SECOND EDITION

FLAVOR CHEMISTRY and TECHNOLOGY

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SECOND EDITION

FLAVOR CHEMISTRY and TECHNOLOGY

GARY REINECCIUS

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Dedication

To my teachers

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Preface

This book combines the essentials of both flavor chemistry and flavor technology. Modern flavor chemistry as an academic area of study has existed for over 40 years. The advent of gas chromatography in the early 1960s (rapidly coupled with mass spectrometry) heralded the beginning of this research area. These instruments gave scientists the ability to separate and identify the host of volatiles found in our foods that contribute to the aroma of foods.

Flavor chemistry flourished in the U.S., Europe, and Japan in the late 1960s and early 1970s. Since money was readily available for flavor research, great strides were made in understanding the biosynthetic pathways of flavor formation and the chemical constituents that are important to flavor. Government support for flavor research in the U.S. started to decline in the early 1980s and stabilized at a level that has permitted a limited number of laboratories to exist. Fortunately, flavor research in Europe has continued to be funded, and Europe has become the leader in flavor research. In recent years, the food industry and various commodity groups such as the dairy industry have come to recognize the value of flavor to the consumer and have provided increasing support to the academic community in the U.S. Thus, we find a growth in flavor research once again in the U.S.

Our journey of understanding has taken us from providing long lists of volatile chemicals found in foods (thought to be the way to replicate nature), to studies to determine the key volatile compounds, to studies on food/aroma interactions, flavor release, and most recently, to taste, perception, and cognitive functioning. It is unlikely that this is the final frontier — for we thought we were there nearly 40 years ago! Nature is not readily giving up its secrets to flavor and the pleasure it provides in our daily lives.

Flavor technology is an ancient area of study. Man has searched for a means of making food more pleasurable or palatable since time began. The flavor industry had its beginnings with the search for, and trading of, spices many centuries ago. This searching and trading has been responsible for the discovery of new worlds. Today this industry is nearly invisible to the average consumer since only a few members of the industry sell directly to the consumer: most are ingredient suppliers only known to their client companies who interface with the ultimate consumer.

The flavor industry today has a world market of approximately 16.3 billion dollars (www.Leffingwell.com). The total global sales are only somewhat larger than a single major food company. The flavor industry is comprised of a few large companies and a host of small companies (perhaps 500+). There is little published about today's flavor industry because of its highly competitive and secretive nature. It is difficult and sometimes impossible to protect a flavoring via patents, so flavoring materials, synthetic chemicals, and manufacturing procedures are kept secret to avoid supporting competitive companies.

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The author has used the best information available to try to give a view of this industry. Often patents or personal experience have been called on due to a dearth of public information on the industry. While only a sampling of the literature available could be included in many areas, the author has provided abundant referencing to provide direction for the reader. This book is intended to serve as a suitable text for both undergraduate and graduate level courses as well as a reference text for those in academia or industry.

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Acknowledgments

I wish to acknowledge the host of individuals referenced in this text for their efforts to provide understanding to a field that is ever so slowly giving up its secrets. I wish to thank my many teachers for sharing their knowledge and helping me down a career path that has been most challenging and exciting. I have to thank my students also for educating me in their specific research topics — I am certain they taught me more than I taught them. I will forever be in debt to Henry Heath for what he has given to the field and to me personally through his writing. Several sections of this book still reflect his contributions.

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The Author

Gary Reineccius, Ph.D., is a professor in the Department of Food Science and Nutrition at the University of Minnesota. He has been actively involved in flavor research for more than 35 years. During this time he has published over 190 research articles. Dr. Reineccius has spent sabbatical leaves with Fritzsche Dodge and Olcott (New York, flavor creation and production), Nestle (Switzerland, reaction flavors), and most recently Robertet S.A. (France, taste modifiers and manufacturing).

Dr. Reineccius teaches courses in Chemical and Instrumental Analysis of Foods, Food Processing, and Flavor Chemistry and Technology. He has written a college textbook on food flavors with Henry Heath. This was the first textbook in the flavor area that combined both flavor chemistry and technology. Dr. Sara Risch and he edited and were major contributors to two books on flavor encapsulation. He is the editor of the *Source Book of Flavors* and an ACS symposium proceeding titled "Heteroatomic Aroma Compounds."

Dr. Reineccius' achievements have been recognized by several local and international organizations. He is an honorary member of the Society of Flavor Chemists. He has been granted the Palmer Award for his contribution to chromatography by the Minnesota Chromatography Forum. He has received the Distinguished Achievement and Service in Agricultural and Food Chemistry Award from the American Chemical Society and been presented the Stephen S. Chang Award by the Institute of Food Technologists. These are the highest awards given to individuals in the flavor area. He is a Fellow of the American Chemical Society.

He often speaks at public schools and other groups. His favorite lay topics are chocolate (he spent three years researching chocolate flavor for his Ph.D. thesis) and the chemistry of gourmet cooking. From a professional standpoint, his favorite topic is flavor encapsulation. He has been actively engaged in research in this area since 1964.

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Contents

PART I Flavor Chemistry 1

Chapter 1 An Overview of Flavor Perception 3

1.1 Flavor Perception 3
1.2 Taste Perception 3
  1.2.1 Anatomy of Taste 4
  1.2.2 Synopses of Tastes 6
1.3 Chemesthesis 10
  1.3.1 Chemesthetic Responses 10
  1.3.2 Tactile Response 12
1.4 Olfaction 13
  1.4.1 Anatomy of Olfaction 14
  1.4.2 Odor Receptor Functioning 14
  1.4.3 Signal Encoding 15
1.5 Summary 18
References 18

Chapter 2 Flavor and the Information Age 23

2.1 Introduction 23
2.2 History of Flavor Literature 23
2.3 Journals 24
2.4 Professional Societies 25
2.5 Internet 27
  2.5.1 Internal Communication 27
  2.5.2 External Communication 28
  2.5.3 General Information About the Industry 28
  2.5.4 Societies/Organizations 28
  2.5.5 Internet Searches 28
  2.5.6 Literature Retrieval 29
  2.5.7 Locating Materials and Equipment Suppliers 30
  2.5.8 Idea Generators/Promotions 31
  2.5.9 Competition 31
  2.5.10 How to Use the Internet Effectively 31
2.6 Summary 31
References 31

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Chapter 3 Flavor Analysis 33

3.1 Introduction 33
3.2 Aroma Compounds 33
3.2.1 Introduction 33
3.2.2 Sample Selection/Preparation 35
3.2.3 Principles of Aroma Isolation 36
3.2.3.1 Introduction 36
3.2.3.2 Solubility 36
3.2.3.3 Sorptive Extraction 38
3.2.3.4 Volatility 39
3.2.4 Methods of Aroma Isolation 41
3.2.4.1 Static Headspace 41
3.2.4.2 Headspace Concentration Methods (Dynamic Headspace) 43
3.2.4.3 Distillation Methods 45
3.2.4.4 Solvent Extraction 48
3.2.4.5 Sorptive Extraction 49
3.2.4.6 Concentration for Analysis 51
3.2.4.7 Aroma Isolation Summary 52
3.2.5 Analysis of Aroma Isolates 53
3.2.5.1 Prefractionation 53
3.2.5.2 Gas Chromatography 54
3.2.5.3 GC/Olfactometry (GC/O) or GC-MS/Olfactometry (GC-MS/O) 54
3.2.5.4 Mass Spectrometry 56
3.2.6 Specific Analysis 58
3.2.6.1 Key Components in Foods 58
3.2.6.2 Aroma Release During Eating 61
3.2.7 Electronic Noses 63
3.3 Taste Compounds (Nonvolatiles) 64
3.3.1 Introduction 64
3.3.1.1 Taste Compounds 64
3.3.1.2 Other Nonvolatile Components of Foods 65
3.3.2 The Analysis of Taste Substances 65
3.3.2.1 Sweeteners 65
3.3.2.2 Salt 66
3.3.2.3 Acidulants 66
3.3.2.4 Umami 66
3.3.2.5 Bitter Substances 66
References 67

Chapter 4 Flavor Formation in Fruits and Vegetables 73

4.1 Introduction 73
4.2 Biogenesis of Fruit Aroma 74

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4.2.1 Aroma Compounds from Fatty Acid Metabolism 74
4.2.2 Aroma Compounds from Amino Acid Metabolism 77
4.2.3 Aroma Compounds Formed from Carbohydrate Metabolism 80
4.3 Biogenesis of Vegetable Aroma 83
4.3.1 The Role of Lipids in Vegetable Aroma Formation 85
4.3.2 Aroma Compound Formation from Cysteine Sulfoxide Derivatives 86
4.3.3 Glucosinolates as Aroma Precursors in Vegetables 88
4.3.4 Additional Pathway for Vegetable Flavor Formation 88
4.4 Glycosidically Bound Aroma Compounds 88
4.4.1 Glycoside Structure 91
4.4.2 Freeing of Glycosidically Bound Flavor Compounds 91
4.5 Location of Flavor in Plant 92
4.6 Influence of Genetics, Nutrition, Environment, Maturity, and Storage on Development of Flavor 92
4.6.1 Plant Products 93
4.6.1.1 Genetics 93
4.6.1.2 Environmental and Cultural Effects on Flavor Development on the Plant 93
4.6.1.3 Influence of Maturity and Postharvest Storage on Flavor Development off the Plant 94
4.7 Animal Products 97
4.8 Conclusions 97
References 98

Chapter 5 Changes in Food Flavor Due to Processing 103

5.1 Introduction 103
5.2 The Maillard Reaction 103
5.2.1 General Overview of the Maillard Reaction 103
5.2.2 Pathways for Flavor Formation via the Maillard Reaction 104
5.2.3 Factors Influencing the Maillard Reaction 105
5.2.3.1 Heating Time/Temperature 106
5.2.3.2 Influence of System Composition 108
5.2.3.3 Influence of Water Activity 109
5.2.3.4 Influence of pH 109
5.2.3.5 Influence of Buffer/Salts 110
5.2.3.6 Influence of Oxidation/Reduction State 110
5.2.4 Kinetics of the Maillard Reaction and Flavor 111
5.2.4.1 Pyrazines 111
5.2.4.2 Oxygen-Containing Heterocyclic Compounds 112
5.2.4.3 Sulfur-Containing Compounds 113
5.2.4.4 Miscellaneous Compounds 113
5.2.4.5 Summary 114
5.2.5 Flavor Formation via the Maillard Reaction 114

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5.2.5.1 Carbonyl Compounds 115
5.2.5.2 Nitrogen-Containing Heterocyclic Compounds 115
5.2.5.3 Oxygen-Containing Heterocyclic Compounds 117
5.2.5.4 Sulfur-Containing Heterocyclic Compounds 118
5.2.5.5 Oxygen-Containing Compounds 119
5.3 Flavors from Lipids 119
5.3.1 Deep Fat Fried Flavor 119
5.3.2 Lactones 121
5.3.3 Secondary Reactions 121
5.4 Flavors Formed via Fermentation 123
5.4.1 Esters 123
5.4.2 Acids 124
5.4.3 Carbonyls 125
5.4.4 Alcohols 127
5.4.5 Terpenes 128
5.4.6 Lactones 129
5.4.7 Pyrazines 129
5.4.8 Sulfur Compounds 130
5.4.9 Conclusions 130
References 132

Chapter 6 Flavor Release from Foods 139

6.1 Introduction 139
6.2 Lipid/Flavor Interactions 140
6.2.1 Effect of Fat: Flavor Interactions on Aroma 141
6.2.1.1 Equilibrium Conditions 141
6.2.1.2 Dynamic Conditions 143
6.2.2 Effect of Fat: Flavor Interactions on Taste 144
6.3 Carbohydrate: Flavor Interactions 145
6.3.1 Simple Sugars: Aroma Interactions 145
6.3.2 High Potency Sweeteners: Aroma Interactions 146
6.3.3 Complex Carbohydrate (Polysaccharide): Aroma Interactions 148
6.3.3.1 Chemical Interactions 149
6.3.3.2 Resistance to Mass Transfer 151
6.3.4 Carbohydrate: Taste Interactions 152
6.4 Protein: Flavor Interactions 153
6.4.1 Protein: Aroma Interactions 153
6.4.1.1 Chemical Interactions 153
6.4.1.2 Resistance to Mass Transfer 155
6.4.2 Protein Hydrolysate: Aroma Interactions 155
6.4.3 Protein: Taste Interactions 155
6.5 Minor Food Components: Aroma Interactions 155
6.5.1 Melanoidin: Flavor Interaction 156
6.5.2 Hydrogen Ion Effects (pH effects) 156

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6.6 Summary 157
References 157

Chapter 7 Off-Flavors and Taints in Foods 161

7.1 Introduction 161
7.2 Sensory Aspects of Off-Flavor Testing 161
7.3 Taints in Foods 163
7.3.1 Airborne Sources 164
7.3.2 Waterborne Sources 166
7.3.3 Disinfectants, Pesticides, and Detergents 167
7.3.4 Packaging Sources 168
7.4 Off-Flavors Due to Genetics or Diet 174
7.4.1 Genetics 174
7.4.2 Diet 174
7.5 Off-Flavors Due to Chemical Changes in the Food 176
7.5.1 Lipid Oxidation 176
7.5.2 Nonenzymatic Browning 183
7.5.3 Photo-Induced Off-Flavors 184
7.5.4 Enzymatic Flavor Changes 185
7.6 Microbial Off-Flavors 189
7.7 Summary 192
References 193

PART II Flavor Technology 201

Chapter 8 Flavoring Materials 203

8.1 Introduction 203
8.1.1 Definitions 203
8.1.1.1 Flavoring 203
8.1.1.2 Natural Flavoring 203
8.1.1.3 Artificial Flavoring 204
8.2 Natural Flavoring Materials (Plant Sources) 204
8.2.1 Herbs and Spices 205
8.2.1.1 Definitions and Markets 205
8.2.1.2 Historical Associations 206
8.2.1.3 Classification of Herbs and Spices 209
8.2.1.4 Flavor Character of Herbs 211
8.2.1.5 Preparation of Herbs for Market 212
8.2.1.6 Introduction to Spices 213
8.2.2 Derivatives of Spices 216
8.2.2.1 Essential Oils (Distillation) 216
8.2.2.2 Oleoresins (Solvent Extraction) 220

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8.2.2.3 Expressed "Essential" Oils (Citrus Oils) 224
8.2.2.4 Mint Oils 230
8.2.2.5 Fruit, Fruit Juices, and Concentrates 236
8.2.2.6 Vanilla 241
8.2.2.7 Cocoa, Coffee, and Tea Flavorings 249
8.2.2.8 Aromatic Vegetables 253
References 256

