species over the entire course of the study.

Fifteen of 189 possible intersite comparisons (8 percent) showed a significant difference in the abundance of common species during 1992. The abundance of common species was higher on control segments in 11 of the cases (73 percent). In 1992, the abundance of common species was greater on the treatment transects than on the control transects.

If EM fields affected bird distribution patterns, investigators would expect to detect a change in the patterns of abundance between treatment and control transects as the power of the transmitter was gradually increased. To investigate this possibility, the distribution of abundance for 40 common species was examined over the period 1986-1992 using repeated-measures ANOVA. Only three of the 40 species (7.5 percent) showed a significant difference in abundance between treatment and control transects.

Analysis of Guilds. Although transects were selected randomly, there are more lowland-coniferous and early successional habitats on treatment transects than on control transects, and control transects cross more upland-deciduous habitats. In monitoring for possible effects to birds from the ELF Communications System, it is important to determine whether significant findings are related to EM exposure or to habitat differences between treatment and control transects. Therefore, habitat-related differences, as well

as the possibility that effects found to be significant for some species also affect other species, are being addressed by guild analyses.

All species of birds found on study sites have been classified into guilds on several bases, including foraging location and preferred breeding habitat. The total number of individuals for each guild on treatment and control transect segments were then compared for the 1992 season. During 1992, only a few significant differences were noted when comparing the distribution of foraging (three of 25) or habitat (three of 30) guild members. The number of significant differences are few, and such differences could be expected to occur by chance alone.

<u>Conclusions</u>. Results to date indicate relatively few significant differences between sites in bird species and community parameters. Most of the significant differences show no pattern relatable to ELF EM exposures, and appear to be occurring as chance events. Species abundance and guild distributions show several consistent patterns; however, these are attributed to habitat differences between transects and to bird preferences for them. Trends in abundance on treatment and control transects have not been significantly altered since the antenna became fully operational in 1989. Investigator conclusions are presented in Table 20.

AND MIGRATING BIRDS	
Parameter	ELF EM Effect
Total abundance	None
Species richness	None
Abundance of (40+) common species	None
Abundance within (4) guild types	None

TABLE 20. COMMUNITY CHARACTERISTICS OF RESIDENT AND MIGRATING BIRDS

Investigators conclude that there is no evidence of effects on bird species from either intermittent or full operation of the ELF Communications System in Wisconsin or Michigan.

# 3.8 Aquatic Biota

Aquatic biota, particularly fish, have been reported to use or react to weak EM fields. Therefore, a riverine ecosystem is being monitored for effects from exposure to the low-intensity EM fields produced by the ELF Communications System. Populational aspects as well as the functional and structural components of three major aquatic communities (periphyton, aquatic insects, and fish) have been monitored by researchers from MSU.

All studies have been performed on similar (fourth-order) sections of the Ford River in Dickinson County. Treatment sites are located adjacent to the NS leg of the NRTF-Republic, while control sites are located more than 10 km from the antenna. At each site, ambient environmental factors are monitored. Statistical analyses of natural ambient factors such as discharge, temperature, solar radiation, and nutrients show that treatment and control sites are well matched.

## 3.8.1 Periphyton

Periphyton are a community of microscopic plants and animals associated with the surfaces of submerged objects. Unlike organisms suspended in the water column, the structural and functional aspects of the periphyton community at a given location are governed by conditions at that point. Because they are important primary producers and show responses immediately at the source of a perturbation, periphyton were used to assess for possible changes due to the operation of the ELF Communications System.

The periphyton community is dominated by diatoms. Diatoms were emphasized in the monitoring of structural aspects of the periphyton; however, functional aspects such as chlorophyll, biomass, photosynthesis, and respiration were determined for the entire community (i.e., diatoms, plants other than diatoms, and microscopic animals). Quantitative determinations were made by collecting periphyton that had colonized glass slides of a known surface area. Preliminary studies in the Ford River have shown that the periphyton established on these artificial substrates were representative of the periphyton community found on natural substrates.

Statistical comparisons between sites used the paired t-test; however, BACI techniques were emphasized in most analyses. The BACI technique compared the mean of the "before" differences between control and impact (treatment) sites to the mean of the "after" differences between sites by using an unpaired t-test. Investigators consider observations made between June 1983 and June 1986 as preoperational data; between July 1986 and September 1989 as intermittent data; and between October 1989 and September 1992 as operational data. Randomized intervention analysis (RIA) was used when BACI techniques could not be used.

