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Annual Research Progress Report No. 11

July 1976



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TABLE OF CONTENTS

	Page
I. SCIENTIFIC AND ENGINEERING INVESTIGATIONS	
A. Department of Aeronautics	1
1. Aerodynamic Characteristics of Glide Bomb Configurations	1
2. Glide Bomb Aerodynamics	1
3. Rolling Moment Induced by a Rotating Wing	2
4. Wind Tunnel Tests of a 0.2 Scale Maneuvering Air to Surface Submunition (MASS)	2
5. Flow Field Behind Aerodynamic Fences	3
6. Roll Reversal in Canard-Configured Missiles	4
7. The Flow Dynamics of Unsteady Separated Regions	4
8. An Undergraduate Propulsion Sequence.	5
9. Boundary Layers in Axially Symmetric Nozzle Flows	6
10. Jet Engine Particle Separators	6
B. Department of Astronautics and Computer Science	7
1. The Data Administrator's Handbook	7
2. The Effects of Response Time on User Performance and Satisfaction in a CAI Environment	8
3. Development of a 4-Way Analysis of Variance System.	9
4. ALGOL Procedures for Detection of Deadlocks among Parallel Processes	9
5. Visual Representation of Network Symmetry	10
6. Configurable Microprocessor Array	10

7.	Artificial Intelligence Research	11
C.	Department of Chemistry and Biological Sciences. . .	12
1.	Chemiluminescent Gas Phase Reactions	12
2.	Energetic Plasticizer and Binder Synthesis Via Organic Triflate Intermediates	13
3.	Pelletized Thermal Batteries	14
4.	Electrode and Electrolyte Physical and Electrochemical Measurements	15
5.	Aluminum Matrix-Trialuminum Nickelide Separation	17
6.	Detonation Property Prediction and Modeling. . .	18
7.	Simultaneous Monitoring of Climate and Air Pollutants at USAFA	19
8.	Lead as an Indicator of Environmental Quality in Airport Environs	19
9.	Lipids and Lipoproteins in USAF Academy Cadets .	20
10.	Determination of Parameters for a Model of Environmental Quality for USAF Installations . .	21
11.	An Evaluation of Current Capabilities in Genetics and Molecular Biology	22
12.	Disposal of Herbicide Orange by Soil Incorporation and Biodegradation	22
13.	Ecological Studies on a Herbicide-Equipment Test Area (TA C-52A), Eglin AFB Reservation, Florida	23
14.	Radiochemical Bioassay of TCDD Uptake in Plant Material	25
15.	Ultrastructural Studies of Liver Tissue from TCDD-Exposed Beach Mice (<i>Peromyscus polionotus</i>)	26
16.	Ultrastructural Evaluation of Tissues Removed from Animals Exposed to TCDD	27

D.	Department of Civil Engineering, Engineering Mechanics, and Materials	28
1.	Backing Strength of Masonry Shear Walls	28
2.	Validation of the Soil Stabilization Index System with Manual Development	28
3.	Polymer Tetherline Evaluation for Balloon Technology .	29
4.	Solar Energy	30
5.	Statics and Dynamics of Cables	30
6.	Sublimation of Basal Faces of Zinc Oxide Single Crystals Under Ultraviolet Illumination	31
7.	Finite Element Analysis of Single Span Cables	32
8.	Lateral Dynamics Optimization of A Conventional Railcar	33
E.	Department of Electrical Engineering	33
1.	Troposcatter Communications Simulator	33
2.	Micro-Computer Solutions to the Time Difference of Arrival Problem	34
3.	Study of Composite Materials in an Electromagnetic Environment	34
4.	USAFSA Solar Energy Program	35
5.	Space Test Program Experiment Prioritization	36
6.	Digital Communications Performance Monitor	37
7.	A Microprocessor Based Controller for an Isolation Pad	37
8.	Effects of Weightlessness on the Cardiovascular System	38
9.	Management Model of the Military Health Care System .	38
F.	Department of Mathematical Sciences	39