Chapter 9 Flavoring Materials Made by Processing 261

9.1 Introduction 261
9.2 Natural Products Made by Roasting: Cocoa/Chocolate 261
9.2.1 Production of Cocoa Powder 262
9.2.2 The Dutch Process 262
9.2.3 Chocolate 262
9.3 Process Flavors: Meat-Like Flavors 263
9.3.1 The Evolution of Process Meat-Like Flavorings 264
9.3.2 The Creation of Process Flavorings 265
9.3.2.1 Reaction System Composition 265
9.3.2.2 Reaction Conditions 271
9.3.2.3 Final Flavor 272
9.3.3 Hydrolyzed Vegetable Proteins (HVP) 273
9.3.4 Autolyzed Yeast Extracts (AYE) 273
9.4 Enzymatically Derived Flavorings 274
9.4.1 Introduction 274
9.4.2 Properties of Enzyme Catalyzed Reactions 275
9.4.3 Enzyme Modified Butter/Butter Oil 278
9.4.4 Enzyme-Modified Cheese (EMC) 279
9.4.4.1 Enzymes Used 280
9.4.4.2 General Processes Employed 281
9.4.5 Further Processed EM Dairy Products 282
9.5 Flavors Made by Fermentation 283
9.5.1 Yeasts 283
9.5.2 Vinegar/Acetic Acid 283
9.5.3 Dried Inactive Yeast Powder 285
9.6 Flavors Made by Pyrolysis: Smoke Flavors 285
9.6.1 The Smoking of Foods 285
9.6.2 Natural Liquid Smoke Flavorings 286
9.6.3 Pyroligneous Acid 286
9.6.4 Smoke Condensates 286
9.7 Biotechnology to Produce Flavoring Materials 287
9.7.1 Introduction 287
9.7.2 Production of Natural Flavoring Materials by Enzymatic Action 287
9.7.2.1 Ester Formation 288
9.7.2.2 Resolution of Racemic Mixtures 290

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9.7.3 Production of Natural Flavoring Materials by Microbial Action 290
9.7.3.1 Fermentation to Produce Flavoring Materials 291
9.7.3.2 Bioconversions Via Microorganisms 293
9.7.3.3 Resolution of Racemic Mixtures 293
9.7.4 Economics of Biotechnology 294
References 294

Chapter 10 Artificial Flavoring Materials 299

10.1 Artificial Flavorings 299
10.2 Synthetic Flavoring Materials 299
10.2.1 Introduction 299
10.2.2 Consumer Attitudes Toward Synthetic Chemicals 300
10.3 Classification of Aroma Compounds by Molecular Structure 300
10.4 Sensory Threshold Values 303
10.5 Sensory Characters of Odor Compounds 305
10.5.1 Hydrocarbons 306
10.5.2 Carboxylic Acids 307
10.5.3 Acetals 307
10.5.4 Alcohols 308
10.5.5 Carbonyls 308
10.5.5.1 Aldehydes 309
10.5.5.2 Ketones 309
10.5.6 Esters 309
10.5.7 Ethers 310
10.5.8 Heterocyclic Compounds 310
10.5.9 Lactones 311
10.5.10 Nitrogen-Containing Compounds 311
10.5.10.1 Amines 311
10.5.10.2 Amides 311
10.5.10.3 Imines 312
10.5.10.4 Amino Acids 312
10.5.10.5 Isothiocyanates 312
10.5.11 Phenols 312
10.5.12 Sulfur-Containing Compounds 312
10.6 Nomenclature of Organic Chemicals 313
References 313

Chapter 11 Flavor Potentiators 317

11.1 Introduction 317
11.2 Chemical Properties of L-Amino acids and 5′-Nucleotides 318
11.2.1 Structures 318
11.2.2 Stability 319

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11.3 Sensory Properties of MSG and 5′-Nucleotides 320
11.3.1 Influence of MSG and 5′-Nucleotides on Taste 320
11.3.2 Influence of MSG and 5′-Nucleotides on Aroma 321
11.3.3 Synergism Between MSG and the 5′-Nucleotides 322
11.4 Traditional Flavor Potentiators in Foods 322
11.4.1 MSG and 5′-Nucleotides in Foods 322
11.4.1.1 Yeast Extracts 323
11.4.1.2 Hydrolyzed Vegetable Proteins (HVP) 324
11.4.2 MSG and 5′-Nucleotides Added to Foods 325
11.4.3 Sources of MSG and 5′-Nucleotides 326
11.4.4 Table Salt as a Flavor Potentiator 329
11.5 Toxicity of MSG and 5′-Nucleotides 329
11.5.1 Monosodium Glutamate 329
11.5.2 5′-Nucleotides 330
11.6 Other Potentiators 330
11.6.1 Beefy Meaty Peptide 330
11.6.2 Umami Tasting Glutamate Conjugates 331
11.6.3 Alapyridaine 331
11.6.4 Sweetness Potentiators 332
11.6.4.1 Maltol and Ethyl Maltol 332
11.6.4.2 Cyclic Enolones 332
11.6.5 Other Potentiators 332
References 332

Chapter 12 Flavorists and Flavor Creation 337

12.1 Introduction 337
12.2 The Flavorist 337
12.2.1 Selection of Individuals to Become Flavorists 337
12.2.2 Training of Flavorists 338
12.3 Working Environment 338
12.4 Flavor Creation 340
12.4.1 Imitation Flavorings 340
12.4.2 Blending of Seasonings (Culinary Products) 345
12.5 Sensory Assessment 346
12.5.1 Sample Evaluation 347
12.6 Conclusions 348
References 348

Chapter 13 Flavor Production 351

13.1 Introduction 351
13.2 Liquid Flavorings 351
13.3 Emulsions 352
13.3.1 Beverage Emulsions 353
13.3.2 Baker's Emulsions 360

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13.4 Dry Flavorings 360
13.4.1 Extended or Plated Flavors 360
13.4.2 Inclusion Complexes (Cyclodextrins and Starches) 362
13.4.2.1 Cyclodextrins 362
13.4.2.2 Starch 364
13.4.3 Phase Separation/Coacervation Processes 364
13.4.4 Dehydration Processes 366
13.4.4.1 Spray Drying 367
13.4.4.2 Freeze, Drum, and Tray Drying 375
13.4.5 Extrusion 377
13.4.5.1 Traditional Processes and Formulations 377
13.4.5.2 Modern Extrusion Processes 380
13.4.5.3 Shear Form Process 382
13.5 Controlled Release 383
13.6 Summary 383
References 384

Chapter 14 Flavor Applications 391

14.1 Introduction 391
14.2 The People 391
14.3 The Laboratory 392
14.4 Specific Flavoring Applications 392
14.4.1 Culinary and Meat Products 393
14.4.1.1 Soups and Stocks 394
14.4.1.2 Sauces, Seasonings, and Marinades 395
14.4.1.3 Meat Products 396
14.4.2 Baked Goods and Bakery Products 397
14.4.2.1 Problems in Flavoring Baked Goods 398
14.4.2.2 Flavoring Baked Goods 398
14.4.2.3 Heat Resistant Flavorings 399
14.4.3 Snack Foods 400
14.4.3.1 Problems in Flavoring Snack Foods 401
14.4.3.2 Snack Flavorings 401
14.4.3.3 Flavoring Materials 402
14.4.3.4 Means of Applying Flavorings 405
14.4.4 Sugar-Based Confectionery Products and Chewing Gum 406
14.4.4.1 Hard Candies 406
14.4.4.2 Caramels (Toffees) 407
14.4.4.3 Pressed Tablets 408
14.4.4.4 Starch-Deposited Chews 408
14.4.4.5 Chewing Gum 409
14.4.5 Dairy Products 410
14.4.5.1 Flavored Milks 410
14.4.5.2 Flavored Yogurts 411
14.4.5.3 Flavored Dairy Desserts 412

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14.4.6 Soft Drinks 414
14.4.6.1 Introduction 414
14.4.6.2 Carbonated Beverages 415
14.5 Summary 416
References 417

Chapter 15 Flavor Legislation and Religious Dietary Rules 419

15.1 Introduction 419
15.2 Legislation Limiting the Use of Flavor Compounds 419
15.2.1 U.S. Flavor-Related Legislation 420
15.3 Religious Dietary Rules 421
15.3.1 Kosher Dietary Laws 422
15.3.2 Halal Rules 423
15.4 Labeling of Food Flavorings 424
15.4.1 Bulk Labeling Requirements 424
15.4.2 Labeling for the Consumer 424
15.5 Summary 430
References 431

Chapter 16 Quality Control 433

16.1 Introduction 433
16.2 Analytical Tests 433
16.2.1 Overview of Physicochemical Tests 433
16.2.1.1 Natural Plant Materials 433
16.2.1.2 Essential Oils 434
16.2.1.3 Oleoresins 436
16.2.1.4 Plated or Dispersed Spices 436
16.2.1.5 Synthetic Chemicals 436
16.2.1.6 Finished Flavorings 437
16.2.1.7 Vanilla Extract 438
16.2.1.8 Fruit-Based Products 438
16.2.2 Generally Used Analytical Testing Methods 439
16.2.2.1 Density/Specific Gravity 439
16.2.2.2 Refractive Index 440
16.2.2.3 Optical Rotation 440
16.2.2.4 Alcohol Content 440
16.2.2.5 Residual Solvent 440
16.2.2.6 Particle Size of Emulsions 441
16.2.2.7 Volatile Oil 444
16.2.2.8 Surface Oil 445
16.2.2.9 Moisture Content 445
16.2.2.10 Gas Chromatography 447
16.2.2.11 Spectroscopic Analysis 449

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16.2.2.12 Microbiological Analysis 450
16.2.2.13 Electronic Noses 450
16.3 Sensory Analysis 450
16.3.1 Incoming Raw Ingredients 450
16.3.2 Finished Flavors 451
16.3.2.1 Sensory Evaluation 451
16.3.2.2 Changes in Finished Flavors with Age 453
16.3.2.3 Sample Rejection 454
16.3.3 Colorings 455
16.3.4 Scoville Heat Units 455
16.3.5 Summary 455
16.4 Adulteration Testing 456
16.4.1 Gas Chromatography/Mass Spectrometry 456
16.4.2 Chiral Compounds 458
16.4.3 HPLC 459
16.4.4 Carbon 14 Dating 460
16.4.5 Stable Isotope Ratio Analysis 461
16.4.6 Comments on Adulteration 463
References 463

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Part I

Flavor Chemistry

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7 Off-Flavors and Taints in Foods

7.1 INTRODUCTION

The major cause of consumer complaints to the food industry is an objectionable flavor [1]. Objectionable (or unacceptable) flavor may be due to incidental contamination of the food from environmental sources (e.g., air, water, or packaging materials) and is termed tainted, or may arise in the food itself as a result of degradation of some food component (e.g., lipid oxidation, nonenzymatic browning, or enzymatic action) and is considered off-flavored [2]. A third cause of unacceptable flavor, which is not generally recognized and is much more poorly defined, is when the flavor is not satisfactory due to the loss of characteristic flavor (evaporation or reaction of characterizing flavor components with the food itself) [3]. The food has neither been tainted nor developed an off-flavor. This source of an unacceptable flavor is difficult to diagnose and correct.

While there are numerous articles in the literature on objectionable flavor in foods, an article by Goldenberg and Matheson [4], despite being dated, and a book edited by Saxby [2] have to be singled out as indispensable references. There has been less published on off-flavors in foods in recent years than in the 70s and 80s. In recent years most of this type of work has been done in industry, and industry is reluctant to publish their problems or solutions. Thus, many of the references will be twenty or more years old. Unfortunately, they are as relevant today as then.

In the real world, one is concerned with determining if a food has an objectionable flavor (sensory methods), and what is causing it (combined sensory/instrumental methods) since this ultimately suggests how to eliminate it in the future. In this chapter we will consider the approaches for determining if a food has an objectionable flavor and its source. The instrumental aspects of this problem have been considered in Chapter 3.

7.2 SENSORY ASPECTS OF OFF-FLAVOR TESTING

Kilcast [5] has provided an excellent discussion on this topic and will serve as the primary reference for this chapter. The reader is most strongly advised to read his work for the detail he provides.

Some of the more frustrating aspects of the task of dealing with off-flavors are the difficulties one may have to determine if the food truly has an off-note, how to describe it, and determine the importance to a company. The event is usually initiated by a consumer complaint (but may also originate within the company, e.g., in the quality assurance laboratory). Consumers are notoriously poor at describing an off-note

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in a food. They know that product flavor does not meet their satisfaction but they generally lack the experience to accurately describe the off-flavor. The descriptions they provide come from their experiences, which vary from person to person. Thus, since the descriptions are so varied and inconsistent, it may be difficult to determine if a series of complaints are the result of the same problem. Thomson [6] illustrated this problem by having 32 untrained subjects describe the character of pork that had a boar taint. The 32 subjects used 22 different terms to describe a note that is generally characterized by the sensory descriptors of urinous, sweaty, or animal. The primary piece of information that one gets from a consumer complaint is that there is a flavor problem that some segment of the population finds unacceptable.

Unfortunately, there often is very poor consensus even among trained individuals. While the consensus improves as individuals become more familiar with an off-note, there still can be very poor agreement in descriptors used, or estimate of intensity of an off-note. This difference can come from experience of the panelists, innate ability to verbalize a sensory stimulus, and/or sensitivity to the stimulus.

Due to differences in sensitivity to a stimulus, some individuals may not detect an off-note while others may be greatly offended by it. Kilcast [5] illustrates this with a typical plot of the distribution of people who can detect a hypothetical aroma chemical as a function of concentration (Figure 7.1). One can see that at the sensory threshold concentration, 50% of the subjects can detect the chemical — this is the standard definition of detection threshold. If we go down the chart, we find that at a concentration ten times lower than the determined threshold, 5% of the population would be expected to still detect the aroma chemical; 95% cannot sense it. Thus, a subset of consumers may be sensitive to a given defect and have a legitimate complaint while the majority of consumers (or even company tasters) may not detect a problem.