In order to minimize errors associated with a single method approach, investigators collected multiple independent sets of data and analyzed them using several statistical techniques. In addition, researchers calculated a correlation matrix for all ambient and biological/ecological variables.

Microscopic examination of glass slides emplaced at study sites for 28 days was used to identify and enumerate colonizing diatoms and to determine the cell volume of the dominant species. Cell volume indicated the physiological state of the species and, in conjunction with density, provided an estimate of the biovolume or biomass of the diatom community at each study site. The slide data also made possible

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the detection of possible shifts in the makeup of the community through calculation of evenness and diversity indices.

Individual cell volumes of dominant diatom species showed no significant differences between sites or between preoperational and operational periods. During the preoperational period, there were no differences between sites in diatom cell density; subsequently, however, site differences became significant. These results are supported by RIA and BACI analyses, which show significant differences in "before" and "after" comparisons. Derived estimates of biovolume gave mixed statistical results.

Diatom species diversity and evenness showed no significant differences during the preoperational period of the Michigan antenna, but these prarmeters were found to differ between sites after the antenna became fully operational. Other statistical analyses support a change in species evenness between preoperational and operational periods, but did not indicate a change in diversity indices. Investigator conclusions are presented in Table 21.

Parameter	ELF EM Effect
Cell volume	None
Density	Possible
Biovolume	None
Species evenness	Possible
Species diversity	None

# TABLE 21. STRUCTURAL ASPECTS OF THE PERIPHYTON COMMUNITY IN THE FORD RIVER

By themselves, the numbers and types of diatoms did not provide a complete characterization of the periphyton community. Because of this, investigators also monitored parameters that characterize the functioning of the entire community (e.g., photosynthesis, organic matter).

Microscope slides were emplaced in the Ford River for 14 days for determinations of accrual rates and for 28 days for determinations of standing crop estimates of chlorophyll a, phaeophytin a, and organic matter biomass. Fluorometric methods were used for analysis of chlorophyll and phaeophytin. Organic matter biomass was determined using changes in ash-free dry weight (AFDW) per unit area. Changes in dissolved oxygen concentrations in light/dark chambers with periphyton-covered substrates were used to estimate community productivity. Chlorophyll is the primary photosynthetic pigment used by all algae, including diatoms. In this project element, chlorophyll was used to determine the relative physiological state and biomass of the algal community at each study site.

Annual patterns in the accrual and quantity of chlorophyll in the Ford River varied between sites and years. Generally, the annual seasonal pattern was one of winter lows, with a peak value occurring in July or August. Year-to-year variability in standing crop and accrual rates was attributed to the presence (or absence) of secondary peaks occurring in the spring or fall and/or the magnitude of the summer peak. Secondary peaks have been occurring at both sites since 1986; these are thought to be related to low stream flows and relatively warm temperatures.

Prior to energization of the NRTF-Republic, chlorophyll was more plentiful at the control site than at the treatment site. Since the antenna became fully operational, the rate of accrual and total amount of chlorophyll have become greater at the treatment site. A significant change in the intersite relationship was also indicated by RIA and BACI analyses.

Organic matter standing crop (the biomass of the entire periphyton community) and organic matter accrual rates also changed between preoperational and operational periods. Paired t-tests examining site differences showed no significant differences between sites during the preoperational period; but after the antenna became fully operational, the treatment site tended to have higher organic biomass than the control site. RIA and BACI analyses supported the hypothesis that a significant change occurred in the intersite distribution of biomass after the antenna became fully operational.

There were no significant differences in gross primary production rates between treatment and control sites in 1992. However, when data were pooled over the entire period that the antenna was fully operational, t-tests showed a significant difference in production rates between sites. There were no significant differences between sites during the preoperational period. Other statistical tests failed to show a change in the site relationships for pooled "before" and "after" data. Investigator conclusions are presented in Table 22.

Multiple statistical analyses of several productivity parameters indicate a change in intersite periphyton biomass when preoperational data are compared with data collected after the antenna became fully operational.

These findings indicate a change in the overall distribution of biomass between sites, but the differences cannot decisively be attributed to ELF EM exposure. Results from aquatic insect studies (Section 3.8.2) indicate a possible change in the physical makeup at the treatment site over the last few years. Some of the significant differences may be attributable to partitioning of data between preoperational and operational periods. These possibilities are to be addressed, and additional data collected, during 1993.