1.	Efficient Branch-and-Bound Algorithms for Permutation Problems	39
2.	Distributed Parameter Control.	39
3.	An Encke Method Applied to Element Equations	39
4.	Seismic Network Capability Model	40
5.	Detonation Property-Predictions and Modeling	40
G.	Department of Physics.	41
1.	Portable Solar Collector for Sea Survival.	41
2.	Transverse Electric Atmospheric Laser.	41
3.	Unstable Resonator Mirror Misalignment	42
4.	Spontaneous Emission Using Vector-Spherical Harmonics	42
5.	Single Particle Green's Functions.	42
6.	Liquid Metal Equation of State	43
7.	Lamb Shift in Hydrogen	44
8.	Deformable Mirror Control System Parameters.	44
9.	Analysis of Electronic Component Damage Due to EMP	44
10.	Diagnostic Technique for the AFWL SHIVA X-ray Simulator	45
11.	Visual Perception: Nature and Cause of the Horizontal Vertical Bias in the Human Visual System.	46
12.	Numerical Calculation of Explosive Behavior	47
13.	Aircraft Engine Oil Analysis by Neutron Activation.	47
II.	GENERAL RESEARCH IN THE HUMANITIES AND SOCIAL SCIENCES	
A.	Department of Aeronautics and Computer Science	49

1.	Computerized Scheduling of CCQ for the Cadet Wing . .	49
2.	A Layman's Interface to the AID-4 Statistical Computer Program	50
3.	Phase Testing for a First Course in Computer Science.	51
4.	Minimum Paced Self-Instruction in a Two-pass Course .	52
5.	The Computer Science Core Course Grade Score Data Base	52
6.	The Front-end Processing Programs of the Basic Cadet Training Tracking Program and BCT Standing System . .	53
7.	Analysis of the Military Order of Merit (MOM) System	53
8.	Statistical Analysis of the Military Order of Merit (MOM)	54
9.	ELSIE Data Reduction	54
B.	Department of Behavioral Sciences and Leadership	55
1.	The Validity of Various Measures in Predicting Pilot Training.	55
2.	Level of Alertness and the Perceptual Processing of Visually Displayed Information during Simulated Flying Missions.	56
3.	Behavioral and Physiological Correlates of Varying Noise Environments	57
4.	The Use of Biofeedback Techniques in Stress Management Training	58
C.	Department of Economics, Geography and Management	59
1.	Observable Public Good Preferences	59
2.	Centralized Planning.	60
3.	Hahn, Inc.	60
4.	Performance Appraisal: A View From Using Organizations	61
5.	The Technological Gatekeeper--A Boundary Role Communicator	61

6.	Capturing Judgment Policies in Performance Ratings.	62
7.	The Technological Gatekeeper.	63
8.	Tertiary Sedimentation into the Denver Basin, Colorado: A Model for Alluvial Plain Deposition	64
9.	Reconstruction After an Earthquake Disaster	64
10.	Kutahya Province, Turkey.	65
11.	Disaster and Change	65
12.	Inflation Models of Selected NATO Countries	66
13.	Fighter-Maker Base Selection Project	66
14.	An Algorithm for a Pattern Sensitive Data Storage System (PSDSS) (PhD dissertation)	66
15.	Federal Energy Administration Mapping Project	67
16.	Analysis of Hospital Overhead Costs	67
D.	Department of English and Fine Arts.	68
1.	Edward, Lord Herbert of Cherbury: A Preliminary Checklist	68
2.	The Donne-Herbert Family Relationship	68
3.	Radiographic Reproduction of Early American Watermarks	69
4.	The Air Force Academy and Total Environmental Education	69
5.	The Teaching of Old English Literature in Translation.	70
6.	A Descriptive Bibliography of Robert Penn Warren, 1917-72	71
7.	A Bicentennial Edition of Thomas Godfrey's "The Prince of Parthia, A Tragedy" (1765)	71
8.	Writing and the Composition Teacher	72
E.	Department of Foreign Languages.	72