Despite the problem in getting good sensory characterization of an off-note, this characterization is very important in determining the source of the problem and its

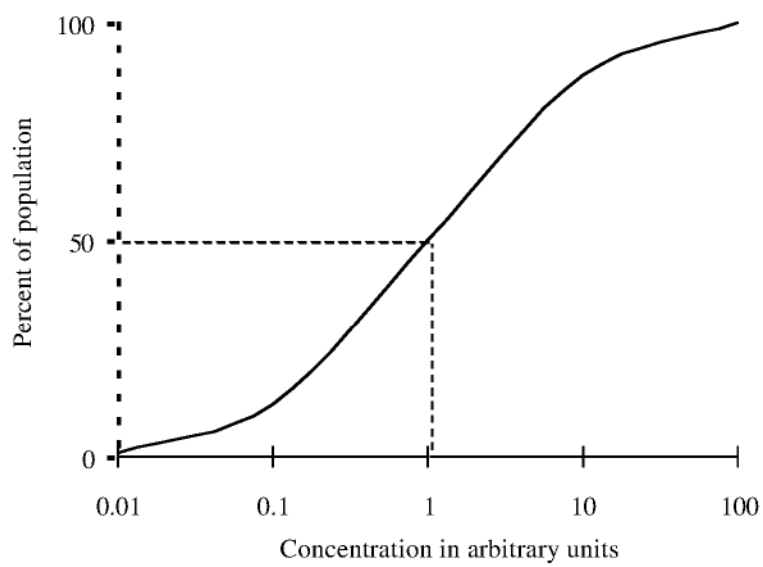

FIGURE 7.1 A typical relationship between concentration of an odorant and percentage of the population that can perceive it. (From Kilcast, D., *Food Taints and Off-Flavours,* M.J. Saxby, Ed., Blackie Acad. Prof., London, 1996, p. 1. With permission.)

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ultimate resolution. If the off-note is commonly a problem in the product (e.g., lipid oxidation), a sensory description by an experienced individual may be adequate to determine the source of the off-note and lead to its resolution. If not, the description is valuable in selecting and identifying the agents responsible for the offending flavor.

Kilcast [5] provides a brief overview of sensory methods employed in the field overall and then focuses on applying sensory testing to taints and off-flavors. He provides some guidelines for sensory testing that illustrate how this application of sensory differs from traditional sensory testing. His points can be summarized as:

1. When the source of the taint or off-flavor is known, use panelists that are sensitive to the objectionable compounds. He points out that it is difficult to have a sensory panel selected for every potential flavor defect, so one may have to select panelists generally known to be sensitive and reliable. (This is not so different from traditional sensory testing.)
2. If a sensory panel cannot be selected that is known to be sensitive to the flavor defect, use a large panel to improve the probability that panelists will be in the group that are sensitive. He suggests a minimum of 15 people (preferably 30) should be used for a triangle test. He cautions that replication is not useful in this type of testing — for if an individual is not sensitive to a flavor defect, testing again does not help.
3. Use a high sensitivity sensory test. He suggests using difference tests as opposed to profile-type tests. Difference tests are more rapid and require less training.
4. Maximize the amount of information gained from a sensory test. Traditional sensory testing has strict rules about getting a limited amount of information from a given test. Yet it is desirable to use a difference test and then ask for preference information (does the panelist prefer a given sample? He or she may like the objectionable sample) and how confident the panelist was in his or her choice.
5. Use statistical methods appropriate to off-flavor testing. This may require that the level of significance may be relaxed to 20% as opposed to the traditional 1 or 5% for accepting there is an off-flavor.
6. Irrespective of the testing outcome, look closely at minority opinions, especially if from typically reliable judges.

From personal experience, most food companies do not apply formal sensory testing to off-flavor problems unless litigation is involved. The purpose is then to establish that there is in fact a problem and establish the magnitude of it. If litigation is not involved, the sensory work is generally done informally by people familiar with the product or off-flavor testing in general.

7.3 TAINTS IN FOODS

This section is organized by means of contamination of a food. While it could have been organized around characteristic off-notes or chemical compounds, discussing sources may make the reader more aware of how foods can become contaminated. This could lead to better control of contamination.

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7.3.1 Airborne Sources

The air can be a very effective means of conveying a taint to a food product. Airborne contamination can enter a food in production (e.g., into milk through odors in the barn, or chicken meat/eggs in chicken coups), in manufacturing, through packaging contamination (will be discussed under packaging related tainting in Section 7.3.4), or during storage and distribution. We will provide brief descriptions of some of these mechanisms of contamination.

Taints can easily enter food products via the lungs of animals and thereby taint eggs, milk, or meat. The dairy industry has long recognized that milk is readily tainted in this manner [7–8]. A milk taint described as "barny" or "cowy" is known to come from cows housed in a poorly ventilated barn. This taint has become relatively rare due to better, cleaner cow environments. A second milk taint related to the environment is a "feed" taint. If cows are fed odorous feeds (e.g., silage, green grass, or oxidized feeds) within four to five hours of milking, the odor of the feed may be transferred to the milk.

An example of how poultry products can become tainted due to air contamination relates to finding 2,3,4,6-tetrachloroanisole in eggs and broiler meat. It was found that the wood shavings used in the hen house came from treated wood (phenol-containing) and microorganisms in chicken litter converted the phenols to the corresponding chloroanisoles [9]. The broilers and eggs assumed a musty taint from breathing the chloroanisole-contaminated air of the hen house.

The finished food products susceptible to airborne tainting are generally limited to goods that are baked and then cooled open to the environment. While the taint may enter the product during cooling, it may also be a problem once packaged. The packaging on these products typically presents little barrier to the transmission of volatile organic compounds. Table 7.1 lists some of the food taints reported in the literature due to airborne contamination. It is of interest that in one case, the source of contamination was 8 km from the food production facility. An examination of the compounds contributed to foods by airborne contamination in Table 7.1 shows that a variety of sources may be involved in the production of airborne taints.

Saxby [2] focused on the role of chlorophenols and chloroanisoles in this context. He cited examples where di-, tri-, tetra- and pentachlorophenols/anisoles were found in wooden flooring, pallets, or shipping containers. Much of the soft timber in the U.S. and Britain (perhaps the world) was at one time treated with chlorophenols (technical grade pentachlorophenol) as antisapstain agents [2]. Therefore, shavings as byproducts or the wood product itself (as noted above) contained significant levels of these chemicals. Chlorophenol contamination has been occasionally traced to accidental contamination through spillage. A third route is that they may arise due to treatment of wood with hypochlorite. In this latter case, naturally occurring phenols in the wood (or paint) may be converted to chlorophenols by hypochlorite treatment [11]. Chloroanisoles may exist as such but generally arise due to microbial conversion of the corresponding chlorophenol [9].

Chloroanisoles and chlorophenols are responsible for musty and medicinal taints, respectively, in foods. If one searches the literature, the majority of reported cases of contamination with these chemicals are in Europe, Australia, and Japan; there are

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TABLE 7.1
Examples of Airborne Contamination of Foods

Food	Compound(s) or Sensory Character	Mechanism of Entry	Ref.
Biscuits	Herbicides (Chlorophenols)	Carried by wind from a herbicide factory (8 km)	4
Biscuits	Tar	Tar boiler gave smell due to gaps in the wall	4
Chocolates		Anise seed-type odor from adjacent factory	4
Cakes	Catty off-flavor	Reaction of ketones with sulfur compounds in food	4
Wrapped bakery goods	Diesel oil fumes	Depots, loading docks, warehouses. Fumes penetrate packaging to contaminate product	4
Cakes, biscuits, meat pies, chocolate, etc.	Paint	Painted floors, walls, etc.	4
Biscuits	Oxidized oil	Oil used for spraying biscuits was oxidized. Contaminated product and other products	4
Cocoa mass	Chlorophenols—musty character	Contaminated shipping container	10

few cases reported in the U.S. This does not mean that tainting by them has not occurred in the U.S., but simply we have few reported cases of such contamination. The U.S. food industry is less likely to make public the occurrence of off-flavor problems or their resolution than European food processors.

An example of chloroanisole/chlorophenol contamination involved the occurrence of a musty taint in dried fruit [1]. It was found that the outer fiberboard cartons were made from recycled waste paper which contained high levels of chlorophenols that were converted (microbially) to the corresponding chloroanisoles during shipping. These chloroanisoles permeated the cartons and contaminated the dried fruit. Interestingly, when the industry changed to virgin fiberboard, the problem decreased but did not disappear. Tindale and Whitfield [12] found an incident where cartons were stored in an area that was cleaned with disinfectants high in available chlorine. The chlorine reacted with the degraded lignin in the board to produce chloroanisoles.

Chloroanisoles are frequently responsible for the tainting of wines. Cork taint is a well-known problem to the wine industry. This topic will be discussed under a subsequent heading titled "Microbial Off-Flavors"(Section 7.6) since this specific tainting is generally associated with the microbial conversion of phenolic substances in cork to chloroanisoles.

A most challenging tainting problem was the occurrence of a "catty" note in some foods. "Catty" is used to describe the odor of tom cat urine. Pearce et al. [13] found that an unsaturated ketone, mesityl oxide, (4-methylpent-3-en-2-one) can react with sulfur compounds in a food, particularly hydrogen sulfide, to form a compound with a catty odor. This compound has been identified as 4-mercapto-methylpentan-2-one [14]. This reaction has been responsible for several tainting events in the industry. In one case, the mesityl oxide was a trace component (0.5%) in a solvent

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for a polyurethane paint that was applied in a meat chill room [15]. While xylene and pentoxone (major components of the paint solvent) were found in the beef carcasses, 4-mercapto-methylpentan-2-one was found to be responsible for the offensive (catty) taint. Apparently the mesityl oxide reacted with sulfur-containing amino acid degradation products in the meat to form the catty note. The unique difficulty this taint provides is that the offending compound is not present in the environment but is formed in the product. Mesityl oxide has been linked to other occurrences of taints including a variety of canned goods [16]. This taint will be discussed under packaging contamination later in this chapter (Section 7.3.4).

Airborne contamination is generally exceedingly difficult to identify as the source of product contamination. This is partly due to the fact that the taint cannot be reproduced in the product at will for studying. This is contrasted with an off-flavor arising from lipid oxidation, for example, during storage of food. The oxidized off-flavor can be reproduced and subjected to analysis, identification, and eventual management. The airborne taint may occur randomly, depending on the production schedule of the offending source of the taint, wind direction and speed, production schedule of the food processing plant, etc. The offending chemicals also typically have very low sensory thresholds, so they are difficult to isolate and identify by established analytical techniques. An example is the sensory threshold of 6-chloro-*o*-cresol in biscuits (5×10^{-5} ppm). This compound was found to contribute a disinfectant taint to the biscuits [4].

7.3.2 Waterborne Sources

Large amounts of water are typically used in most food processing operations. If this water contains significant quantities of tainting components, the potential exists for the taint to be transferred into the food being processed. However, this is generally not a problem unless the water becomes a significant component of the food product or the taint is very intense. Water becomes a significant part of the food if the food is reconstituted or diluted with water prior to processing (e.g., spray drying of cheese or reconstitution of concentrated juices for retail sale), or if direct steam injection is used for product heating.

Taints may also be introduced into a food via water in an indirect manner, e.g., the absorption of taints from habitat water by fish and shellfish. There has been a substantial amount of work on taints in fish and shellfish due to water contamination [17]. Fish are very susceptible to absorbing chemicals from their environment resulting in a taint. The taint may come from water pollution (chemical) or bacterial growth in the water.

In terms of fish tainting from pollution, Vale et al. [18] and Shipton et al. [19] have reported finding a "kerosene" taint in mullet. The tainted mullet were found to contain high levels of hydrocarbons from polluted waters. Bemelmans and den Braber [20] reported an iodine-like taint in herring from the Baltic Sea. They traced this taint to o-bromophenol. While the source of the o-bromophenol was not determined, it is possible that it came from industrial water pollution.

The ease with which chemicals are absorbed into fish (particularly fresh water fish) is a major problem facing aquaculture. While water pollution is not typically

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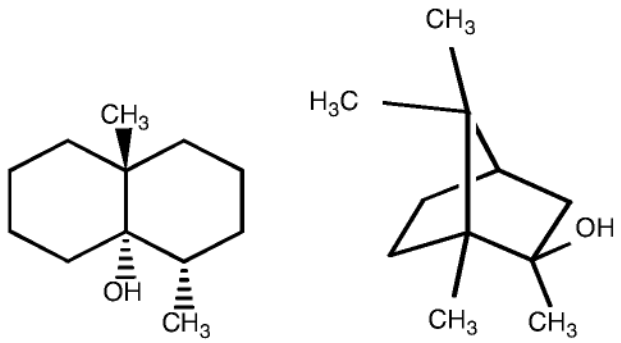

Geosmin Isoborneol

FIGURE 7.2 Geosmin (*trans*-1,10-dimethyl-*trans*-9-decalol) (left) and 2-methylisoborneol (right), causes of musty, earthy taints in water.

a problem, the growth of bacteria or tainting algae in the pond does present taint problems. Lovell and Sackey [21] reported that catfish developed an objectionable earthy-musty taint within two days when holding in tanks containing odor-causing algae. This taint was associated with the presence of *Anabaena*, *Ceratium*, and *Pediastrum* algae [22–23]. The tainting component was likely geosmin (Figure 7.2, trans-1,10-dimethyl-trans-9-decalol) and/or 2-methylisoborneol (Figure 7.2) since these compounds have been identified in muddy tasting fish [24]. Geosmin and 2-methylisoborneol have a very low odor thresholds, 0.05 ppb in water and 6 ppb in fish) and 0.029 ppb in water, respectively, which contribute greatly to their tainting potential. Magligalig et al. [25] demonstrated how quickly water taints can be transferred to fish muscle. They found an exposure of 10 min to 15 ppm 2-pentanone or dimethyl sulfide resulted in significant taint. Recent work by Jirawan and Athapol [26] on the tainting of Nile tilapia by geosmin showed that this earthy taint is acquired in less than two hours when fish are placed in water containing only 5 ng/l geosmin. The fish had to be held in clean water at least 16 days to then eliminate this taint. This very rapid absorption of odorants makes fish very susceptible to picking up taints from their environment.

Some taint problems appearing in the literature from water sources are presented in Table 7.2. The causes of these taints are quite diverse.

7.3.3 Disinfectants, Pesticides, and Detergents

Disinfectants, pesticides, and detergents comprise a group of chemicals that occasionally contribute taints to foods. This may be the result of inadequate care in their application by the end user or errors on the part of the cleaning compound manufacturers. The summary of taints observed at Marks and Spencer (Table 7.3) by Goldenberg and Matheson [4] provides a useful illustration of how these chemicals may enter a food and produce a taint. They noted several incidences of taints in food due to detergent contamination, which are not included in this table.