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Parameter	ELF EM Effect	
Chlorophyll		
Standing crop	Possible	
Accrual rate	Possible	
AFDW		
Standing crop	Possible	
Accrual rate	Possible	
Gross productivity	None	

#### TABLE 22. FUNCTIONAL ASPECTS OF THE PERIPHYTON COMMUNITY IN THE FORD RIVER

Because of these issues, investigators conclude that results to date are only indicative of possible ELF EM effects on periphyton biomass.

#### 3.8.2 Aquatic Insects

As part of the integrated studies of the aquatic ecosystem, insects were monitored as representative of the primary and secondary consumer levels in the aquatic food chain. These studies examined the important functional insect groups, such as shredders, collectors, grazers, and predators. Both functional and structural aspects of the entire insect community organization were monitored. Community aspects included colonization patterns on benthic substrates and leaf litter; leaf litter processing; and structural descriptors of community change such as species richness, individual abundance, and species diversity.

As benthic insects are a major component of the primary and secondary trophic levels of the Ford River, their community structure and function have been monitored for possible effects from the operation of the ELF Communications System.

Sample baskets containing riverine substrates were placed at study sites for one-month periods at intervals during the spring, fall, and summer. The colonizing insects were separated from the substrates, and then 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 biomass for functional feeding groups were determined. For those insects with high numerical abundance, mean dry weight per individual was also computed.

Data for biotic parameters were sorted into three seasonal categories: spring (April, May), summer (June-August), and fall (September-December). All seasonal data (27 data sets) were first analyzed by

using two-way ANOVA, then multiple linear regressions were used to determine those ambient factors that were most correlated with the monitored biotic parameters. Preoperational-to-operational (1983-1986 to 1989-1992) comparisons were performed using RIA or BACI analyses.

ANOVA showed significant site-year differences, most of which were later related to river discharge and/or water temperature. None of the 21 data sets examined using BACI procedures showed significant differences between data collected during preoperational years and data collected during operational years. RIA of the remaining six data sets showed significant differences between preoperational and operational periods for numbers of individuals, chironomid dominance, and total insect biomass. The former two parameters were highly variable and were not analyzed further. Sequential multiple regression of total insect biomass showed that temporal differences were related primarily to differences in river discharge and not to ELF EM exposure.

Overall, investigators conclude that there was no statistically significant pattern of results indicating ELF EM effects on structural or functional aspects of the benthic insect community (see Tables 23 and 24).

Parameter	ELF EM Effect
Species diversity	None
Species evenness	None
Species richness	None
Number of individuals	None
Chironomid dominance	None

#### TABLE 23. STRUCTURAL ASPECTS OF THE BENTHIC INSECT COMMUNITY IN THE FORD RIVER

#### TABLE 24. FUNCTIONAL ASPECTS OF THE BENTHIC INSECT COMMUNITY IN THE FORD RIVER

Parameter	ELF EM Effect
Total mass	None
Chironomid mass	None
Collector-gatherer mass	None
Predator/prey ratio	None
Growth/recruitment	None

In headwaters such as exist in the ELF System area, only a small portion of the energy input to the river is provided by aquatic plants and algae. The input of leaves from riparian vegetation is also recessary to maintain the existing community structure. Consumers (mainly insects) process the leaf litter, and in doing so make their biomass available to insect and fish predators. A "leaf pack" bioassay technique was used to monitor for possible EM effects to this aspect of the aquatic community.

Tag alder leaves were collected from a grove adjacent to the Ford River. After leaf packs were weighed, they were tied to bricks and placed at riverine study sites. Prior to 1990, both fresh and autumnabscised alder leaves were used in the packs; since 1990, only fresh alder leaves have been used. Leaf packs were periodically retrieved and replaced over a four-month period. Insects colonizing the retrieved packs were identified and enumerated, and the loss in leaf mass was determined. These data were then used to characterize the structural aspects and functioning of the community.

Data collected over the period 1984-1992 were analyzed using ANOVA and ANCOVA, multiple regression, or t-tests. BACI tests could not be performed on these data, because only one value was generated per year.

