

DE GRUYTER

*Daniel J. Vogt, Joel P. Tilley,
Robert L. Edmonds*

SOIL AND PLANT ANALYSIS FOR FOREST ECOSYSTEM CHARACTERIZATION

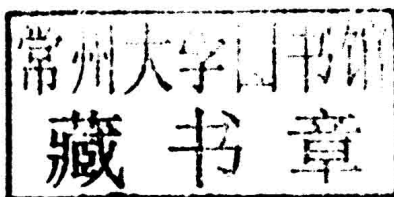
ECOSYSTEM SCIENCE AND APPLICATIONS



Higher
Education
Press

Daniel J. Vogt, Joel P. Tilley, Robert L. Edmonds

Soil and Plant Analysis for Forest Ecosystem Characterization



DE GRUYTER



Higher
Education
Press

This work is in the *Ecosystem Science and Applications* co-published by Higher Education Press and Walter de Gruyter GmbH.

ISBN 978-3-11-029029-5
e-ISBN (PDF) 978-3-11-029047-9
e-ISBN (EPUB) 978-3-11-038176-4
ISSN 2196-6737

Library of Congress Cataloging-in-Publication Data

A CIP catalog record for this book has been applied for at the Library of Congress.

Bibliographic information published by the Deutsche Nationalbibliothek

The Deutsche Nationalbibliothek lists this publication in the Deutsche Nationalbibliografie; detailed bibliographic data are available on the Internet at <http://dnb.dnb.de>.

© 2015 Higher Education Press and Walter de Gruyter GmbH, Berlin/Boston

Cover image: Julian Weber/Hemera/thinkstock

Printing and binding: CPI books GmbH, Leck

♻️ Printed on acid-free paper

Printed in Germany

www.degruyter.com



Daniel J. Vogt, Joel P. Tilley, Robert L. Edmonds
Soil and Plant Analysis for Forest Ecosystem Characterization

Ecosystem Science and Applications

Editors

Jiquan Chen

Heidi Asbjornsen

Kristiina A. Vogt

Acknowledgments

The authors would like to thank Dr. Kristiina A. Vogt, Patricia A. Roads and Roxanna Lewis for their editorial help on this book. However, any errors are not due to their editorial help but solely due to the authors.

Acronyms

ADP	Adenosine Diphosphate
AFDM	Ash-free Dry Mass
AM fungi	Arbuscular Mycorrhizal Fungi
ATGC	nucleotide bases: Adenine, Thymine, Guanine, Cytosine
ATP	Adenosine Triphosphate
BLAST	Basic Local Alignment Search Tool program for finding regions of local similarity between sequences
CB	Citrate-Bicarbonate
CDB	Citrate-Dithionite-Bicarbonate
cDNA	complementary DNA (cDNA) is DNA synthesized from a messenger RNA (mRNA) template in a reaction catalyzed by the enzymes reverse transcriptase and DNA polymerase, and is often used to clone eukaryotic genes in prokaryotes
CEC	Cation exchange capacity
CFV	Coarse Fraction Volume (as in the > 2 mm particles in the bulk soil)
CTAB	Cetyltrimethylammonium Bromide—a positively charged detergent
CV	Coefficient of Variation—statistical parameter that is the ratio of the standard deviation to the mean
dATP	Deoxyadenosine triphosphate (dATP)—a nucleoside triphosphate used in cells for DNA synthesis (or replication)
DBH	Diameter at breast height (1.3 m) of a tree
dCTP	Deoxycytidine triphosphate (dCTP)—a nucleoside triphosphate that contains the pyrimidine base cytosine
dsDNR	double stranded Deoxyribonucleic Acid (DNA)
DDI	Distilled De-Ionized (water)
DDW	Double Distilled Water
DEA	Dentrification Enzyme Activity
dGTP	Deoxyguanosine triphosphate (dGTP)—a nucleoside triphosphate, and a nucleotide precursor used in cells for DNA synthesis
DNA	Deoxyribonucleic Acid
dNTP	deoxy-nucleotide-tri phosphate—used by the DNA polymerase to add nucleotides to the elongating DNA strand (during replication). dNTP is a generic term referring to the four deoxyribonucleotide triphosphates: dATP, dCTP, dGTP and dTTP