The compounds responsible for these types of taints are often phenols or chlorinated phenols. As noted earlier, phenols can contribute taints to foods but their

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TABLE 7.2
Taints Due to Waterborne Contaminants

Food	Contaminant	Method of Transmission	Ref.
Canned fruits and canned vegetables	Chlorophenols	Chlorinated water used for cooling lithographed cans subsequently used as boiler feed water. Chlorine reacted with phenol-based lithographic print. Resulting chlorophenols volatized in steam subsequently used for heating brines and syrups by direct steam injection	4
		Peas transported to cannery in open steel troughs on newly tarred road. Peas contaminated by tar covered stones flying up from road. Peas contaminated by tar blanched in chlorinated water and chlorophenol	4
Canned products	Boiler water treatment chemicals	Can cause off-flavors if live steam is used for direct steam-injection for heating	4
Spray-dried cheese	Disinfectant?	Source of off-flavor water powder borne. Problem solved by use of activated carbon water filter	4
Beer	Iodinated trihalomethanes	Water in city reservoir was low, resulting in decay of organic matter. Combination of chlorination of water, organic matter, and geological abundance of iodine and bromine	27

relatively high sensory thresholds make them less problematic than their chlorinated counterparts. The chlorinated phenols are soapy, medicinal, or disinfectant in character and have very low sensory thresholds (often < 1 ppb). While the chlorinated phenols may occur as a contaminant in a disinfectant or cleaner, they are also formed from the chlorination of free phenols. Saxby et al. [28] have demonstrated that o-cresol can react with hypochlorite to form 6-chloro-o-cresol (0.05 ppb threshold in biscuits) and the chlorination of mixed phenols typically yields significant quantities of 2-chlorophenol (threshold of about 2 ppb in milk). There are several examples in the literature illustrating how this reaction has caused taints in foods. One example was where a meat processor was washing carcasses with chlorinated water. The water, on occasion, contained trace levels of phenols that were converted to the chlorinated phenols during canning of the meat [2]. Another example involved the transfer of beer through Nylon tubing [30]. The Nylon had been sterilized with a hypochlorite solution. Apparently, the hypochlorite formed a N-chlorinated derivative of the Nylon, which then served as a good chlorinating agent for phenols in the beer. Disinfectant taints in coffee sold in vending machines may have been formed by similar mechanisms [2].

7.3.4 Packaging Sources

Food packaging materials are frequent contributors to taints found in food products [31]. Virtually the only food packaging material that will not potentially contribute a taint to foods is glass. Since food is not sold in glass ampoules (i.e., sealed entirely

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TABLE 7.3
Taints Due to Disinfectants, Pesticides, and Detergents

Food	Contaminant	Method of transmission	Ref.
Cake	Chlorophenol	Due to sultanas, contaminated on board ship by chlorophenol. On a previous voyage, holds had been used for cargo of herbicide containing 0.5% chlorophenol as impurity. Hold washed out but timber still impregnated. Finally timber parts had to be destroyed	4
Spray dried milk	Chlorophenol	Water contaminated with chlorophenol-cleaner	4
Canned raspberries	Pesticide	"Catty" taint due to mesityl oxide in solvent	4
Pork	Disinfectant	Preventive spraying with disinfectant in slaughterhouse against spread of swine vesicular disease	4
Tomatoes	Pesticide	Appeared to be chlorophenol type contaminant	4
Smoked bacon	Disinfectant	Hypochlorite used as sterilizing agent; excess used and inadequately washed off. Chlorine combined with phenolic compounds present in smoked bacon to give chlorophenol. Bacon uneatable. Sterilizing agents containing chlorine should not be used where smoked meats or fish are handled, unless special precautions are taken	4
Beer	Permethrin (pesticide)	High levels of pesticide on malt carried taint into beer	29

in glass) but needs a closure of some type, a small potential exists for food contamination due to this closure even for glass packaging.

Some occurrences of food contamination by packaging materials resulting in tainting are presented in Table 7.4. The contamination seldom comes from the major packaging component. Most often the taint is due to trace amounts of residual solvents, unanticipated contaminants, or the degradation of some packaging component. This can involve the bulk packaging material (e.g., styrene monomer), trace contaminants in the packaging material (e.g., ethyl benzene in plastics or residual inks in recycled paper), solvents used in printing inks, or solvents in adhesives. In some cases the contaminant responsible for the taint is a normal constituent of the packaging system but has not been adequately removed by the processing. This typically occurs due to pushing production too fast. The alternative is that the solvents may, on occasion, be contaminated with higher boiling constituents that are not removed by established process conditions. Tice [31] has presented a good discussion of printing processes and their potential to contribute taints to foods, which will be summarized in the following section.

A variety of printing processes and thus, materials are used to print food packages. One printing system, litho printing, uses a petroleum-based solvent system to carry the inks (resins). While the petroleum-based solvent is chosen to be low in odor, inadequate solvent removal or trace contaminants can be problematic. The

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TABLE 7.4
Taints Due in Foods Due to Packaging Materials

Food	Packaging Material	Source and Method of Transmission
	Printing Systems	
	Printed packaging film[47]	Musty odor in printed film. Found to be due to 4,4,6-trimethyl-1,3-dioxane. Formed by reaction of printing components
Yogurt containers[a] and confectionery[4]		Inadequate curing of inks used in printing
Chocolate cakes[4]	Coated paper-print on one side	Inadequate curing of inks used in printing
Cook in the bag ham[48]	Plastic wrap	Mesityl oxide formed from the dehydration of diacetone alcohol — a component in red printing ink — catalyzed by ionomer layer in bag
	Paper	
Cakes, biscuits, chocolate[4]	Chipboard	Off-flavor due to either preservatives added to repulped waste paper or compounds in print in waste paper
Doughnuts[a]	Coated paper divider plus cellophane overwrap	Inadequate removal of hydrocarbon solvents used to apply divider coating
Cold cereals [43,44]	Glassine liner coupons	Residual solvents
	Plastics	
Orange and lemon drinks[4]	PVC bottle	Mercaptide stabilizer used in PVC-split off in processing of beverage
Chocolate and lemon cookies[45]	Polystyrene trays wrapped in printed cellophane	Residual styrene monomer
TV dinners[a]	Polystyrene trays	Residual styrene monomer
	Laminates	
Fruity soft drinks[45]	Polyester/aluminum foil/polyethylene laminate	Residual toluene in laminate adhesive-faulty drying process
	Metal-Paper	
Refrigerated doughs[a]	Paper tube with metal caps	Residual hydrocarbon solvents in triglycerides used to lubricate metal caps to facilitate packaging operation
	Metal	
Canned pork products[4]	Lacquered can	"Catty" taint due to use of lacquer to cover side seam to prevent blackening. Solvent used contained mesityl oxide which reacted with sulfur compounds in food to cause taint
Refrigerated milk beverage[a]	Lacquered can	Trace contaminant (isophorone) in solvents used in lacquer which did not "flash off"
Beer[46]	Can	Lubricants used in can manufacture

[a] Reineccius, unpublished.

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resins themselves can also be responsible for tainting. These resins are alkyd types and thus can oxidize in the drying process to form unpleasant aldehydes and ketones.

An alternative printing process, flexographic printing, does not use hydrocarbon solvents but alcohols (ethanol, isopropyl alcohol, and n-propyl alcohol) or esters (ethyl acetate, isopropyl acetate and n-propyl acetate) as solvents. Ethyl acetate and ethanol are the most commonly used solvents. The ink formulation will also contain plasticisors and a slow drying solvent (e.g., propylene glycol monomethyl ether). The resins are polyamides, cellulose nitrates, or acrylics. The tainting potential is primarily from residual solvent (ethyl acetate) since the resins are quite stable. Gravure printing systems use similar solvents and inks as flexographic printing, however, water-based systems are generally used for food packaging applications.

The solvent-less UV printing systems use acrylate monomers with a photo initiator such as benzophenone. The acrylate monomers are polymerized under UV light to form a hard, dry print. Residual odors in this printing system would likely come from unpolymerized acrylate, reaction byproducts, and residual benzophenone. Benzophenone has a geranium-like odor. Potential byproducts that may taint a food include benzaldehyde and alkyl benzoates.

In recent years, there has been a movement to eliminate organic solvents from food manufacturing operations. Various packaging components may be delivered in a water-based or a solvent-less system, the latter employing ultraviolet irradiation to cure and polymerize the resin components (photo initiators are incorporated into the resin). This change has resulted in less food tainting with the usual volatiles but has resulted in a wide range of new taints. Since the new water-based packaging materials (e.g., coatings, inks, overlays, and adhesives) are not *solubilized* in an organic solvent but *dispersed* in an aqueous system, new chemicals are needed to keep these packaging components dispersed. The solvent-less UV printing systems may afford fewer possibilities for contamination, but yet they employ new materials (monomers and photo sensitizers), which again offer possibilities for tainting.

One example of contamination from the newer materials is an off-taste that was found in cookies that were packaged in a clear film. The cookies gave a mouth-tingling, numbing effect where they touched the clear packaging. After substantial research effort, it was found that the packaging was contaminated with spilanthol. Spilanthol is a very unusual compound that is found in a rare essential oil, jambou. This is the first time spilanthol was found as a food contaminant. The implications of this finding are noteworthy.

In the past, a limited number of known solvents (and common contaminants) were likely to be found in food packaging materials. With the advent of water-based delivery systems, the potential list of contaminants has been expanded upon greatly. More significantly, these materials are likely to be nonvolatile and thus not measurable by standard methods used in quality assurance laboratories. Traditionally, food companies have established standards for volatile residues and the testing methods are routine [31]. Unfortunately, there are no routine or all inclusive methods for the testing of nonvolatiles. Thus, the food industry is starting to find new packaging related tainting problems and lacks suitable methods for their study.

There are three examples of food tainting from paper-based packaging materials listed in Table 7.4. Paper or paper-based products come in contact with or form the

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outer package of a host of food products. For example, liquid dairy products (plastic/board composite), breakfast cereals (variable inner barrier but board exterior), flour (kraft bag), butter and related products (greaseproof paper), chocolates (glassine), cookies (kraft wrap), numerous dry mixes (inner barrier plus outer board), and frozen foods (outer box) have either direct contact or form a secondary package. Even when a contaminated paper product forms a secondary or outer barrier, tainting of the food is possible since the primary, or food contact package, is seldom a good barrier to organic volatiles.

Paper packaging may become tainted in several ways. Paper generally contains various sizing agents, fillers, defoaming agents, and slimicides. The paper may be chemically bleached to provide whiteness. Paper surfaces may also be coated with china clay or calcium carbonate imbedded in a synthetic resin (e.g., a styrene/butadiene copolymer or a styrene/acrylate copolymer) to give the desirable appearance, print surface, or durability [31]. Often we see multilayered packaging where a paper is laminated onto a plastic or foil layer (e.g., milk cartons). Lamination requires the use of adhesives. Each of these packaging materials may be printed upon and further coated with a varnish to give the desired surface properties. Thus, there are many opportunities for the paper-based product to become contaminated and for this contamination to reach the food product. It should be noted that paper-based materials may become more odorous on storage as opposed to less odorous. This is related to the oxidation of resins or lipids in the paper.

The use of plastics in food packaging has become extremely pervasive. Plastics have replaced paper, glass, and metal packaging in a host of food packaging applications. Plastics always begin with the polymerization of monomeric materials. Typically very small quantities of these monomeric materials remain in the polymer, and thus offer little opportunity for food tainting unless the monomer has a very low sensory threshold and is easily recognized as foreign to a food product, e.g., styrene. In addition to the base polymer, additives such as antioxidants, stabilizers, slip agents, antistatics, and colorants may be present in the finished polymer. Each of these components may bring a taint to a food product [31].

Some of the polymers used in food packaging include: PVC (polyvinyl chloride), PET (polyethylene terephthalate), PC (polycarbonate), polystyrene, and PE (polyethylene — either low or high density). Of these polymers, PVC was found in a study of mineral waters to provide the least off-notes. Other than one occurrence (noted in Table 7.4) where a PVC stabilizer was found to produce a taint in beverages, there are *no reports* in the literature of taints by this material. PET is widely used for packaged liquids (mineral water, carbonated beverages, ciders, and beers) and ovenproof food trays. PET is based on the polymerization of either terephthalic acid or dimethylterephthalic acid and ethylene glycol. Both of these monomers have little odor so they cause little problem in terms of tainting. However, a significant amount of acetaldehyde may be formed during the manufacturing or processing of the PET into a final package. This potential is well recognized in the industry, and therefore specifications are in place for its permitted levels. The polyolefins (various low and high density products) are the most common of all food packaging polymers [31]. The odors associated with the polyolefins are seldom from residual monomers since hydrocarbons have high sensory thresholds but arise from the oxidation of the

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polymers. Koszinowski and Piringer [32] found that two components of heated low density polyethylene, 1-nonene and 2-ethyl-hexanol, are readily oxidized to 2-nonenal and 1-heptene-3-one, respectively. Both of these oxidation products have very low sensory thresholds and have been identified as tainting compounds in LDPE-coated paper board. The packaging most commonly associated with food taints is polystyrene. Styrene can be present in the polymer as a result of incomplete polymerization or thermal degradation. Styrene monomer readily migrates to a food and causes a very characteristic easily recognized plastic taint.

Several other cases of packaging related taints are listed in Table 7.4. These taints involved laminates, metal-paper combinations, and metal cans. Laminates need adhesives to bind the layers together, and the adhesives were contaminated with residual solvents. The paper-metal tube taint involved closing the package before the solvent used to apply a triglyceride lubricant had an opportunity to evaporate from the metal tube lid. The solvent was sealed inside the tube with the product. The final examples in Table 7.4 are tainting from metal cans. In each of these cases, the contamination came from the lacquer lining used on the inner can surface. Foods must be protected from the can and the can from the food. This necessitates the application of a polymer film on the inside of the can surface. Any residual solvent or trace contaminants in this lacquer would potentially taint the can contents.

A final consideration within this topic heading is the loss of flavor either to absorption of the flavor by the packaging (scalping) or its loss through the packaging to the environment (permeation). These processes do not result in a tainted product, but the loss of desirable flavor can expose off-notes to decrease product quality or simply result in a product lacking in flavor.

One has to recognize that permeation can be a two-way process: the loss of flavor from a food product to the environment or the intrusion of odors in the environment into the product. Intrusion of off-odors from the environment may result in tainting of foods and has been discussed earlier in Section 7.3.1 Airborne Sources. One should recognize that food packages range in quality from dust covers, i.e., those packages that present little or no barrier to moisture, oxygen, or organic vapor permeation, to those that are very good barriers to all of these potential migrants. Unfortunately, packaging materials, i.e., polymer-based materials, that offer a good barrier to organic vapors are expensive and seldom used in the industry. Thus, if one is using a polymer-based food package, it is likely to be involved in flavor scalping and offers little to no protection against tainting by permeation.