1.	FRANCE - Land and People: A Handbook	72
2.	GERMANY - Land and People: A Handbook.	73
3.	Criterion-Referenced Foreign Language Testing	73
F.	Department of History	74
1.	A History of the United States Air Force.	74
2.	General Hoyt S. Vandenberg, Sr.	75
3.	USAF Oral History Program.	75
4.	The Harmon Memorial Lectures in Military History.	76
5.	The Military and Society: The Proceedings of the 1972 History Symposium.	76
6.	The Military History of the American Revolution: The Proceedings of the 1974 History Symposium	76
7.	The Marquess Wellesley in Irish and British Politics: 1781-1842	77
8.	Air Force Images of Research and Development and Their Reflections in Organizational Structure and Management Policies	77
9.	The Development of Aircraft and Doctrine in the Royal Flying Corps, 1912-1914	78
10.	Blacks in the Army Air Forces During World War II: The Problem of Race Relations	78
11.	U.S. Army Aviation 1963-Present	79
12.	Biographical Study of Sergei Mikhailovich Soloviev, 1820-1879	79
13.	Intruduction to the "Zapiski" of S. M. Soloviev	79
14.	S. M. Soloviev's Role in Russian Historiography	80
15.	Publication History of S. M. Soloviev's "Istoriia Rossii c drevneishikh vremen"	80
16.	The Creation of the GHQ Air Force	80

17.	The Divergence of Policy and Power: The American Military and Postwar Foreign Policy (1945-47)	80
18.	The Army and Its Air Corps	81
19.	U.S. Army Air Corps: 1926-1941.	81
20.	Nevada's Fighting Irishman: Senator Pat McCarran of Nevada	81
21.	On the Road Toward Termination: The Pyramid Lake Paiutes and the Indian Attorney Controversy.	82
22.	American Military Missions to Korea: 1882-1896.	82
23.	Studies in English on Modern Korean History, 1876-1910: A Critical Bibliographic Survey of Primary and Secondary Sources.	82
24.	Stalag Luft III: An American Experience in a World War II German Prisoner of War Camp	83
25.	Black Spies in America's Wars.	83
26.	Negro Migration to Colorado: 1870-1930.	84
G.	Department of Law	84
1.	Due Process Requirements in Cadet Disenrollment Proceedings	84
2.	Teaching by a USAF Academy Faculty Member During the Summer at a Civilian Educational Institution	84
3.	Legitimated Interposition and International Law	85
4.	Legality of Orders.	85
H.	Department of Political Science and Philosophy.	86
1.	A Comparison of Military Elite Role Perceptions in Indonesia and the Philippines	86
2.	Israel and the PLO: Persistence or Transformation.	86
3.	Insurgent Strategies: An evaluation of Four Approaches	87

4. Petroleum Security: the Role of the Military Instrument of Statecraft	87
5. Revolutionary Warfare in Oman: A Strategic Appraisal	87
6. An Analysis of Possible Policy Changes in Terms of Selected Defense Policy Issues	88
7. Decision-making in the Organizational-Bureaucratic Context	88
8. Final Report: Colloquium on NATO's Southern Flank. .	89
9. Japan: The 1975 Debate Over Ratification of the NPT.	89
10. The Ethics of Leadership.	90
11. Security Implications of Normalization of Relations Between the United States and Cuba	90
12. Chinese Military Power and Foreign Policy: The Case of the Paracel Islands	91
13. The Minorities and the Military in China.	91
14. Inner Mongolia: The Haos and Huais of Chinese Policy Toward the Mongols	91
15. Thailand: A Case Study in Counterinsurgency.	92
16. The Political Element in Military Expertise	92

III. RESEARCH AND ANALYSIS OF SPACE AND WEAPONS SYSTEMS

A. Department of Astronautics and Computer Science.	94
1. Air-to-Air Fire Control Research	94
a. Laboratory Test of the ASCOT Electro-Optical Tracker Aided by a Digital Estimator	94
b. Development of a Director Gunsight for Flight Test	95
c. Director Gunsight for an Advanced Fighter	95
d. Scoring Algorithm for Advanced Fighter Gunsights .	96

e.	Digital Simulation of One-on-One Air-to-Air Combat	96
f.	F-16 Gunsight Evaluation	97
g.	Gunsight Heads-Up Display for the Aerospace Simulator.	97
h.	Air-to-Air Combat Simulation Support to Tactical Air Command.	98
2.	Principles of Airborne Fire Control Text	98
3.	Independent Stability and Control Analysis for the Navigation Demonstration Satellite-1 of the Global Positioning System - (GPS) - NAVSTAR	99
4.	The Time Rate Changes due to Relativistic Effects on the Global Positioning System.	100
5.	GPS Satellite Selection Criteria	101
6.	GPS Deployment Using the Space Shuttle	101
B.	Department of Mathematical Sciences	101
1.	Strategic C ³ Study Group	101
2.	Effect of Transition on Aerodynamic Coefficients of Hypersonic Reentry Vehicles.	102
IV.	MANPOWER, PROCUREMENT, AND LOGISTICS STUDIES	
A.	Department of Astronautics and Computer Science.	103
1.	Automation of the Cadet Status Records	103
2.	Cadet Time Analysis System	103
3.	Analysis of the Military Order of Merit (MOM) System	104
B.	Department of Economics, Geography and Management	104
1.	Variable Obsolescence Rates.	104
2.	Procurement Productivity Indices	105