DON	Dissolved Organic Nitrogen
DTPA	Diethylene Triamine Pentaacetic Acid
dTTP	Deoxythymidine triphosphate (dTTP)—one of the four nucleoside triphosphates that are used in the <i>in vivo</i> synthesis of DNA
DW	Distilled Water
EC	Electrical Conductivity
ECEC	Effective Cation Exchange Capacity
EDTA	Ethylene Diamine Tetraacetic Acid
EF-1 α	Eukaryotic Elongation Factor (also known as EF1A) catalyzes aminoacyl-tRNA binding by the ribosome during translation
EM	Ectomycorrhizas or ectomycorrhizal (fungi or root)
EMF	Electromotive Force (or Voltage)
EPA	The United States Environmental Protection Agency
EtBr	Ethidium Bromide
FAA	Formalin Acetic Acid
FAO	Food and Agriculture Organization of the United Nations
FF	Forest Floor, or sometimes litter or surface organic horizon in forests
FID	Flame Ionization Detector
GC	Gas Chromatograph
GC-rich	Guanine-Cytosine rich means that the GC as a percentage of nitrogenous bases on a DNA molecule is high (from a possibility of four different ones also including adenine and thymine)
GELCOMPAR II	Software Package—a program that makes it possible to link multiple electrophoresis fingerprints to the strains or samples studied and generate multiphasic groupings and identifications with databases of unlimited size
I.D.	Inner Diameter
IC	Ion Chromatography
ICP	Inductively Coupled Plasma Spectroscopy
ID	Identification Number
IRGA	InfraRed Gas Analyzer system
ITS	Internal Transcribed Spacer refers to a piece of non-functional RNA situated between structural ribosomal RNAs (rRNA) on a common precursor transcript
LCI	Ligno-cellulose Index
L-DOPA	L-3, 4-dihydroxyphenylalanine, a chemical that is made and used as part of the normal biology of humans, some animals and plants
LECO CHN	LECO corporation equipment that analyzes Carbon, Hydrogen, Nitrogen (CHN)
LOI	Loss on Ignition
LR0R/LR16	Small conserved subregion for the large subunit (LSU) rDNA gene
LSU rRNA	Large Sub Unit of the ribosomal ribonucleic acid (rRNA)
ML5/ML6	Conserved primer sequence for PCR amplification
MPN	Most Probable Number
NIST	National Institute of Standards and Technology

Oa Horizon	Organic horizon composed of highly decomposed organic material-humus or muck (can see very few signs of the original organic tissues) (USDA Soil Taxonomy [31])
OD	oven dried
Oe layer	Organic horizon composed of intermediate decomposed organic material-peat (can see some signs of the original organic tissues) (USDA Soil Taxonomy [31])
Oi Horizon	Organic horizon composed of slightly decomposed organic material (can still see evidence of the original organic tissues) (USDA Soil Taxonomy [31])
OM	Organic Matter
OSHA	United States Occupational Safety and Health Administration
PCB	Polychlorinated Biphenyl
PCR	Polymerase Chain Reaction
PDAB	p-dimethylaminobenzaldehyde
pH	The decimal logarithm of the reciprocal of the hydrogen ion activity in a solution
PHYLIP	PHYLogeny Inference Package—a free computational phylogenetics package of programs for inferring evolutionary trees
PLFA	PhosphoLipid ester-linked Fatty Acids
PVA	Polyvinyl Alcohol
PVC	Polyvinyl Chloride—rigid forms of PVC are frequently used in the construction of pipes or tubes
QA/QC	Quality Assurance/Quality Control
QC	Quality Control
QF-PCR	Quantitative Fluorescent Polymerase Chain Reaction
qPCR or QPCR	real-time quantitative Polymerase Chain Reaction
QRT-PCR	Quantitative Reverse-Transcriptase Polymerase Chain Reaction
rDNA	Ribosomal Deoxyribonucleic Acid
RFLP	Restriction Fragment Length Polymorphism
RNA	Ribonucleic Acid
RPB2	A gene that encodes for RNA polymerase
RQ-PCR	See RT-PCR
rRNA	ribosomal ribonucleic acid is the RNA component of the ribosome and is essential for protein synthesis in all living organisms
%RSD	Relative Standard Deviation, or also referred to as the absolute value of the Coefficient of Variation (CV)
RT-PCR	Reverse Transcription Polymerase Chain Reaction (not to be confused with Real Time PCR or qPCR)
SI unit	The International System of Units (abbreviated SI from French: Le Système international d'unités) is the modern form of the metric system
SOM	Soil Organic Matter
T	Temperature Correction Factor
TCA	Trichloroacetic Acid