The amount of scalping or permeation one experiences is related both to the properties of the aroma compounds (polarity and molecular size) and those of the packaging polymer (chemical makeup). Small aroma molecules diffuse better through polymers as do aroma compounds of similar polarity as the packaging polymer. This combination of solubility and molecular dimensions determine the diffusion of a given aroma molecule into and through a package. Since there are numerous references on this topic including symposia proceedings [33–35,29], we will not deal with the details of this process. Our point is that flavor scalping occurs and can result in significant flavor loss and potential flavor imbalances (selective scalping of parts of a flavor). These effects may result in a food product being perceived as being off-flavored when in fact it is not: the food has lost its characterizing flavor.

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7.4 OFF-FLAVORS DUE TO GENETICS OR DIET

It is obvious to each of us that genetics and diet have a strong influence on the flavor of both plants and animals. We appreciate the differences in flavor found between the numerous varieties of apples, grapes, oranges, etc. We do not, however, accept differences in meat flavor due to either genetic or diet differences between animals. We have come to expect all meats (of the same species) to taste the same. For example, if one beef steak tastes different from another, we consider it to have an off-flavor.

7.4.1 Genetics

Two animal species in which genetics plays an important role in producing an off-flavor are swine and ovine. Swine products (pork) occasionally contain an off-flavor characterized as early as 1936 by Lerche as being perspiration or onion-like in character. More recent sensory studies have noted that this off-note will vary depending upon the animal. Sensory descriptors including urine, sweaty, chemical, rancid, turpentine, stable, viscera, pig/animal, manure, and naphthalene-like have been used to describe this off-note [36]. This off-flavor (most commonly referred to incorrectly as a taint) occurs primarily in sexually mature uncastrated male hogs (boars). It has received the most attention of any off-flavor or taint in the literature (194 references in the literature from 1969 to 2002). This is due to its severe impact on the economics of producing pork. Barrows and gilts (no off-flavor) do not gain weight nearly as quickly or efficiently as a boar (uncastrated animal). Thus, substantial research has been directed towards finding means to test for the defect at the time of slaughter to isolate the meat from off-flavored animals or find ways to prevent its occurrence in the male animal [37]. Prevention has typically focused on vaccination or breeding programs.

It is of interest that 46% of the male population and 92% of the female population can smell this off-odor [38], and thus females are most commonly offended by this off-note. It is caused by some combination of 5-α-androst-16-ene-2-one [39], 3-hydroxy-5-α-androst-16-ene [40], indole, skatole [41] and potentially, 4-phenyl-3-buten-2-one [42]. Despite the large amount of research devoted to this sensory defect, the problem persists throughout the industry.

Like swine, the mature ovine animal is also considered to have an off-flavor. (However, this is not a true off-flavor in the sense that the offending odor is a normal characteristic of all mature ovine). This off-flavor is termed "mutton flavor" and is most commonly described as being "sweaty-sour" in nature [49]. The source of this off-flavor has been traced to the unique synthesis of 4-methyl octanoic and 4-methyl nonanoic acids in the mature animal [50]. Jamora et al. [51] have provided a review on this defect and the factors that influence its intensity in meats.

7.4.2 Diet

Animal diet can have a pronounced influence on the flavor quality of meats, especially in nonruminant species. Earlier in this chapter, mention is made of musty

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taints in broilers due to the inhalation of chloroanisoles. While the wood litter was primarily responsible for chloroanisoles in the broiler, Bemelmans and Noever de Brauw [52] also found chloroanisoles in poultry feed. Thus, a tainted diet may also contribute musty taints to the poultry meat. Taints may also enter poultry meat via the diet from the consumption of significant levels (>2%) of highly unsaturated fats [53]. For example, turkeys fed 2% tuna oil develop a fishy defect when cooked [54]. The fishy character was found to be due to oxidation of the highly unsaturated fatty acids during cooking of the turkey. Poultry very readily deposit dietary fat. Goldenberg and Matheson [4] reported similar tainting problems with the feeding of fish oil to hogs. They recommended limiting fish oil to less than 1% of the animal diet.

Tainting of fish via the diet has already been partially discussed in this chapter. Mention was made of muddy taints being the result of the fish eating certain algae *(Anabaena, Ceratium,* and *Pediestrum).* Petroleum-like taints have been found in fish consuming *Limacina helicina* as a major part of their diet [55]. Earthy taints have been associated with the consumption of certain *Actinomyces* species [56]. In addition to off-flavors arising in fish from certain algae in their diet, may also occur if the fish are fed turkey livers or cereal. Maligalig et al. [57] found turkey liver notes in pond-reared catfish after 19 days of feeding livers and cereal notes in the fish following 33 days on a cereal diet. Minimizing taints in fish farming operations is a challenging task.

The contribution of bromophenols to seafood flavor and tainting has been clarified [58–59]. It appears that bromophenols are natural components of seabed algae and other sea creatures (e.g., worms). They are considered to contribute to the characteristic desirable flavor of the seafood when at low levels. However, high levels of these bromophenols in the diet result in tainted product. Boyle et al. [59] found about 3 ppb was a normal bromophenol level in seafoods. However, levels as high as 38 ppb in herring, 30 ppb in salmon, and 100 ppb in shrimp have been found. These levels would be expected to produce taints. Whitfield et al. [58] found 2,6-dibromophenol as the primary source of taint in prawn with 2- and 4-bromophenol, 2,4-dibromophenol, and 2,4,6-tribromophenol as secondary contributors. Whitfield et al. [60] identified various bromophenols as the source of an iodine-like taint in blackfish (Australian origin).

There are numerous studies showing how diet can produce taints in lamb [17]. Park et al. [61] reported on a sickly and nauseating aroma in lamb that had been fed rape. They postulated that the natural glucosinolates of the rape were metabolized to aliphatic sulfides, disulfides, or mercaptans that caused the taints. Park et al. [62] also noted that feeding vetch had an influence on lamb flavor. The vetch-fed lambs had a sweeter and stronger meat flavor. Melton [63] has provided a review of the effect of diet on the flavor of red meat.

Jeon [7] has presented a summary of taints found in milk as a result of pasture weeds. He has noted that land cress (tainting compound, benzyl mercaptan), penny cress (also known as French weed or stinkweed, tainting compound, allylisothiocyanate), penny royal (tainting compound, pulegone), pepper grass (tainting compound, indole), and onion/garlic (tainting compounds, di-n-propyl sulfide, isopropyl mercaptan, and propionaldehyde) have been found to produce tainted milk.

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7.5 OFF-FLAVORS DUE TO CHEMICAL CHANGES IN THE FOOD

Foods contain a number of constituents that may either react with other constituents or simply degrade into off-flavor components. The major reactions leading to off-flavors include lipid oxidation, nonenzymatic browning, enzymatic changes, and photo-catalyzed reactions. This section will present some examples of off-flavors arising in foods from these reactions.

7.5.1 Lipid Oxidation

Lipid oxidation (LO) is with few exceptions (e.g., American milk chocolate and deep fat fried flavor) a detrimental reaction. This reaction is probably the most common source of off-flavors in foods during storage. Off-flavors resulting from LO are described as being: cardboard, beany, green, metallic, oily, fishy, bitter, fruity, soapy, painty, rancid, grassy, buttery, tallowy, or oxidized [64]. The most common descriptors are stale (reaction has not progressed to stage it is recognized yet), cardboard (mild oxidation), tallowy (moderate), metallic, painty, and/or fishy (severe). The sensory character depends upon the degree of oxidation as well as the food product. While dry foods are most susceptible to off-flavors from lipid oxidation, nearly all foods are susceptible to deterioration via this reaction. Grosch [65] and Kochhar [64] have presented excellent reviews of lipid oxidation as it relates to off-flavor production. The reader is recommended to either of these references for detail beyond what is presented here.

Lipid oxidation typically involves the reaction of molecular oxygen with unsaturated fatty acids via a free radical mechanism. The mechanism may be presented as follows:

Initiation

$$R_1H \rightarrow R_1\bullet + H\bullet$$

$$R_1H + O_2 \rightarrow R_1OO\bullet + H\bullet$$

Propagation

$$R\bullet + O_2 \rightarrow R_1OO\bullet$$

$$R_1OO\bullet + R_2H \rightarrow R_1OOH + R_2\bullet$$

Termination

$$ROO\bullet + R\bullet \rightarrow ROOR$$

$$R\bullet + R\bullet \rightarrow RR$$

$$ROO\bullet + ROO\bullet \rightarrow ROOR + O_2$$

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where RH is unsaturated lipid; R·, lipid radical; and ROO·, lipid peroxy radical.

Alternatively, lipid oxidation can be initiated by light (photosensitized oxidation). This process is presented below.

$$\text{Sens} \rightarrow {}^{1}\text{Sens}^{*} \rightarrow {}^{3}\text{Sens}^{*}$$

$${}^{3}\text{Sens}^{*} + {}^{3}\text{O}_2 \rightarrow {}^{1}\text{O}_2^{*} + {}^{1}\text{Sens}$$

$${}^{1}\text{O}_2^{*} + \text{RH} \rightarrow \text{ROOH} \rightarrow \text{decomposition to form free radicals that continue autoxidation}$$

The Sens is a sensitizer that acts as a means of transferring light energy to oxygen, in essence, activating the oxygen so it is able to directly attack an unsaturated fatty acid. The sensitizer is often chlorophyll, pheophytin, myoglobin, or erythrosine [66–67]. The excited singlet oxygen is extremely reactive, reacting with methyl linoleate, for example, 10^3 to 10^4 times faster than normal oxygen. The hydroperoxide formed will decompose, resulting in free radicals thereby initiating free radical autoxidation. Thus, light serves as a good initiator of oxidation in foods.

Foote [68] has proposed a slightly different mechanism for the initiation of photo-sensitized oxidation. In this mechanism, the sensitizer forms an active complex with oxygen, which then attacks the fatty acid. This process is shown below and is believed to function in the riboflavin sensitized oxidation process.

$${}^{3}\text{Sens} + {}^{3}\text{O}_2 \rightarrow {}^{1}[\text{Sens-O}_2]$$

$${}^{1}[\text{Sens-O}_2] + \text{RH} \rightarrow \text{ROOH} + {}^{1}\text{Sens}$$

One can see that this is a self-propagating reaction, i.e., once started it continues itself. It is interesting that an initial radical will produce about 100 hydroperoxides before its termination occurs [69,70].

Off-flavors arise from LO via secondary reaction products. Lipid hydroperoxides are very unstable and break down to produce short chain volatile flavor compounds. Keeney [70] has shown some of the general reactions leading to flavor production:

(A)
$$\begin{array}{ccccc} \text{R–CH–R} & \rightarrow & \text{R–CH–R} & + & \bullet\text{OH} \\ | & & | & & \\ \bullet\text{O–OH} & & \text{O}\bullet & & \end{array}$$

(B)
$$\begin{array}{ccccc} \text{R–CH–R} & \rightarrow & \text{R–CH} & + & \text{R}\bullet \\ | & & \| & & \\ \text{O}\bullet & & \text{O} & & \end{array}$$

(C)
$$\begin{array}{ccccc} \text{R–CH–R} & + & \text{R}_1\text{H} & \rightarrow & \text{R–C–R} + \text{R}_1\bullet \\ | & & & & \| \\ \text{O}\bullet & & & & \text{O} \end{array}$$

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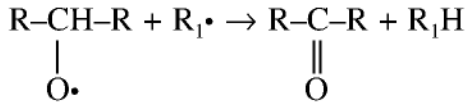
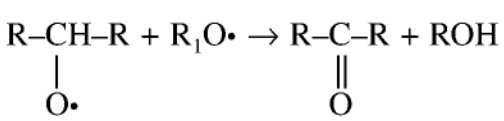
(D)



(E)



Steps B through E result in the production of short chain saturated or unsaturated aldehydes, ketones, and alcohols. These primary reaction products can undergo further oxidation if unsaturated or secondary reactions to yield a host of off-flavor volatiles (see Figure 7.3). These final products are generally aldehydes, ketones, acids, alcohols, hydrocarbons, lactones, or esters (Table 7.5). The unsaturated aldehydes and ketones have the lowest sensory thresholds and are, therefore, most often credited with being responsible for oxidized flavors. The metallic taint in oxidized butter, for example, has been attributed to oct-l-ene-3-one [71].

An insight into the factors that enhance lipid oxidation will help one understand why some food products develop oxidized off-flavors very rapidly. A primary consideration is the number and type of double bonds in a fatty acid. The relative rates of oxidation for arachidonic, linolenic, linoleic, and oleic acid are 40:20:10:1. In some food applications, the highly unsaturated fatty acids, e.g., linolenic acid, must be selectively hydrogenated in order to permit an acceptable shelf life. For example, the presence of any linolenic acid in a dry coffee whitener would limit shelf life to less than three months.

Trace metals, particularly copper, cobalt, and iron, greatly increase the rate of LO and influence the direction of peroxide decomposition [72]. These metals function both to reduce the induction period and increase reaction rate by decomposing hydroperoxides. Trace levels of these catalysts, e.g., as little as 0.3 ppm iron or 0.01 ppm copper, will result in prooxidant effects [73]. Iron may exist in foods in the free form or as a part of an enzyme (contain organically bound haem, Fe^{+2} or haemin, Fe^{+3}). Enzymes containing haematin compounds include catalase and peroxidase (plant tissues) and haemoglobin, myoglobin, and cytochrome C (animal tissues). While heat treatment results in denaturation of the enzymes, it frees the iron to greatly enhance its catalytic properties. This is particularly relevant in the formation of warmed-over off-flavor in cooked meats.

There is substantial literature demonstrating a strong relationship between water activity and rate of LO [66]. This relationship is shown in Figure 7.4. One can see that very dry products or intermediate moisture products are very susceptible to LO. Dry foods such as dry milk, potatoes, potato chips, and dry cereals are particularly susceptible to LO because they have substantial area for oxygen exposure, are permeable to oxygen, and may be at water activities (less than the monolayer) such that oxidation rate is high.