Two-way ANOVA showed significant differences between years (1984-1991) for many structural parameters; however, there were few site differences. Except for taxon diversity, ANCOVA and other tests of the same data indicated neither site nor year differences, or have related such differences to temperature. Even though it has steadily decreased at both sites, diversity has remained significantly greater at the treatment site than at the control site over the term of the study. The principal investigator does not, however, attribute this site difference to ELF EM exposure (Table 25).

Parameter	ELF EM Effect
Taxon richness	None
Taxon evenness	None
Taxon diversity	None
Number of individuals	None
Chironomid dominance	None

# TABLE 25. STRUCTURAL ASPECTS OF THE LEAF-PROCESSING COMMUNITY IN THE FORD RIVER

Functional community parameters for organisms colonizing leaf packs were also examined. These parameters included total insect biomass (adjusted to leaf biomass), and the mean (dry) weight per

individual for three abundant insect species. ANCOVA failed to show significant differences in insect biomass for 21 of 24 growth comparisons. No significant differences in leaf processing rates between sites were found when each year from 1984 to 1992 was examined using Student's t-tests (Figure 30). Investigator conclusions based on these results are presented in Table 26.



FIGURE 30. ANNUAL RATES OF LEAF PROCESSING IN THE FORD RIVER.

# TABLE 26. FUNCTIONAL ASPECTS OF THE LEAF-PROCESSING COMMUNITY IN THE FORD RIVER

Parameter	ELF EM Effect
Leaf processing	None
Growth	None
Total insect mass	None

Statistical analyses showed many significant temporal and spatial differences in aquatic insect parameters. However, significant differences were attributed to river discharge and water temperatures in all but two comparisons. The two parameters showing significant differences between preoperational and operational periods were not further analyzed by sequential regression because of their high variability. Researchers conclude that there were no ELF EM effects to the aquatic insect community.

Over the period 1984-1992, data from treatment and control sites have become more similar (Figure 31). These changes appear to have occurred in response to changes in substrate particle size at the treatment site over the last two years. In 1993, investigators are to analyze substrate particle size



FIGURE 31. RELATIVE BIOMASS OF BENTHIC INSECTS IN THE FORD RIVER.

for comparison with similar data collected at the beginning of the study. Data collection protocols and statistical analyses during 1993 will continue as in previous years.

Other studies have suggested that high-intensity 60 Hz EM fields can increase dispersal and locomotor activity in exposed terrestrial insects. Accordingly, the movement patterns of aquatic insects were examined for possible effects from operation of the ELF Communications System. The movement patterns of displaced dragonfly naiads were monitored from 1985 through 1989; however, no significant effects were noted. Because this parameter proved to be statistically insensitive, study was concluded after the 1989 season. The results and findings have been published in the Journal of Freshwater Ecology.<sup>27</sup>

#### 3.8.3 Fish

Some species of fish have an ability to perceive weak EM fields. Because it has been shown that fish use this perceptive ability to orient themselves and to detect prey, structural aspects and movement characteristics of the mobile fish community are being monitored for possible effects from EM fields generated by the ELF Communications System. As tertiary consumers, fish can also indirectly reflect changes that are occurring at lower trophic levels. In this regard, investigators are also assessing the condition of select fish species.

The fish community was sampled by deploying two fyke nets (one facing upstream and the other downstream) at each of three sites. Nets were locateu at a downstream site (control site), near but downstream of the NS antenna crossing (treatment site), and 400 m upstream of the treatment site. Previously used net sites on the upper Ford River and Two Mile Creek were discontinued after use in 1991.

Netted fish were identified, counted, weighed, and measured for length. Common species had their fins clipped for identification. The fish were then released in the original direction of travel, i.e., either upstream or downstream of the capturing net. Community parameters determined from these samples were species composition and the abundance of common species by numbers and biomass. Several individual species were also examined for stress using derived age and growth data. Movement rates of common species were determined by the distance between nets and the period between captures.

Data were divided into three periods for statistical analyses: preoperational (1983-1985), intermittently operational (1986-1988), and fully operational (1989-1992). Analyses were performed using appropriate statistical techniques, including ANOVA, chi-square, Spearman rank correlation, and BACI analyses.

The number of fish species collected during 1992 was greater at the control site (17 species) than that at the treatment site (15 species). This intersite relationship in species richness has remained the same since studies were initiated in 1983. The slight difference that exists between sites results from the occasional catches of a few uncommon species at the control site. No significant differences between sites or operational periods were found by investigators for fish species diversity; however, diversity has steadily decreased over the term of the study.