TCD	Thermal Conductivity Detector
TDR	Time-Domain Reflectometry
TDS	Total Dissolved Solids
TE	commonly used buffer solution in molecular biology consisting of Tris (pH buffer) and EDTA (cation chelate); used to solubilize DNA or RNA while protecting it from degradation
TKN	Total Kjeldahl Nitrogen (Total Nitrogen using the Kjeldahl method)
TNPP	Total Net Primary Production
UPGMA	Unweighted Pair Group Method with Arithmetic mean
USDA	United States Department of Agriculture
UV	Ultra-violet radiation
W-B	Walkley-Black carbon
wt.	Weight

Contents

Part I Context of Soil and Plant Analysis — 1

1 Overview of Soil and Plant Analysis for Forest Ecosystems — 3

1.1 Soils are Physically, Chemically and Biologically Complex — 8

Part II Introductory Methods in Soil and Plant Analyses — 15

2 Field Characterization of Soils to Establish Sampling Protocols — 17

2.1 Soil Sampling Design and Methods — 17

2.1.1 Introduction to Sampling Design — 17

2.1.1.1 Accuracy, Bias, and Precision — 18

2.1.1.2 General Considerations on Soil Sampling — 20

2.1.1.3 Common Sampling Tools and Techniques — 20

2.1.1.4 Soil Sample Preparation — 21

2.1.2 Soil Sample Process Procedure — 23

2.1.2.1 Sample Variability—Number of Samples Required — 24

3 Plant Tissue Characterization — 29

3.1 Tissue Sampling — 30

3.2 Tissue Preparation and Laboratory Extraction — 33

4 Introduction: Laboratory Practices — 35

4.1 General Laboratory Protocol — 35

4.1.1 Safety — 36

4.1.2 Laboratory Water — 37

4.1.3 Clean-up — 37

4.1.4 Waste Disposal — 39

Part III Soil Physical, Chemical and Biological Analyses — 41

5 Methods for Analyzing Soil Physical Characteristics — 43

5.1 Soil Moisture — 43

5.1.1	Direct Methods of Estimating Soil Moisture and Soil Water Potential —	46
5.1.2	Procedure to Determine Gravimetric Water Concentrations —	47
5.2	Soil Bulk Density —	48
5.2.1	Soil Bulk Density Methods —	49
5.3	Soil Texture (Particle Size Analysis or Mechanical Analysis) —	50
5.3.1	Soil Texture Methods —	53
5.3.1.1	Soil Texture Procedure: Bouyoucos Hydrometer Method —	56
5.4	Soil Water Potential —	58
5.4.1	Pressure Plate Apparatus Procedure: Soil Moisture Release Curve —	63

6 Soil Chemical Characterization — 67

6.1	Soil pH —	70
6.1.1	Measuring pH —	71
6.1.2	The Care of pH Electrodes —	74
6.2	Electrical Conductivity (EC) —	75
6.2.1	Saturated Paste Extract Procedure: Electrical Conductivity —	80
6.3	Ion Exchange in Soils —	82
6.3.1	Cation Exchange Capacity —	82
6.3.2	Exchangeable Cations —	86
6.3.3	Extraction Procedures for Exchangeable Cations and Cation Exchange Capacity —	87
6.4	Exchangeable Soil Acidity —	91
6.4.1	Extraction Procedures for Exchangeable Soil Acidity —	92
6.4.1.1	Exchangeable Acidity (Barium Chloride—Triethanolamine Method) —	92
6.4.1.2	Exchangeable Acidity (Potassium Chloride Method) —	93
6.5	Extractable Inorganic Soil Nitrogen —	95
6.5.1	Extraction Methods for Inorganic Soil Nitrogen —	96
6.5.1.1	Single Extraction Procedure: Extractable Inorganic Nitrogen —	97
6.5.1.2	Double Extraction Procedure: Mechanical Vacuum Extractor —	98
6.6	Soil Phosphorus —	100
6.6.1	Methodology for Measuring Soil Phosphorus —	102
6.6.2	Procedure: Extractable Inorganic Phosphorus —	108
6.7	Soil Carbon and Organic Matter —	110
6.7.1	Dry Combustion Procedure: Total Soil Carbon and Nitrogen —	112
6.7.2	Loss on Ignition (LOI) Procedure: Total Soil Organic Matter —	113
6.7.3	Walkley-Black Procedure: Soil Carbon —	114
6.8	Selective Dissolution of Iron and Aluminum —	116
6.8.1	Extraction Procedure: Organically Complexed Iron and Aluminum —	117
6.8.2	Extraction Procedure: Non-crystalline Soil Iron and Aluminum Oxides —	118