3.	General Recruit Quality and Its Relationship with Air Force Technical Training.	106
4.	A Conceptual Model for Evaluating Contractor Management During Source Selection.	106
5.	Proceedings of the Fourth Annual Department of Defense Procurement Research Symposium.	107
6.	USAF Procurement Quality.	107
7.	Improvement of Aeronautical Systems Division Inflation Forecasting Indices	108
8.	R&D Cost Expenditure Pattern Analysis	108
C.	Department of Mathematical Sciences	109
1.	Manpower Modeling in Support of CAREERS	109

correlation between elevated serum cholesterol and coronary disease, this finding was indeed encouraging. A positive correlation was also observed between periods of stress (academics, hell week, and Basic Cadet Training) and average cholesterol and uric acid levels. This finding may be useful in diagnosing emotional stress when the patient (subject) is not aware that he is under severe stress conditions. A third general finding was that there was an overall average weight increase in the subjects of this study. While this may not be unexpected, determinations of body fat revealed there was an unexpected decrease in this parameter. Apparently, this weight increase was due to an increase in muscle tissue rather than in fatty deposits. Overall, the data from this study tend to lend support to the current diet and physical conditioning programs at USAFA. Other findings will undoubtedly be forthcoming after all the data have been evaluated.

10. Determination of Parameters for a Model of Environmental Quality for USAF Installations

Principal Investigators: Captain Lawrence J. Biever and Alvin L. Young, and Major Manuel A. Thomas, Department of Chemistry and Biological Sciences.

Sponsored by Frank J. Seiler Research Laboratory (AFSC).

The major effort on this project this year has been to determine what effects non-potable sewage effluent have on ecosystems (e.g., parade fields and golf course) where it is used for irrigation. Data collected so far indicate that inclusions in the water are causing a unfavorable shift in species composition of grasses and are causing damage to tree species. The non-potable irrigation water includes high concentrations of phosphate, nitrate, chlorine, salt, and many heavy

metals, Soil analysis indicated progressive changes in soil chemical composition that may result in degradation of soil quality. Continuation of the program is being proposed to further delineate cause and effect relationships and to provide corrective measures.

11. An Evaluation of Current Capabilities in Genetics and Molecular Biology

Principal Investigators: Captain Lawrence J. Biever, Captain Alvin L. Young, Lieutenant Colonel Eugene L. Arnold, and Major William C. Wilson, Department of Chemistry and Biological Sciences.

Sponsored by the Defense Intelligence Agency.

This study has been initiated to determine the state-of-the-art of current biological technology. An evaluation of the results of the application of current biological research to agriculture, industry, medicine, and the military has been made. The study included an evaluation of the international political, economic, social, and military impact of technological developments in biology. ^{Genetic Engineering}

Data collected from current literature and contacts with researchers has been evaluated by DFCBS personnel. An interim report has been submitted. The final report will be submitted in June, 1976. A follow-on project has been proposed by the sponsor.

12. Disposal of Herbicide Orange by Soil Incorporation and Biodegradation

Principal Investigators: Captain Alvin L. Young, and Lieutenant Colonel Eugene L. Arnold, Department of Chemistry and Biological Sciences.

Sponsored by Air Force Logistics Command/DS.

In August 1972, a site for soil incorporation of 2,4-D and 2,4,5-T herbicides was selected on the Air Force Logistics Command Test Range Complex, Hill Air Force Base, Utah. The site was considered

remote and the land of low-use potential. It was characterized by sandy loam soil with a pH of 8.0 and an organic carbon content of 0.95 percent. The herbicide formulation contained equal amounts of the n-butyl esters of 2,4-D and 2,4,5-T and was applied by simulating sub-soil injection at rates as high as 4,000 lbs ai/A. The replicated plots were periodically sampled and the concentration of ester and acid forms of the herbicides was determined by gas-liquid chromatography. Levels of the toxic contaminant 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) were also measured in certain selected samples. Degradation of both ester and acid form of each herbicide was initially rapid (1st year) but tended to taper off in succeeding years. Strict exponential decay kinetics were not followed. Application rate had only minimal effect on the rate or pattern of degradation. The rate of disappearance of 2,4-D was slightly higher than that for 2,4,5-T. In those samples in which it was measured, significant degradation of TCDD was also noted. Soil penetration of the herbicides as either acid or ester forms was negligible.