Numerically and by biomass, the fish community at treatment and control sites has been dominated by the same five species since 1983 (Figure 32). These species are brook trout, burbot, common shiner, creek chub, and white sucker. Despite changes in abundance from year to year, there were no significant differences in catch by number or biomass, either between sites or among operational periods.

Most non-salmonid fish (burbot, shiner, chub, and sucker) have been recaptured at the sites where they were tagged. However, a number of each species has been recaptured at other sites, demonstrating their movement upstream and downstream under the antenna. The percent recapture at other than marking sites (1 to 2 percent) was the same during both preoperational and operational periods.

Condition factor analysis was performed using common shiner, creek chub, and white sucker data as indicators of possible stress to the fish community. Analysis of data collected since 1983 and pooled over both treatment and control sites showed that condition factors for common shiners and creek chubs were above those reported in the literature. Consistent with previous years, the condition of white suckers was more than 10 percent below the species means for populations reported in the literature. This below-normal condition existed prior to intermittent or full energization of the antenna.

**IITRI D06205-3** 



FIGURE 32. RELATIVE BIOMASS OF DOMINANT FISH SPECIES IN THE FORD RIVER.

An important sport fish, the brook trout, was also monitored for pattern, rate, and magnitude of movement as well as growth and condition.

The general pattern of trout migration has been upstream through the ELF System area to the confluence of the Ford River and Two Mile Creek. Virtually all trout migrate up Two Mile Creek. Upstream movement occurred with varied intensity in the late spring to early summer. No downstream movement from Two Mile Creek was observed for sampling periods lasting through November. The factors affecting distribution pattern and time of peak catches appear to be water temperature, stream velocity, and trout population size.

Brook trout movement across the NS antenna of the NRTF-Republic was monitored by radiotracking tagged fish as well as by the recapture of marked individuals. ANOVA did not show significant differences in average movement rates between preoperational, intermittently operational, or fully operational periods. Movement rates determined from recapture of trout in nets (2.9 km/day) compared favorably with rates determined from angler catches of tagged fish (2.3 km/day). There were no significant differences between preoperational and operational periods in the number of days between tagging and recapture at another site.

Age and growth analysis indicated that the brook trout in the Ford River exhibit average or better growth than that reported in the literature. Growth rates at both treatment and control sites during the operational period differed significantly from growth rates prior to or during intermittent operation of the

antenna. There were no significant differences between preoperational and intermittently operational periods.

Brook trout were also examined using relative weight condition factors. These factors declined from 1983 through 1986, increased from 1987 through 1990, and again have declined since 1990. Overall, trout from the Ford River were of average to below-average weight when compared to the calculated average reported in the literature. Regression analyses did not show significant differences between sites, and ANCOVA did not detect significant differences in condition between preoperational, intermittently operational, or fully operational periods.

Investigator conclusions are presented in Table 27.

Parameter	ELF EM Effect
Species richness	None
Species diversity	None
Number of individuals	None
Biomass	None
Growth	None
Condition	None
Movement	None

# TABLE 27. CHARACTERISTICS OF THE FISH COMMUNITY IN THE FORD RIVER

Since the initiation of studies in 1983, the richness and abundance of fish species have declined at both treatment and control sites. There were no statistically significant differences in fish diversity between sites. Slight differences between sites appear to be due to occasional catches of uncommon species in nets at the control site.

The condition and growth of fish in the Ford River were determined by combining length and weight data collected at treatment and control sites and comparing the values to those reported in the literature. Generally, the condition and growth of fish varied across species and years. No consistent pattern relatable to ELF EM exposure has been found.

All representative species have demonstrated both upstream and downstream movement beneath the ELF antenna. Low movement patterns from 1986 through 1990 corresponded to the presence of

beaver dams and low river flows experienced in those years. Dams were destroyed by high spring flows in the spring of 1991, and fish have been recaptured at higher rates since then.

Investigators conclude that there were no EM effects to the fish community from the operation of the Michigan ELF antenna.

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# APPENDIX A

# ECOLOGICAL MONITORING PROGRAM: LIST OF PUBLICATIONS/PRESENTATIONS, 1982-1993

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### Upland Flora (Michigan Technological University)

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