7	Total Plant and Soil Nutrient Analysis (Digestion) — 121
7.1	Wet Oxidation Method — 121
7.2	Dry Oxidation Method — 123
7.3	Total Dissolved Carbon and Nitrogen in Water — 125
7.4	Modified Kjeldahl Digest Procedure: Sulfuric Acid Digest for “Total” Nutrients — 126
7.5	“Total” Nutrient Analysis Procedure: Dry Ashing Followed by Nitric Acid Digest — 129
7.6	Total Dissolved Nitrogen in Water Procedure: Persulfate Oxidation — 130
8	Soil Biology Characterization — 133
8.1	Soil Microbes — 134
8.1.1	Archaea and Bacteria — 135
8.1.2	Fungi — 137
8.1.3	Soil Algae and Cyanobacteria (Blue-green Algae) — 138
8.2	Methods for Determining Soil Microbial Diversity and Populations—Numbers and Biomass — 139
8.2.1	Direct Culture, Microscopy and Image Analysis — 139
8.2.2	Microbial Numbers and Microbial Biomass — 139
8.3	Mycorrhizas — 140
8.3.1	Types of Mycorrhizas — 140
8.3.2	Sampling Mycorrhizas — 142
8.3.2.1	Sampling Design — 143
8.3.2.2	Collection of Root and Soil Samples — 143
8.3.2.3	Storage of Samples — 143
8.3.2.4	Determining Mycorrhizas in Samples — 144
8.3.3	Determination of Mycorrhizal Fungal Species — 144
8.3.3.1	Analysis of Sporocarps and Spores — 145
8.3.3.2	Morphotypes of Ectomycorrhizas — 145
8.3.3.3	Trap Cultures for Arbuscular Mycorrhizal Fungi — 145
8.3.3.4	DNA and Biochemical Techniques — 146
8.3.4	Ectomycorrhizal Quantification — 147
8.3.5	Identification of Ectomycorrhizal Sporocarps — 150
8.3.6	Quantification of Arbuscular Mycorrhizal Colonization — 151
8.3.6.1	Staining — 151
8.3.6.2	Grid-line Intersection Method with a Dissecting Microscope — 152
8.4	Indirect Indices for Soil Biological Activity — 154
8.4.1	Soil Respiration — 154
8.4.1.1	CO ₂ Gas Sampling — 155
8.4.1.2	The Soda Lime Technique — 157
8.4.2	Decomposition Rates of Litter — 159
8.4.2.1	Fine Litter — 159
8.4.2.2	Woody Debris — 161
8.4.2.3	Fine Woody Debris — 161

8.4.2.4	Coarse Woody Debris (CWD) —	163
8.4.2.5	Standard Substrates —	165
8.4.2.6	Calculation of Decomposition Rates —	167
8.4.3	Soil Enzymes —	168
8.4.4	Functional Biodiversity—Phospholipid Ester-linked Fatty Acids (PLFA) and Substrate Utilization Profiles —	169
8.4.5	Molecular Tools for Ecological Systems —	170
8.4.5.1	DNA Extraction from Soil —	171
8.4.5.2	PCR —	172
8.4.5.3	Restriction Length Fragment Polymorphism RFLP —	174
8.4.5.4	Primers —	174
8.4.5.5	Gel Electrophoresis —	174
8.4.5.6	DNA Sequencing —	175
8.4.5.7	Metagenomics and Transcriptomics —	176
8.5	Soil Invertebrates —	176
8.5.1	Macrofauna with Emphasis on Earthworms —	178
8.5.1.1	Extraction Methods —	178
8.5.2	Mesofauna —	180
8.5.2.1	Mites and Collembola —	180
8.5.2.2	Enchytraeids —	183
8.5.3	Microfauna —	184
8.5.3.1	Protozoans —	184
8.5.3.2	Rotifers —	184
8.5.3.3	Tardigrades —	184
8.5.3.4	Microfauna-Nematodes —	185
8.6	Nitrogen Transformations —	186
8.6.1	Nitrogen Fixation —	187
8.6.1.1	Acetylene Reduction —	188
8.6.1.2	N Accretion through Time —	190
8.6.1.3	¹⁵ N Based Methods —	190
8.6.2	Denitrification —	191
8.6.2.1	The Acetylene Inhibition Method —	192
8.6.2.2	¹⁵ N Tracer Methods —	192
8.6.2.3	Direct N ₂ Quantification —	192
8.6.2.4	Mass Balance Approaches —	192
8.6.2.5	Stable Isotope Approaches —	193
8.6.2.6	Approaches using <i>in situ</i> Gradients in Environmental Tracers —	193
8.6.2.7	Molecular Approaches —	193