Publications

Arnold, E. L., A. L. Young, and A. M. Wachinski. 1976. Three years of field studies on the soil persistence and movement of 2,4-D, 2,4,5-T and TCDD. Presentation to the Weed Science Society of America, 3 February 1976, Denver, Colorado. Abstract No. 206.

Arnold, E. L. and A. L. Young. 1976. A rapid gas chromatographic method for the determination of phenoxy herbicide residues from soil disposal plots. Journal of Agriculture and Food Chemistry. In Press.

13. Ecological Studies on a Herbicide-Equipment Test Area (TA C-52A), Eglin AFB Reservation, Florida

Principal Investigators: Captain Alvin L. Young, and Majors Charles E. Thalken and Lorris G. Cockerham, Department of Chemistry and Biological Sciences.

Sponsored by Air Force Logistics Command/DS.

During this past year species diversities and food chain studies were conducted in two aquatic ecosystems draining the unique one-square mile military test area (Test Area C-52A, Eglin AFB, FL) that received 160,948 lbs 2,4,5-T and 169,292 lbs 2,4-D herbicide during the period 1962-1970. Significant levels (10-710 parts per trillion, ppt) 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) were found within the top six inches of the test area soil. Erosion of soil occurred into a pond on the test area and into a stream immediately adjacent to the area. TCDD levels of 10-35 ppt were found in silt of the aquatic systems, but only at the point where eroded soil entered the water. Species diversity studies of the stream were conducted in 1969, 1970, 1973 and 1974. Insect larvae, snails, diving beetles, crayfish, tadpoles, and major fish species from both aquatic systems were analyzed for TCDD. Species diversity studies indicated no significant change in the composition of ichthyofauna between these dates or a control stream. Concentrations of TCDD (12 ppt) were found in only two species of fish from the stream, Notropis hypselopterus, Gunther (sailfin shiner), and Gambusia affinis, Baird and Girard (mosquitofish). Samples of skin, muscle, gonads, and gut were obtained from Lepomis punctatus, Valenciennes (spotted sunfish), from the test grid pond. Levels of TCDD in those body parts were 4,4,18 and 85 ppt, respectively. Gross pathological observations of the sunfish revealed no significant lesions or abnormalities.

Publications

Young, A. L., C. E. Thalken, and W. E. Ward. 1975. Studies of the ecological impact of repetitive aerial applications of herbicides on the ecosystem of Test Area C-52A, Eglin AFB, Florida. AFATL-TR-75-142, Air Force Armament Laboratory, Eglin AFB, Florida.

Young, A. L., P. J. Lehn, and M. F. Mettee. 1976. Absence of TCDD toxicity in an aquatic ecosystem. Presentation of the Weed Science Society of America, 3 February 1976, Denver, Colorado. Abstract No. 107.

14. Radiochemical Bioassay of TCDD Uptake in Plant Material

Principal Investigators: Captains James M. Cupello, and Alvin L. Young, Department of Chemistry and Biological Sciences.

Sponsored by Frank J. Seiler Research Laboratory (AFSC), and Air Force Logistics Command/DS.

The Environmental Protection Agency currently limits the commercial use of 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) herbicide due to the lack of environmental data on the teratogenic (birth-deforming) contaminant 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) found in many 2,4,5-T formulations. The consequence of such a use limitation directly affects the fate of a large USAF surplus inventory of Herbicide Orange (a 2,4,5-T formulation).

