FOOD ANALYSIS

Principles and Techniques

Volume 3

Biological Techniques

Edited by

DIETER W. GRUENWEDEL

JOHN R. WHITAKER

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FOOD ANALYSIS

Principles and Techniques

A Treatise in Eight Volumes

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Volume 1: Physical Characterization
Volume 2: Physicochemical Techniques
Volume 3: Biological Techniques
Volume 4: Separation Techniques
Volume 5: Proximate Analysis
Volume 6: Physical Techniques
Volume 7: Spectroscopy
Volume 8: Electroanalytical Techniques

Introduction to the Treatise

Food Analysis: Principles and Techniques was conceived to meet the need for an up-to-date detailed treatment of the methods of modern food analysis. Each chapter, written by an expert in the area, is carefully reviewed to provide as much uniformity of style as possible, yet to allow for individual creativity and the requirements of specific topics. Each chapter addresses first the underlying principles of an area in sufficient detail that those not initially familiar with the method can develop sufficient knowledge of the area to utilize the principles and techniques required. Each chapter then contains numerous examples of the application of the principles in the analysis of compounds of interest in foods.

The treatise is not a handbook of methods of analysis, and the reader should not expect to find a detailed description of the step-by-step procedures to be used in an analysis. The treatise also is not intended as a primary textbook for the instruction of undergraduate students in the principles and techniques of food analysis, although it will prove valuable as a reference source for these students. Rather, the treatise is intended for use by graduate students and by all scientists involved in the analysis of biological materials.

Food analysis has grown by leaps and bounds in recent years, in part due to better instrumentation, but also in part because it has recognized the need to understand and to apply the modern principles of analytical techniques—developed in large part by the physicists, the chemists, and the biochemists—to problems associated with quality assurance, product development, and food safety. In this treatise, we have deliberately sought out authors who are at the

cutting edge of their area of science, who understand what the major advances have been, how they can be applied to analysis related to food products, and what the future needs are in analytical techniques.

It is a truism that we all benefit from the availability and application of proper analytical techniques, whether we be scientist or consumer of food products. The need to know whether a particular compound is present, in what quantity, and any adverse effect it may elicit is an ever-present question that must depend for its answer on the most modern analytical techniques available and applied by those familiar with the advantages, limitations, and statistical significance of methods. Determination of the presence of a compound is often required at the picogram and fentogram levels.

Food analysis is a very broad and rapidly developing science. It is impossible to cover every conceivable method, even in a treatise of eight volumes as planned for *Food Analysis: Principles and Techniques*. Yet, it seemed to us that there are a number of areas of food analysis that must be incorporated and that the potential must be provided for additional volumes when emerging analytical techniques have reached a sufficient level of development.

The eight volumes will deal with the following topics:

Volume 1. Physical Characterization
Volume 2. Physicochemical Techniques
Volume 3. Biological Techniques
Volume 4. Separation Techniques
Volume 5. Proximate Analysis
Volume 6. Physical Techniques
Volume 7. Spectroscopy
Volume 8. Electroanalytical Techniques

The continuous encouragement, advice, and help of Dr. Maurits Dekker and the dedicated assistance of the staff of Marcel Dekker have provided the climate that made this treatise possible.

It is the authors of each of the chapters who have earned our undying gratitude for their dedication to the principles of excellence, for their desire to educate others in the principles and techniques of food analysis and thereby contribute to the quality and adequacy of the food supply, and in particular for their patience with the editors as this project evolved.

We welcome constructive suggestions from the users of the treatise. We hope that it will make a major contribution to the continued improvement in the application of modern analytical techniques in food analysis.

Dieter W. Gruenwedel
John R. Whitaker

Preface

The chapters of Volume 3 of *Food Analysis: Principles and Techniques* describe and discuss in detail a number of biological techniques that are of vital importance to the quality and safety assessment of foods, particularly in the areas of nutrition, food science, and food toxicology. The techniques, broadly speaking, encompass two methodological approaches: one that employs live systems as analytical tools (Chapters 1, 2, and 4) and one that uses extracellular biochemical techniques (Chapters 5 and 6). Chapter 3 may be considered to contain material from both.

The use of the whole animal as an analytical tool is a time-honored technique and one that is of particular importance in the nutritional as well as toxicological evaluation of foods. While expensive and time consuming, whole animal experimentation provides a combined assessment of digestibility, absorption, nutritional quality, and metabolism of foods. A similarly time-tested approach is the employment of microorganisms such as bacteria, fungi, protozoa, and yeasts in the nutritional evaluation of foods. Of more recent vintage is the employment of cell and tissue culture methodology in food research. While, at present, cell and tissue culture are utilized primarily in assessing food safety, particularly regarding the contamination of foods with mutagens, oncogens, and environmental toxicants, indications are that they will soon find more and more employment in the nutritional evaluation of foods as well. The techniques using cultured cells and tissues or microorganisms such as bacteria, fungi, protozoa, and yeasts, are less expensive, less time consuming, and statistically more valid than whole animal experiments.

With the advance of modern instrumentation, nonculture methods for analyzing foods with respect to microbial contamination have become increasingly important and useful in food research. The nonculture tests are rapid and sensitive, and one approach permits even non-destructive sampling. Food quality tests that also combine sensitivity and speed and, in addition, possess high specificity, are immunochemical and enzymatic techniques. The latter have further advantage of being inexpensive and amenable to automation. Both immunochemical and enzymatic techniques permit assays to be completed in a few minutes.

There can be no doubt that biological and biochemical techniques are of increasing importance to food analysis. Each of the chapters of Volume 3 is written by an expert. The reader is assured of an up-to-date and accurate presentation of the principles and methodology of the techniques.

We thank all the authors for their contributions to this volume.

Dieter W. Gruenwedel
John R. Whitaker

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1

The Whole Animal as an Analytical Tool

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I. INTRODUCTION

Nutrition deals basically with the science of food; the nutrients and other substances therein; their analyses, action, interaction, and balance relative to the health or lack of health of the whole animal; and the process by which the animal ingests, digests, absorbs, transports, utilizes, and excretes food substances. Since the science of nutrition began with an emphasis on chemical and biochemical techniques, the organized study of nutrition has been confined to the twentieth century. Interest in the relationship between human beings and their food, however, began much earlier. The history of nutrition can be divided into the following three eras: the naturalistic era (400 B.C.–A.D. 1750), the chemical-analytical era (1750–1900), and the biological era (1900–the present).

In the naturalistic era, food was considered essential for survival but there was no concern for the value of various foods. Hippocrates, in 400 B.C., considered food as one universal nutrient; by the sixteenth century the principle of diet and longevity was well established; and by 1747, in the first controlled nutrition experiment, Lind found that lemon or lime juice cured scurvy whereas other liquids, such as seawater and vinegar, did not. During the chemical-analytical era, Lavoisier, Black, and Priestley studied respiration, oxidation, and calorimetry. Early in the nineteenth century, analytical methods for carbon, hydrogen, and nitrogen in organic substances became available. These methods led Liebig to hypothesize that an adequate diet should contain protein, carbohydrate, and fat. This hypothesis was later tested by Dumas and Lunin with synthetic foods containing