Appendices —195

References —203

Subject Index —217

List of Tables

Tab. 1.1	Processes that form all soils —	10
Tab. 1.2	Processes that form all soils but depend on environmental conditions —	10
Tab. 1.3	Factors that cause soils to degrade —	11
Tab. 2.1	Definitions of some sample measurement terms —	19
Tab. 2.2	An example of calculating the variability and precision associated with sampling —	25
Tab. 5.1	United States Department of Agriculture (USDA) textural classes of soils based on the USDA particle-size classification and potential ranges of percentages of soil separates —	53
Tab. 5.2	Water potential in the soil-plant-atmosphere system —	61
Tab. 6.1	Role of some mineral elements in plant growth and functions and mineral links to human health —	68
Tab. 6.2	Some nutrient concentration ranges in plants and soils —	69
Tab. 6.3	Descriptive terms for ranges of soil pH —	74
Tab. 6.4	Definition of Soil Salinity classes related to Electrical Conductivity (EC) —	76
Tab. 6.5	Soil types related to pH, EC, SAR, and ESP classes —	76
Tab. 6.6	Effects of Electrical Conductivity (EC) on crops —	77
Tab. 7.1	Percent recovery of 11 elements in a modified Kjeldahl digest —	122
Tab. 7.2	Suggested calibration series for Dissolved Organic Nitrogen (DON) determination —	131
Tab. 8.1	Numbers of species and genera in the bacteria phyla —	136
Tab. 8.2	Characteristics of the different types of mycorrhizas —	141
Tab. 8.3	Example calculation for CO ₂ evolution from a 15-cm diameter cylinder in the soil —	159
Tab. 8.4	Calculations for estimating k (yr ⁻¹) for decaying Pacific silver fir needles —	168
Tab. C.1	Example of a Field Data Recording Sheet —	198
Tab. C.2	Soil Data Summary Sheet —	200
Tab. C.3	Soil Water Concentration Data Sheet —	200
Tab. C.4	Soil Bulk Density Data Sheet —	201

List of Figures

- Fig. 1.1 Factors of human land uses and their influences on our natural ecological systems including soils — 4
- Fig. 1.2 Total net primary productivity in tropical moist forests by soil texture classes — 13
- Fig. 5.1 Texture triangle of different proportions of sand, silt, and clay forming 12 texture classes — 51
- Fig. 5.2 Comparison of particle size scales according to different organizations — 52
- Fig. 5.3 Soil texture classes determined by hand — 54
- Fig. 5.4 Schematic diagram of water molecule — 58
- Fig. 5.5 An example of hysteresis in soil water retention curves of wetting and drying curves — 63
- Fig. 6.1 A schematic diagram of a pH glass electrode and expanded view of the electrode bulb — 73
- Fig. 6.2 A schematic of an Electrical Conductivity Cell — 78
- Fig. 8.1 Using the gridline intersection method for estimating total root length and numbers of ectomycorrhizal root tips — 149
- Fig. 8.2 A gridline intersection example using an 8.5 cm diameter round Petri dish with a 1/2 inch (1.27 cm) grid and a 1 m test sample of thread cut into fragments and randomly re-distributed 10 times — 153
- Fig. 8.3 Diagram showing examples of calculating different volumes of a log such as total, wood and bark volumes — 164
- Fig. 8.4 Average ash-free dry mass of Pacific silver fir needles remaining in litter bags at five sampling times in the Cascade Mountains of Washington, USA — 168
- Fig. 8.5 A schematic of the Tullgren or Berlese funnel to extract soil invertebrates from the soil — 181
- Fig. 8.6 High gradient invertebrate extractor — 182
- Fig. A.1 Frequency distribution of 100 normally distributed random observations, with mean = 99.582 and standard deviation = 9.773. — 195
- Fig. A.2 Frequency distribution of 20 means of 5 observations each — 196
- Fig. A.3 Frequency distribution of 10 means of 10 observations each — 197