Lipid oxidation rate is temperature dependent. While LO may be initiated by enzymes (e.g., lipoxygenase), it most often is a pure chemical reaction responding predictably to reaction temperature. The higher the temperature, the faster the rate

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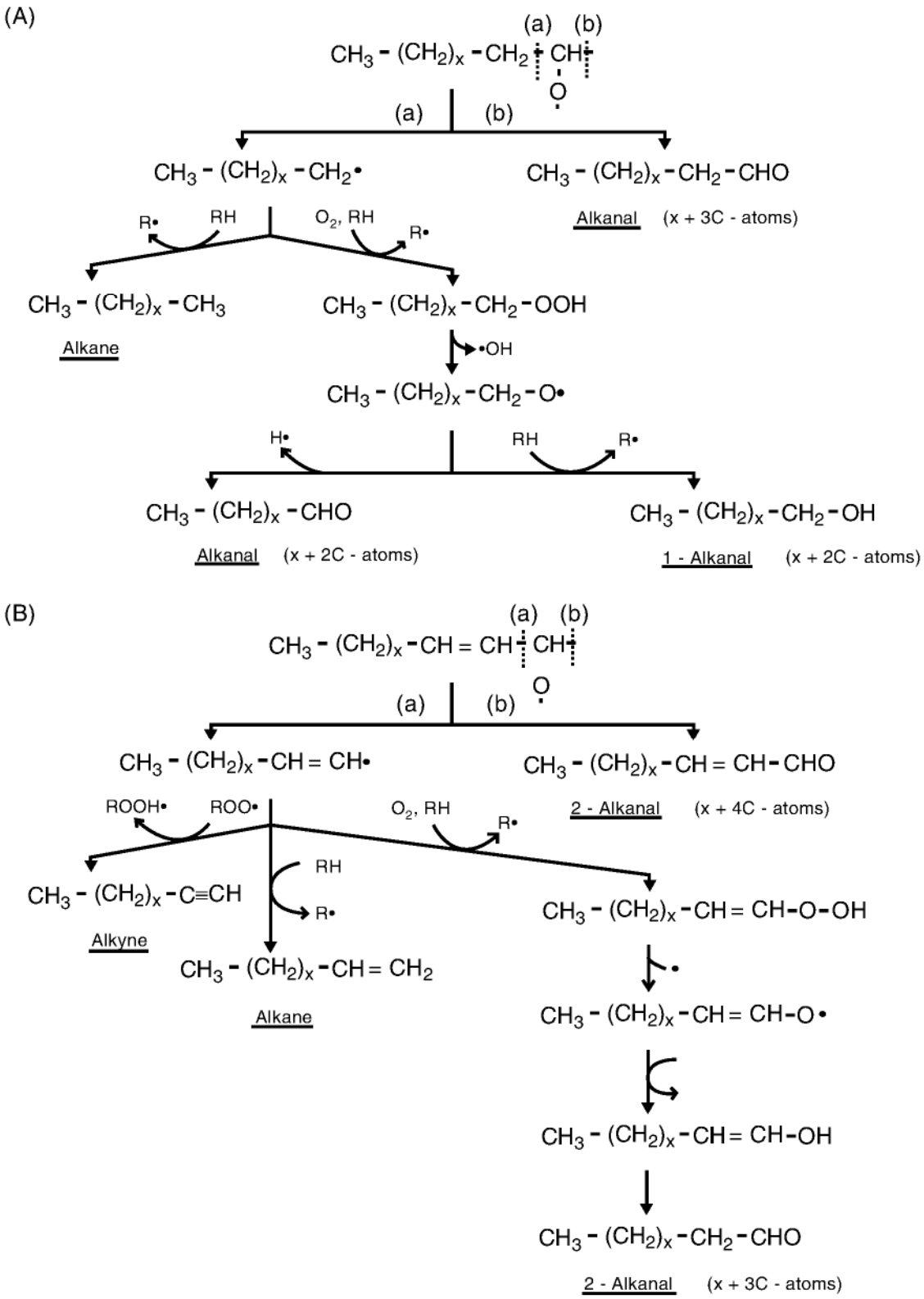

FIGURE 7.3 Mechanisms for the formation of volatile flavor compounds via lipid oxidation. (A) R' is saturated. (B) R' contains an "ene" system. (From Grosch, W., *Food Flavours*, Part A: Introduction, I.D. Morton, A.J. MacLeod, Eds., Elsevier, New York, 1982, p. 325. With permission.)

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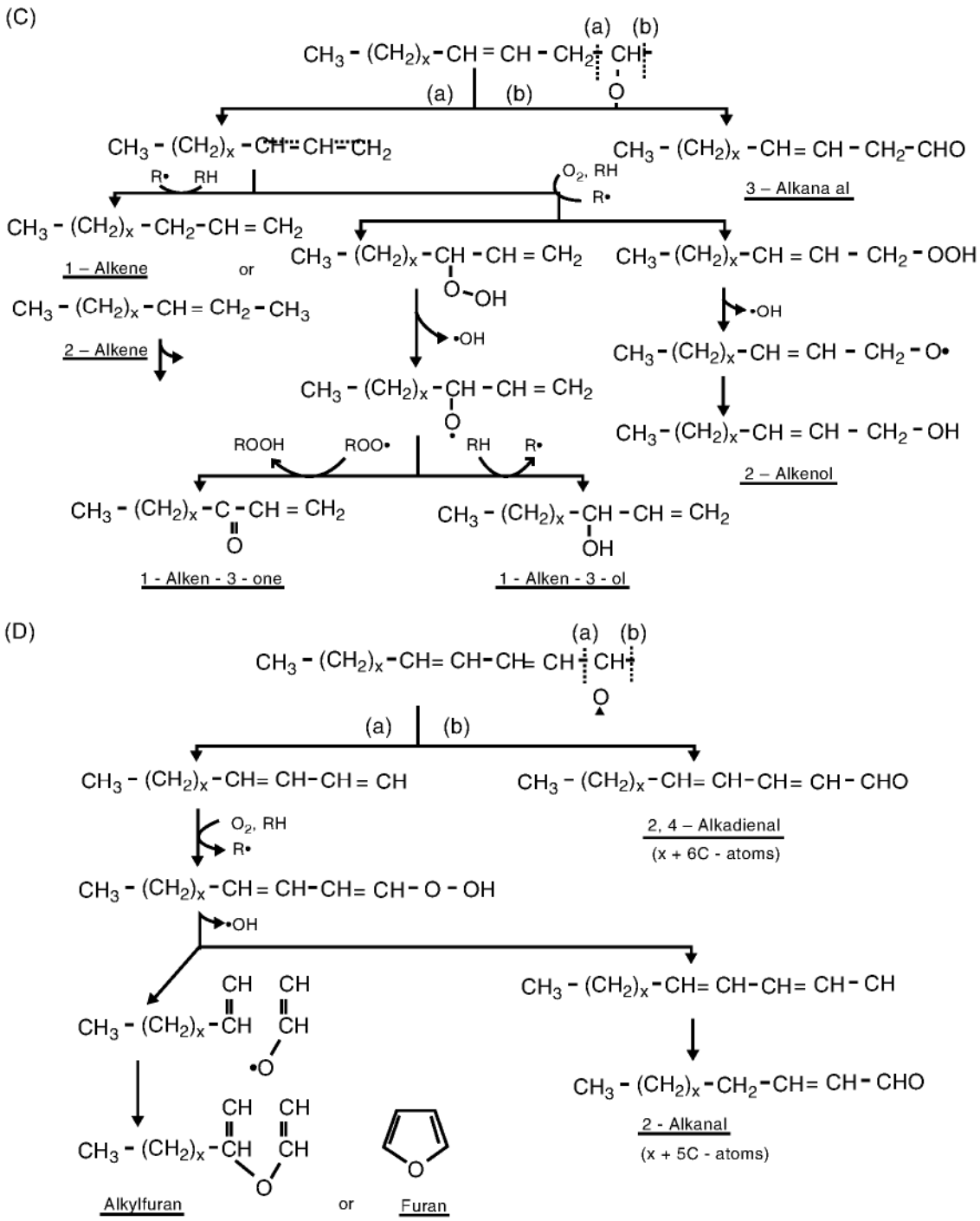

FIGURE 7.3 (Continued). (C) R' contains an "allylic" system. (D) R' contains a "diene" system. (From Grosch, W., *Food Flavours*, Part A: Introduction, I.D. Morton, A.J. MacLeod, Eds., Elsevier, New York, 1982, p. 325. With permission.)

of LO. When LO is catalyzed by enzymes, the reaction will generally have its highest rate of reaction in the 30 to 45°C range with rate decreasing when the enzyme is denatured [64]. The enzyme catalyzed reaction is pH dependent and fatty acid specific (i.e., methylene interrupted, cis, double bonds).

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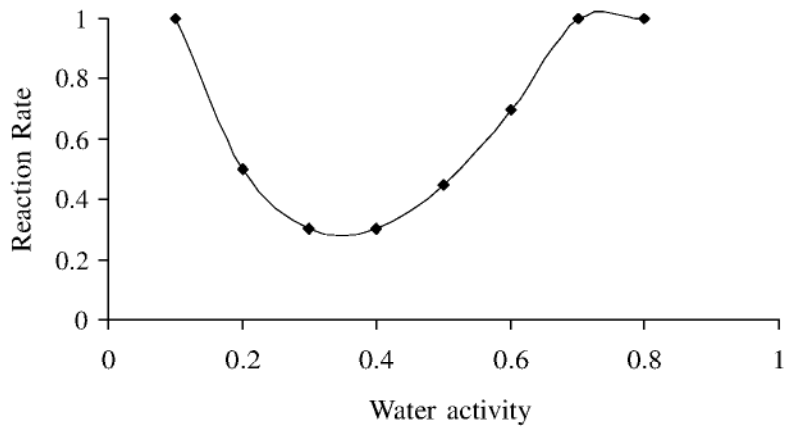

FIGURE 7.4 Relationship between rate of lipid oxidation and water activity.

As discussed later, light is an effective initiator of LO. The shorter wavelengths are higher in energy and therefore, are more effective in enhancing the rate of LO. A final factor determining rate of LO is the presence of antioxidants. Most of the vegetable oils contain some tocopherols and flavonoids that are quite effective antioxidants. The amount of natural (or added synthetic) antioxidant will strongly influence rate of LO. Kochhar [64] has provided considerable discussion of the role of antioxidants (primary and secondary) in inhibiting off-flavors (OF) in foods.

If one considers the factors that influence rate of LO, one can see that very dry products high in polyunsaturated fatty acids are most likely to develop off-flavors due to LO. However, one must also recognize that virtually all foods contain sufficient lipid to develop off-flavors via LO. It is only a question of whether some other mode of product failure occurs before LO becomes significant. It is a question of time. It was thought that low-fat (or no fat) foods would be less likely to suffer from LO. However, it has been found that these products are actually *more* susceptible to off-flavors from LO than the full fat versions of these products since fat tends to mask off-flavors. Reducing the fat content reduces any masking, making off-flavors more visible to the consumer.

While we usually think of LO in terms of dry milk, breakfast cereals, and snacks, occasionally, we find fish and meats developing off-flavors via oxidation. Fish are very susceptible due to their very highly unsaturated fatty acids. Mendenhall [74] reported significant oxidation in fresh mullet and bluefish after three and five days storage, respectively. However, most fresh fish spoil via microbiological means before oxidation becomes significant. Frozen fish have a sufficiently long shelf-life that oxidation becomes the major mode of deterioration. While other meats are not nearly as susceptible to oxidation (nor bacterial spoilage), one occasionally finds oxidized, frozen meats, particularly from foreign countries where the meat has been stored for several months during shipping, etc.

It should be mentioned that foods containing carotenoids also undergo oxidative deterioration. Carotenoids contain two end groups (often cyclic) joined by a series of conjugated trans, double bonds. The oxidation of carotenoids yields highly unsaturated compounds plus cyclic derivatives of the carotenoids. Ayers et al. [75] were the first to identify α-ionone, β-ionone, and β-ionone-5,6-epoxide as off-flavor

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TABLE 7.5
Volatile Compounds Responsible for Off-Notes Resulting from Lipid Oxidation

Flavor	Compounds
Cardboard tallowy	Octanal; alkanals (C9–C11): alk-2-enal (C8–C9); 2,4-dienals (C7–C10); nona-2-*t*-6-t-dienal
Fatty/oily	Alkanals (C5–C7): hex-2-enal; 2,4-dienals (C5–C10); 2-*t*-pentenylfuran
Painty	Alkanals (C5–C10); alk-2-enals (C5–C10); hepta-2-t,4-t-dienal; 2-heptanone; pent-2-*t*-enal; but-2-*t*-enal
Oxidized	Oct-1-ene-3-one; octanal; hept-2-enal; 2,4-heptadienal; alkanols (C2–C9)
Fishy	Alkanals (C5–C10); alk-2-enals (C5–C10); hepta-2-*t*,4-*t*-dienal; 2-alkanones (C3–C11); oct-1-en-3-one; deca-2,4-*c*-7-*t*-trienal; pent-1-en-3-one
Grassy	2-*t*-Hexenal; nona-2,6-dienal; 2-*c*-pentenylfuran; hexanal; heptanal
Mild pine-like	3-*t*-Hexenal
Rotten apple	2-*t*,4-*c*-Heptadienal
Rancid (hazelnut)	2-*t*,4-*t*-Heptadienal
Green-beany	3-c-Hexenal
Beany	Alkanals; non-2-enal
Deep-fat fried	2-t,4-*t*-Decadienal
Sweet aldehydic	2-*t*,4-*c*-Decadienal
Mushroom moldy	Oct-l-en-3-one: oct-1-en-3-o1
Cucumber-like	Nona-2-*t*,6-*c*-dienal; non-2-*t*-enal
Melon-like	Nona-3-*c*,6-*c*-dienals; non-6-*c*-enal
Potato-like	Penta-2,4-dienal
Lemon	Nonanal
Sharp	Octanal; pentanal
Brown beans	Oct-2-enal
Metallic	Pent-l-en-3-one; oct-l-en-3-one; 2-*t*-pentenylfuran; l-*c*,5-octadien-3-one
Rancid	2-*t*-Nonenal: volatile fatty acids (C4–C10)
Nutty	2-*t*,4-*t*-Octadienal; 2-*t*,2-*c*-octenal
Creamy	4-*c*-Hcptcnal
Buttery	2-*c*-Pentenylfuran; diacetyl; 2,3-pentanedione
Fruity	Alkanals (C5, C6, C8, C10); aliphatic esters; isobutyric acid
Green putty	3-*t*-Hexenal; 4-*c*-heptenal
Hardened hydrogenation	6-*t*-Nonenal
Liquorice	2-*t*,1-Pentenylfuran; 2-*c*,1-pentenylfuran; 5-pentenyl-2-furaldehyde
Soapy/fruity	Alkanals (C7–C9)
Bitter	Pentanal; hexanal; 2-*t*-heptenal

Data compiled by Kochhar [64] from Smouse and Change [80]; Badings [81]; Forss [82]; Kochhar and Meara [83]; Swoboda and Peers [84]; Chang et al. [85]; Grosch [86]; Hsieh et al. [87].

components of dehydrated carrots. The ionones have a floral character and are typically used in the formulation of raspberry flavorings. If dehydrated yellow or red vegetables (containing carotenoids) are to be used in a food product, an antioxidant must be used to provide adequate shelf life. Products such as dry carrot cake

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mixes and foods containing dehydrated tomato products commonly suffer from carotenoid oxidation. It is also interesting that frozen tomato products, e.g., pizza, will undergo substantial carotenoid deterioration during the average distribution and storage system. It is in fact carotenoid deterioration that typically limits the shelf life of frozen pizzas.