In a study to determine potential plant uptake of TCDD, a 2,000 pounds per acre equivalent application of Herbicide Orange, containing 14 parts per million (ppm) C-14 labelled TCDD, was placed 4" beneath the soil surface in specially constructed growth boxes, containing 100 plants of Sorghum, Sorghum vulgare, per box. The plants were grown under controlled environmental conditions for a period of nine weeks, at which time they were harvested and assayed for TCDD uptake. Analysis of the plant tissue (exclusive of root systems) prior to hexane extraction, and

after hexane extraction for four hours in a Soxhlet extraction apparatus, indicated the presence of an amount of C-14 activity that was equivalent to 430 parts per trillion (ppt) of TCDD in the plant material. This represents the maximum amount of TCDD that could be present in the plant material, but could also represent (1) the preferential plant uptake of a minor impurity in the TCDD formulation that was applied to the soil; (2) a metabolic breakdown product that was formed in the plant material after TCDD incorporation; or (3) the preferential uptake of a soil decomposition or soil microbial degradation product of the original TCDD placed in the soil.

15. Ultrastructural Studies of Liver Tissue from TCDD-Exposed Beach Mice (*Peromyscus polionotus*)

Principal Investigators: Major Lorris G. Cockerham and Captain Alvin L. Young, Department of Chemistry and Biological Sciences.

Sponsored by Air Force Logistics Command/DS.

In support of research on the biological effects of animals exposed to TCDD (2,3,7,8-tetrachlorodibenzo-p-dioxin), ultrastructural studies were initiated on beach mice, *Peromyscus polionotus*, collected from a herbicide-equipment testing range (Test Area C-52_A, Eglin AFB, Florida). Liver tissue from 30 mice (from the test and a control area) were examined with an electron microscope for possible ultrastructural changes in mitochondria and smooth and rough endoplasmic reticulum. Similar data were collected from 22 mice brought from the field into the laboratory and exposed to 30 days of external dusting with alumina gel (with or without 2.5 ppb TCDD).

Five representative electron micrographs were made from the liver tissue of each animal and the data were obtained from each micrograph

using a technique known as "point counting". With this method, a transparent grid of intersecting lines was placed at random over the micrographs and all the line intersections which were over the required cell structures were counted. The total area of the cytoplasm was then measured in the same manner. An analysis of variance was conducted to determine whether there was a statistically significant difference between populations of cells, i.e., percentage mitochondria and smooth and rough endoplasmic reticulum, removed from experimental and control animals. Analysis of data indicated no statistical differences between field control and field treatment animals, although differences were noted between field and laboratory animals.

16. Ultrastructural Evaluation of Tissues Removed from Animals Exposed to TCDD

Principal Investigator: Major Glenn M. Buchanan, Department of Chemistry and Biological Sciences

Sponsored by the Frank J. Seiler Research Laboratory (AFSC)

A long-term study of the possible biological hazards of pelage exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) has been initiated on the beachmouse, Peromyscus polionotus. Animals were divided into three experimental groups: untreated controls, a "treated" control group dusted with diatomaceous earth, and a "treated" group dusted with diatomaceous earth containing 2.5 parts per billion TCDD. The animals were dusted three times per week; the TCDD concentration simulating an exposure that was twice that expected from normal rangeland application rates of 0.1 parts per million TCDD contaminated 2,4,5-T herbicide. The effect of TCDD on animal weight, blood components, and

cellular ultrastructure (adrenal cortex, testes, liver and kidney) will be investigated following completion of the study. Animal weight data collected over a nine-week period indicated no effect due to TCDD applications, relative to controls.

D. Department of Civil Engineering, Engineering Mechanics, and Materials

1. Backing Strength of Masonry Shear Walls

Principal Investigator: Captain Richard H. Jolley, Department of Civil Engineering, Engineering Mechanics and Materials

Sponsored by the USAF Academy

Increased interest in using masonry for load-bearing structural systems has necessitated review of existing code design criteria and establishing improved design techniques. The objectives of this project are to (1) formulate a failure criteria for walls subjected to "in plane" loads and (2) establish a deflection prediction criteria for walls subjected to "in plane" loads.

The final report is complete and available to any agency or interested individual. Contact USAF Academy, DFCEM.

2. Validation of the Soil Stabilization Index System with Manual Development

Principal Investigators: Captains John J. Allen, and Dallas Little, and Technical Sergeant Glenn K. Kaneyuki, Department of Civil Engineering, Engineering Mechanics and Materials

Sponsored by the Air Force Civil Engineering Center and the Frank J. Seiler Research Laboratory, Air Force Systems Command (AFSC)

The project scope as detailed in Annual Research Progress Report No. 10 was extended to include required values of residual strength for