Warmed-over flavor (WOF) is an off-flavor associated with precooked meats [76]. This off-flavor is generally accepted as being due to the oxidation of intramuscular, phospholipids fatty acids. Oxidation proceeds at a very rapid rate following heating of the meat due to denaturation of heme proteins and destruction of cellular structure thereby permitting intimate mixing of cellular constituents. While the reaction is probably catalyzed by both heme and nonheme iron, the nonheme iron is considered to be of greater importance. WOF can be inhibited by the same methods that are used for lipid oxidation. This includes the use of metal chelators, primary and secondary antioxidants (including natural spice extracts), nitrite, Maillard products, vacuum packaging, etc. [77]. Grosch and coworkers [78,79] have determined that the characteristic sensory defect is due to the loss of desirable aroma components (e.g., two furanones) and the formation of oxidation products, primarily n-hexanal and trans-4,5-epoxy-(E)-2-decenal.

7.5.2 Nonenzymatic Browning

Nonenzymatic browning (ascorbic acid browning and the Maillard reaction) is a common source of off-flavor in foods. While the Maillard reaction is very important to the production of desirable flavors (Chapter 5), it is also a source of undesirable flavors in foods. (Ascorbic acid browning has no benefits — it is only negative.) Undesirable flavors may be the result of unwanted Maillard reactions during processing or in subsequent storage. An example of the formation of undesirable flavors during processing is the characteristic flavor of many canned goods. Whether one wishes to say that canned peas, for example, have an off-flavor due to the thermal processing is a matter of semantics. Canned foods do not taste like their fresh counterparts. The primary reason for a flavor change is Maillard reactions, which occur during the canning process. Virtually any food product that undergoes substantial heat treatment develops a flavor from the Maillard reaction.

It is fascinating that the Maillard reaction that occurs during storage is undesirable while that which occurs during heating (e.g., toasting of cereal) is desirable. Why is it that a loaf of bread does not develop a better flavor as it is stored? Browning is occurring in the bread during storage, and browning produces much of the characteristic desirable flavor of the bread during baking. The reason, of course, goes back to reaction kinetics. As explained in Chapter 5, the Maillard reaction is exceedingly complex with a host of possible pathways. The particular pathways active depend on many factors, including water activity, reactants present, temperature, and pH. The primary difference between developing desirable flavors via the Maillard reaction as contrasted to developing off-flavors during storage is temperature. At storage temperatures, the reactions leading to toasted, roasted, nutty, and meaty flavor compounds are not favored, and therefore, these notes ultimately disappear from the product during storage. Unfortunately the Maillard pathways

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leading to stale, sour, green, sulfury notes can and do progress at typical storage temperatures. Therefore, off-flavors develop in foods during storage due to this reaction. Like lipid oxidation, nearly all foods have sufficient reactants (reducing sugars and amino acids) for the Maillard reaction to occur. Therefore, it will ultimately result in an off-flavor unless some other mode of deterioration renders the product off-flavored first.

The off-flavors due to the Maillard reaction are most typically labeled as being stale in character. Stale is one of the more nebulous terms used in sensory evaluation. Stale is a rather nondescript term denoting a lack of fresh character. Benzothiazole and O-aminoacetophenone are two compounds believed to be formed via Maillard browning, which are responsible for this stale flavor. These compounds were found in stale, dry milk by Parks et al. [88]. O-aminoacetophenone along with some furans were also found to be partially responsible for the "gluey" flavor of old casein [89]. There is also some work in this area related to fruit juices.

Fruit juices generally develop undesirable color changes and a stale flavor during storage [90]. While off-flavors due to the Maillard reaction are quite common, few studies other than those noted have been successful in identifying the components responsible for staleness.

7.5.3 Photo-Induced Off-Flavors

Some quite unique off-flavors may be produced via photo-catalyzed reactions in foods. "Skunkiness" in beer and "burnt feathers" in dairy products are two of the more unique examples of off-flavors in foods due to light exposure.

At one time, light-induced (alternatively called light-activated or sunlight) off-flavors in milk were very common since milk was bottled in clear bottles and left on home doorsteps exposed to light. Changes in packaging (plastic-coated paper) and distribution (supermarkets) nearly eliminated this problem. However, several years ago the industry decided to package milk in clear or translucent plastic jugs and display the product in brightly illuminated diary displays. This resulted in the revival of this off-flavor. Fortunately, the industry is now using opaque plastic packaging with good light blockers, and the off-flavor is again on the decline.

Light-induced off-flavors in dairy products can be considered as two very different off-flavors. One is due to typical lipid oxidation and the other to protein (amino acid) degradation [91]. Amino acid degradation produces the light-activated off-flavor while lipid oxidation produces rather typical oxidized notes. The burnt feather or light-activated flavor initially predominates (as little as two hours light exposure); however, the milk would be judged as typical oxidized after longer light exposure.

Amino acid degradation is catalyzed by light and riboflavin as is shown:

$$\text{Amino acids} \xrightarrow[\text{Riboflavin}]{h\eta} \text{Volatile compounds} + NH_3 + CO_2$$

While all amino acids undergo this degradation, the extremely low sensory thresholds of compounds arising from the sulfur-containing amino acids (particularly methionine) are believed to be most responsible for this defect. Patton [92] and Allen and

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Parks [93] initially suggested that methional was the key contributor to this defect. However, Samuelsson [94] and Forss [152] and more recently, Jung et al. [95] have suggested that dimethyldisulfide is the key contributor with other sulfur compounds (methanethiol and dimethylsulfide) potentially contributing as well.

Off-flavors due to light exposure have also been reported in champagne [96] and beer [97]. Similar to dairy products, the photo-induced changes in champagne involve the riboflavin catalyzed degradation of methionine. Light-induced changes in beer flavor are the result of more complex reactions involving photo-oxidation of lipids (later stages of the off-flavor), photo-catalyzed decarboxylation and deamination of amino acids, and hydrolysis of the isohumulones. Beer will take on an initial off-flavor often characterized as being "skunky" or "leek-like" [98]. This off-flavor is attributed to the photo-catalyzed degradation of the isohumulones [99]. Tressl et al. [100] have provided a pathway illustrating the photo degradation of isohumulone (Figure 7.5). Note that the 4-methyl-3-pentenoyl side chain of isohumulone is cleaved to yield 2,7-dimethyl-2,6-octadiene and 4-methyl-2-pentenal as major end products. This pathway, in the presence of cysteine, has been shown to yield 3-methyl-2-butene-l-thiol (prenyl mercaptan) via free radical mechanisms. Prenyl mercaptan has a sensory threshold of 7 ng/l in beer and is considered to be the offending aroma compound [101]. Control of this off-flavor is typically accomplished by packaging the beer in light impermeable bottles or cans. The alternative is to use hops, which have had the iso-α-acids (isohumulones) reduced to their saturated counterparts. Reduced α-acids will not form the objectionable thiols. They will, however, still undergo photo-catalyzed cleavage into "green" off-notes and result in a somewhat less objectionable off-flavor [100].

A final example of a unique off-note coming from light-induced reactions involves cottonseed oil. Potato chips that are fried in cottonseed oil and then exposed to light develop a distinctive off-flavor also termed "light struck." Fan et al. [153] found this defect to be due to the formation of 1-decyne that arises from the light-induced oxidation of sterulic acid (a cyclic fatty acid) common to cottonseed oil. They found that chlorophyll was a photo sensitizer for this reaction.

Light is recognized as a catalyst for other chemical reactions, most importantly, lipid oxidation. There are numerous examples in the literature of off-flavors arising in foods due to light-induced lipid oxidation (see review by Borle et al. [154]). The sensory character of this defect varies with the food product.

7.5.4 Enzymatic Flavor Changes

One must remember that virtually all foods were once living. These living entities depended on enzymes for metabolism and subsequent growth. Therefore, foods contain a variety of enzymes. While many are inactivated by processing or may be inactive due to inadequate conditions for functioning (e.g., low-water activity, temperature, and pH), some foods do contain active enzymes, which may cause OF to develop in the food product. The three enzymes most commonly associated with OF in foods are lipoxygenase, lipase, and various proteases.

Lipoxygenase is an enzyme that will attack cis-cis double bonded, methylene interrupted fatty acids (most commonly linoleic and linolenic acids). This enzyme

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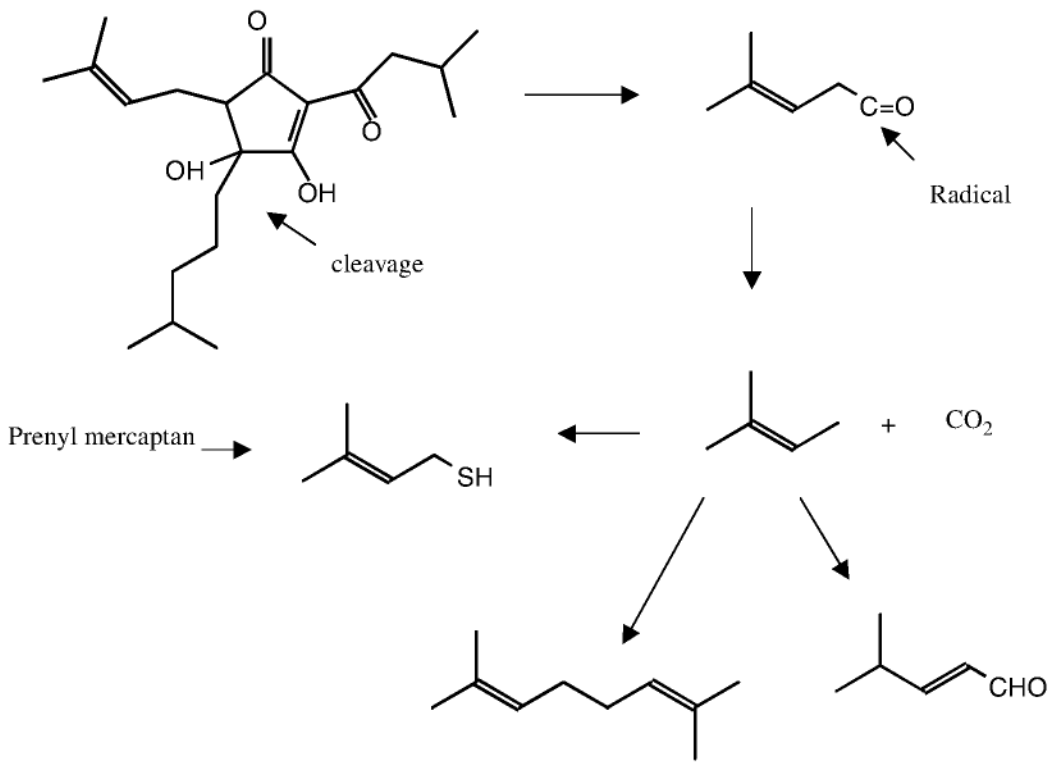

FIGURE 7.5 Photodegradation of iso-α- humulones in a cysteine model system. (From Saxby, M.J., S. Wragg, *Ind.*, p. 310, 1985. With permission.)

initiates lipid oxidation by abstracting a hydrogen radical from fatty acids. Once lipid oxidation has been initiated by this enzyme, the process of oxidation follows the typical auto-oxidative process leading to OF, which has been previously described in this chapter.

Lipoxygenase enzymes are quite common to plant tissues. The legumes (especially soybeans) contain substantial quantities of lipoxygenase enzymes. The beany flavor of soybeans is believed to be due to lipoxygenase activity in the bean once tissue damage has occurred [102–104]. The reversion flavor of soybean oil is also believed to be due to lipoxygenase activity in the oil. Hsieh et al. [155] have shown that 2-pentylfuran is the primary oxidation product responsible for this OF.

Lipoxygenase also plays a role in the deterioration of frozen vegetables. Many vegetables must be blanched prior to frozen storage in order to inactivate lipoxygenase enzymes, which otherwise result in oxidized OF [105] during storage.

Off-flavors may arise from lipase activity in foods. Lipolyzed flavor is due to the hydrolysis of fatty acids from triglycerides. Since lipase is quite widely occurring in food products, one would expect to find lipolyzed OF more commonly than encountered. The major reason lipolyzed OF is not encountered more frequently is that most lipids in foods contain only mid- to long-chain fatty acids. These fatty acids (>12 carbons) are too large to make a significant contribution to either odor or taste. Therefore, the only foods where lipolysis produces significant OF are those that contain short chain fatty acids (e.g., dairy or coconut oil products, Table 7.6).

As one would expect based on the flavor descriptors of the fatty acids (Table 7.6), the OFs arising from lipase activity in dairy products are characterized as being

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TABLE 7.6
Flavor Characteristics and Thresholds of Free Fatty Acids in Selected Media

Fatty acid	Flavor Character	Threshold (ppm) Milk	Threshold (ppm) Oil
Butyric acid	Butyric	25	0.6
Caproic acid	Cowy (Goaty)	14	2.5
Caprylic acid	Cowy (Goaty)	—	350
Caprylic acid	Rancid unclean bitter soapy	7	200
Lauric acid	Rancid unclean bitter soapy	8	700

"butyric," "goaty," and/or "bitter" in nature [7,106]. There appear to be two lipases in dairy products: one associated with the fat globule membrane and the other with the proteins. While the point is controversial, it seems that these two lipases are identical but located in different parts of the milk. Milk lipases are highly specific for the primary positions of the triglyceride. Unfortunately, nature has also shown a preference to put the most sensorially significant fatty acids (i.e., butyric and caproic acids) in the primary positions.

The lipases of milk are rather unique in that they are inactive until activated by some treatment (the exception being rare that a cow will produce spontaneously rancid milk). Activation of the lipase enzyme system may be accomplished in several different ways. Occasionally, fluctuations in temperature such as cooling, warming and then cooling, will cause the lipase to become active. Mechanical agitation or foaming can also activate the lipase systems. This was a particular problem when pipeline milking systems were initially introduced. Homogenization of raw milk is especially effective in producing lipolyzed flavor. In a matter of seconds, the homogenization of raw milk results in so many free acids that it will nearly burn the mouth. The milk fat globule membrane is inadequate to cover the new surface generated upon homogenization of the milk. This unprotected fat is particularly susceptible to lipase attack.

It is of interest that lipase activity can be influenced by the presence of certain chemicals [107]. Aureomycin, penicillin, streptomycin, and terramycin were shown to provide 7.6 to 49.8% inhibition of lipase.

While dairy products may become lipolyzed due to indigenous enzymes, the lipases may also be from outside sources. Bacterial activity, spices, and some fruits contain lipases that may attack the triglycerides of milk. It is relevant that different lipases (different sources) will have a different preference for lipolysis of fatty acids. Milk lipase and other ruminant sources of lipases tend to preferentially lyse butyric acid from triglycerides. Lipases from *Pseudomonas* fluorescens and all porcine lipases have little preference for short chain fatty acids. Lipases from molds and *Chromobacterium* viscosum produce less butyric acid but more capric acid [108]. Lipases that preferentially lyse the short chain fatty acids have a greater impact in the production of off-flavors than do those lipases that lyse the longer chain fatty.

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This suggests that an analytical method that measures total free fatty acids (e.g., acid degree value) may be a poor measure of the off-flavor potential of these fatty acids.

The other food products (ingredients), which are subject to lipolyzed OF are the tropical oils, most notably coconut oil. Coconut oil contains lauric acid (C12) as a major fatty acid. Lauric acid tastes soapy when hydrolyzed from the triglyceride. While coconut does not contain a significant amount of lipases, coconut is often used in foods with other ingredients that do contribute lipases. It should be mentioned that lipases are reasonably heat stable enzymes. They will often survive a thermal treatment that one would expect to be adequate to denature the lipase.

A beverage where "soapy" flavors are a problem is the pina colada. Pina colada is an alcoholic drink consisting primarily of coconut and pineapple juice. The pineapple is an excellent source of quite heat stable lipases. If one encounters a soapy OF in a food product, the first step is to search the ingredients for coconut oil and then a source of enzyme.

Bitterness in foods is an additional OF that may occur due to enzyme activity. In this case, we are considering bitterness arising from proteolytic activity on food proteins. The organisms that produced the enzymes responsible for bitterness may have been killed by a heat treatment but some proteolytic enzymes may survive rather severe heat treatments to remain active in the food thereby causing off-notes. Murry and Baker [109] are often credited with being the first to determine that this bitterness was due to short chain peptides.

Bitter peptides have been found to produce OF in soy products [110–112], zein [114], casein [115–117], and Cheddar cheese [119]. Arai [113] pointed out that most of these peptides have leucine at the C-terminal end. Otherwise there appears to be little similarity between peptide structures (Table 7.7).

Probably the largest amount of research on bitterness due to peptides has focused on aged cheeses [120–124]. Proteolytic activity in the cheese comes from the rennin, starter organisms, and/or contaminating microorganisms. Rennin initially breaks a

TABLE 7.7
Bitter Peptides Identified in Protein Hydrolysates and Cheese Products

Soy globulin hydrolyzed with pepsin
Arg-Leu, Gly-Leu, Leu-Lys, Phe-Leu, Gln-Tyr-Phe-Leu, Ser-Lys-Gly-Leu
Zein hydrolyzed with pepsin
Ala-Ile-Ala, Gly-Ala-Leu, Leu-Val-Leu, Leu-Pro-Phe-Ser-Gln-Leu
Casein hydrolyzed with trypsin
Gly-Pro-Phe-Pro-Ile-Ile-Val, Phe-Ala-Leu-Pro-Gln-Tyr-Leu-Lys
Cheddar cheese
Pro-Phe-Pro-Gly-Ile-Pro
Pro-Phe-Pro-Gly-Pro-Ile-Pro-Asn-Ser
Leu-Val-Tyr-Pro-Phe-Pro-Gly-Pro-Ile-Pro

Source: From Arai, S., *The Analysis and Control of Less Desirable Flavors in Foods and Beverages*, G. Charalambous, Ed., Acad. Press, New York. p. 1, 1980. With permission.

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portion of the protein down into large peptides. These peptides then are further degraded into smaller peptides that may cause bitterness unless they are further metabolized by the starter organisms [125]. Therefore, starter failure, retardation via chemicals or contaminating organisms may end up causing bitterness in a cheese. The proper starter organisms and their growth is essential to minimize the possibility of bitterness during aging of cheese [126].

7.6 MICROBIAL OFF-FLAVORS

Microorganisms (MO) are a common source of OF in foods [127]. If the product is not sterile (via heating or irradiation), or preserved via antimicrobial agents (e.g., humectants or chemical preservatives), microorganisms may grow in the product and produce OF.

Off-flavors may arise in food products via microbial sources in many different ways. The OF may be the result of a planned fermentation going wrong. The fermented product may then develop an OF. An example of this would be a starter culture failure in a cheese vat. Failure of the starter culture would result in poor acid production and the opportunity for the growth of other organisms. These contaminating organisms often result in bitterness and unclean OF. This type of OF development is possible in virtually all types of fermented foods. Only through very careful handling of the starter cultures and manufacture of the fermented products can this problem be minimized.

A second way in which microorganisms may produce OF in foods is somewhat indirect. Enzymes from lysed MO can catalyze reactions in foods leading to OF. Microbial enzymes are frequently quite stable to heat denaturation surviving pasteurization and sterilization processes [7,128]. Therefore, a thermal process may kill the MO but leave active enzymes (e.g., lipases or proteases), which can produce OF on storage. One can appreciate that a food may develop an OF in this manner but yet show very low *viable* cell counts. The low viable cell count could mislead the investigator into believing that the OF was not due to MO.

The final means by which MO can produce OF in foods is the most common. That is from growth in the food and contamination with metabolic products. Fresh fish [17] and dairy products [129] are two foods that are very susceptible to OF from microbial sources. Fresh fish is generally considered to have very little odor. However, if fish is stored above freezing temperatures, the fish develops a "fishy" OF initially that later becomes putrid and foul in character. Fish is particularly susceptible to microbial growth since the fish muscle has substantial soluble material and cells tend to lyse once the fish has been killed. The fish muscle provides a rather ideal growth medium for MO.

The fishy OF is primarily due to the generation of trimethylarnine via bacterial action [130,131]. Trimethylamine is formed from trimethylamine oxide, which is a natural constituent of fish muscle. This reduction is accomplished through bacterial enzymes and involves a coupled oxidation of lactic acid to acetic acid and CO_2 [132]. The latter stages of fish spoilage involve the production of various nitrogen- and sulfur-containing compounds. These compounds produce putrid, sulfury notes in the fish.

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It should be noted that different MO vary in their ability to produce OF in foods. The ability to produce an OF is dependent upon the metabolic compounds which are produced and excreted into the growth medium (food) by the MO. Work on OF in fish demonstrated this very nicely. Herbert and Shewan [133] have shown that Gram-negative bacteria of the genera Pseudomonas, Achromobacter, and Vibrio are most often responsible for OF in fresh fish. Within these *genera*, only a few species can cause spoilage odors. Adams et al. [134] found that typically less than 10% of the bacterial population present on fish can cause OF. This should make one cautious in concluding that high plate counts (or the converse) necessarily mean an OF came from microbial sources. It is the specific organisms and populations that are significant in determining the contribution to OF.

Shipe [129] and more recently Joen [7] have reviewed some of the OFs that may occur in milk due to MO. Prior to wide usage of pasteurization and refrigeration, milk would most commonly spoil due to *Streptococcus* lactis growth. This would result in an acid, sour OF. Pasteurization quite effectively kills *S.* Lactis, and its growth is not favored by storage at refrigeration temperatures, so this organism (and OF character) is no longer a common means of spoilage.

Today the psychrotrophic bacteria are typically responsible for OF in milk. These MOs can multiply at or below 7°C so they will grow at typical refrigeration temperatures. Even though pasteurization destroys these MOs, they produce substantial quantities of heat stable lipases and proteases that may produce OF later during storage. They also are common post pasteurization contaminants that will grow in the milk and produce OF due to metabolic activity. The OFs arising from psychrotrophic bacteria are often characterized as being unclean, foreign, fermented, or bitter.

Two quite characteristic OFs found occasionally in milk are fruity and malty OFs. Fruity defects have been often traced to the presence of *Pseudomonas* fragi. *Pseudomonas* fragi produces a lipase, which initially hydrolyzes short chain fatty acids from milk lipids and then esterifies these acids to form their corresponding ethyl esters. Ethyl butyrate and ethyl caproate have been found to be the OF compounds responsible for this defect [91].

Malty OF occurs occasionally in milk, which has had some temperature abuse (storage at 10°C or above). The malty OF is caused by *Streptococcus* lactic subsp. Maltigenes [135] or *Lactobacillus* maltaromicus [136]. The malty character itself is due primarily to 3-methyl butanal production from leucine.

While geosmin has been mentioned earlier as the compound responsible for earthy taints in fish, it has also been linked to taints in bread [137], wheat flour 138], navy beans [139], and clams [140]. Molds and Actinomycetes growing in these products were found to produce this taint. These are similar organisms to those that produce earthy taints in soils that often end up in root crops such as beets or sugar beets (and sugar made there from).

It is somewhat ironic that molds have been found to grow on foods that have been treated with sorbic acid (a mold inhibitor), degrading the mold inhibitor to 1,3-pentadiene, which has a petroleum-like odor [141]. While sorbic acid is effective in inhibiting molds, if a mold can grow on parts of the food where the inhibitor may

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$$CH_2CH{=}CHCH{=}CHCOOH \rightarrow CH_3CH{=}CHCH{=}CH_2 \text{ (kerosene taint)}$$

FIGURE 7.6 Degradation of sorbic acid to 1,3-pentadiene.

be too low to be effective, the mold will potentially produce this taint (Figure 7.6). While the majority of such taints have been in cheese (Feta — a plastic, paint, or kerosene defect [142]; cheese spread — kerosene defect [143]), other foods that are treated with sorbic acid and are high in moisture have also been found to have "kerosene" taints due to this compound [2].

Sorbate has also been implicated as the precursor of a flavor defect in wine that has a geranium character. Sorbate may be added in the winemaking process as a secondary yeast inhibitor to reduce the amount of sulfur dioxide used. While sorbate is a good yeast inhibitor, it does not inhibit the growth of bacteria. Crowell and Guymon [144] have found that certain bacteria in wines can convert sorbate to 2-ethoxyhexa-3,5-diene, which has a geranium character.

It is interesting that styrene in food products is generally assumed to arise from packaging contamination (polystyrene polymers), but it has been found to also arise from microbial action. Saxby [145] described an off-flavor problem where a yeast, *Hypopichia burtonii,* growing in the presence of a cinnamon flavoring produced styrene. Cinnamon flavor is based on cinnamic aldehyde: decarboxylation of cinnamic aldehyde yields styrene. This defect was found in a spiced, baked good.

A microbial conversion of sensory significance already discussed to some extent is the conversion of phenols to chloroanisoles (airborne taints). A defect in wine recognized as cork taint is considered to be primarily due to this conversion. It was estimated that cork taint results in the rejection of 2 to 6% of commercial wines [146]. While this reference is somewhat dated, it is of relevance that this is the second most researched (based on number of publications) sensory defect in foods (after boar taint), which suggests that the defect is still of importance to the industry. Chloroanisoles can be formed from the microbial conversion of pentachlorophenol (a component of some insecticides and wood preservatives) in the cork or from hypochlorite treated corks (forms chlorophenols) with subsequent microbial methylation to the anisoles. In either event, the chloroanisoles (primarily 2,4,6-trichloroanisole) are formed on/in the cork and leach into the wine [127]. This has lead to a movement within the wine industry to use synthetic bottle stoppers in place of cork.

One must recognize that microorganisms can accomplish a wide range of chemical transformations thereby potentially producing a host of different off-flavors. Earlier in this section, several off-notes have been described as being the result of microbes. One can add other sensory notes such as: faceal (indole and skatole in rotting potatoes [147]), musty potato (2-methoxy-3-isopropylpyrazine [148]), mousy (wine, 2-acetyl-1,4,5,6-tetrahydropyridine [149]), and musty, "dirty dishcloth" (machine oils 2,6-dimethyl-3-methoxypyrazine [150]).

Additional examples of OF produced in foods from MO are provided by Goldenberg and Matheson [4]. A summary of OF they have encountered at Marks and Spencer is presented in Table 7.8. It is quite obvious from their tabulation that OF due to MO can occur in a wide variety of products.

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TABLE 7.8
Off-Flavors Due to Microbial Spoilage

Food	Type of Spoilage	Comments
Cakes	Bacteria	Off-flavor due to bad egg; either use of poor quality egg or careless handling during or after thawing of frozen egg
Cakes; angel cakes; apple tarts; marzipan	Mold	Mold due to contamination by flour dust in bakery
Cakes	Lipolytic bacteria	Soapy off-taste in coconut
Apricot jam	Bacteria-producing butyric acid from sugar	Inadequate processing or inadequate cleaning or washing of equipment
Fresh cream	Bacteria	Contamination by yogurt
Cream	Lipolytic bacteria	Cheesy off-flavor due to growth of lipolytic bacteria either in milk or during processing before pasteurization
Cream	Bacillus *cereus*	Causes bitter flavors
Chocolate tea cakes	Osmophilic yeasts	Caused fermentation and beery off-flavor
Biscuits	Molds	Musty flour used and not examined before used
Chicken	Molds	Wood shavings used as litter in chicken broiler-house made from wood preserved with chlorophenols. This metabolized to chloroanisoles and absorbed by chicken flesh
Canned ham	Fecal streptococci	Cheesy off-flavors caused by fecal streptococci that survive heat process, especially in large 15-lb packs
Wilshire bacon	Bacteria	Development of sulfide off-flavor caused by contamination of bacon brines by foreign bacteria
Minced beef pies	Lactic acid bacteria	Contamination of cooked beef mix by lactic acid bacteria
Sausages	Lactic acid bacteria	Souring of sausages caused by selective growth of lactic acid bacteria in presence of sulfite
Ice Cream	Bacteria	Smoky defect noted in rework. Conversion of vanillin to guaiacol and 2-ethoxyphenol

Source: From Saxby, M.J., *Food Taints and Off-Flavors*, 2nd ed., Blackie Acad. Prof., London, 1996, p. 326, and Goldenberg, N., H.R. Matheson, *Chem. Ind.* (London), (13), p. 551, 1975. With permission.

7.7 SUMMARY

This chapter briefly discusses some of the OF that have been found to occur in foods. Unfortunately, the occurrence of OF is quite common and the causes are very diverse. It is impossible to present a detailed discussion of all of the OF reported in the literature. In some cases, the source of the OF has never been found. Despite the sophisticated instrumentation available to the flavor chemist, identification of OF in foods is literally like looking for a needle (one or two OF compounds) in a

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haystack (the normal flavor profile of the food). When one appreciates the very low sensory thresholds one may encounter in OF flavor compounds (e.g., bis-(2-methyl-3-furyl)-disulfide, 2 parts in 10^{12} parts water [151]), the task of finding and then identifying the OF becomes very formidable. Hopefully, this chapter provides a better appreciation for the diversity and the host of sources of OF in foods.

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