Chapter 1	The Role of DNA Analysis in the Determination of Food Authenticity	1
	S. B. Primrose	-
	1.1 A Brief History of Food Adulteration	1
	1.2 Food Fraud in the 21st Century	2
	1.3 Challenges in Detecting Food Misdescription	
	and Fraud	4
	1.4 The Role of DNA in Food Authenticity Determination1.5 Application of DNA-based Analytical Methods to	5
	Different Commodities and Food Authenticity	
	Problems	6
	1.6 Pushing the Boundaries: Specialist Techniques for Breed or Variety Identification and Determination	
	of Geographical Origin	8
	1.7 Fitness for Purpose and a solution of the	9
	1.8 Final Comments	9
	References	9
	3.5.5 Evaluation of a DNA Sample's Integrity	
Chapter 2	Forensic DNA - Criminal and Paternity Methods and	
	Applications – How Can This Help in Verifying Food	
	Authenticity?	12
	Victoria Moore	
	References References	
	2.1 Some History	12
	2.2 Applying the Forensic Code of Conduct2.3 Key Issues - Complexities Within Forensic	14
	Regulatory Bodies and Associations	16

Food Chemistry, Function and Analysis No.16 DNA Techniques to Verify Food Authenticity: Applications in Food Fraud Edited by Malcolm Burns, Lucy Foster and Michael Walker © The Royal Society of Chemistry 2020 Published by the Royal Society of Chemistry, www.rsc.org

XX		Contents
	2.4 Key Issues – Databases and Communication	18
	2.5 Document Recording	21
	2.5.1 Receipt of Materials	21
	2.5.2 Examination, Standard Operating	
	Procedures and Quality Control	22
	2.6 Conclusion	24
	References	25
Chapter 3	DNA Extraction from Food Matrices	29
•	Timothy Wilkes	
	3.1 Introduction	29
	3.2 Factors Influencing the Choice of Extraction	
	Methodology	30
	3.2.1 Sample Source and Processing	30
	3.2.2 Sample Collection and Storage	30
	3.2.3 Homogenisation	30
	3.2.4 Presence of Contaminants	31
	3.3 General DNA Extraction Methods	33
	3.3.1 Phenol-Chloroform Extractions	34
	3.3.2 Detergent and Protease-based Extractions	34
	3.3.3 Solid-phase Extraction Methods	35
	3.3.4 Concentrating DNA Post-extraction	36
	3.4 DNA Extraction Methods Frequently Employed	
	with Food and Feed Samples	37
	3.5 Determining DNA Quantity and Purity	39
	3.5.1 Quantification of DNA	39
	3.5.2 Evaluation of DNA Purity	42
	3.5.3 RNA Contamination	44
	3.5.4 Non-nucleic-acid Contamination	44
	3.5.5 Evaluation of a DNA Sample's Integrity	44
	3.5.6 Determining the Presence of Inhibitory	
	Compounds	45
	3.6 Concluding Remarks	46
	Acknowledgements	47
	References	47
	Valisities valisities	
Chapter 4	"DNA Techniques" Case Study: Isothermal Approaches	5 50
onapter i	G. Nixon	
	Regulatory Bodies and Associations	
	4.1 Introduction	50
	4.2 Main Isothermal NAA Technologies	51
	4.2.1 Cross Priming Amplification (CPA)	51
	4.2.2 Helicase-dependent Amplification (HDA)	51
	4.2.3 Loop-mediated Isothermal Amplification	51
	(LAMP)	52
		54

ontents		xxi
	4.2.4 Recombinase Polymerase Amplification	on. [1]
	(RPA)	54
	4.3 PCR vs. Isothermal Technologies	55
	4.4 Food Applications	55
	4.4.1 Meat Speciation	55
	4.4.2 GMOs	57
	C	57
	4.5 Future View	57
	4.6 Conclusion	58
	Acknowledgements	58
	References	59
Chapter 5	Digital Polymerase Chain Reaction (dPCR) –	
1	General Aspects and Applications	63
	S. Pecoraro	
	5.1 Introduction	63
	5.2 General Properties of dPCR	64
	5.3 dPCR Platforms	65
	5.4 Assumptions for Absolute DNA Quantification	
	with dPCR	66
	5.5 Applications of dPCR in Food Analysis	67
	5.6 Future Aspects	67
	References	68
Chapter 6	UK Food Authenticity Programme – The Analytical	
	Tool Box	70
	L. H. Foster and S. B. Primrose	
	The construction of the policy of the policy of the second s	
	6.1 Introduction: Drivers and Rationale for the	
	Programme	70
	6.2 Policy Context: Industry, Enforcement and	
	Consumer Trust	71
	6.3 Technical Challenges Affecting Policy	
	Development and Food Law Enforcement	74
	6.4 The Analytical Toolbox	74
	6.4.1 Chemical Methods	75
	6.4.2 Physical Methods	76
	6.4.3 Proteomic-based Methods	76
	6.4.4 DNA-based Methods	77
	6.5 Upskilling: Knowledge Transfer of Government-	
	funded DNA Techniques to Support Food Law	
	Enforcement	79
	6.6 Forward Look and Future Challenges	79
	6.6.1 Collaboration – a Global Response to	
	Food Fraud	80

111

	6.6.2 Future Tools to Tackle Food Fraud	80
	6.6.3 Food Labelling and Informed Choice	81
	6.7 Conclusion	81
	Acknowledgement	82
	References	82
Chapter 7	Fitness for Purpose of DNA-based Analytical Methods S. B. Primrose and L. H. Foster	86
	7.1 Introduction	86
	7.2 Fitness for Purpose	87
	7.2.1 What is 'Fitness for Purpose?'	87
	7.3 Challenges for Fitness for Purpose for	
	Authenticity	88
	7.4 Sampling and Extraction	88
	7.5 Qualitative Versus Quantitative Analysis	89
	7.6 Reference Materials	91
	7.7 Validation	91
	7.8 Measurement Uncertainty	91
	7.9 How is Fitness for Purpose Achieved?	92
	7.10 Full Validation	94
	7.11 Conclusions	94
	References	94
Chapter 8	GMO Detection and Identification Using Next-generation	
	Sequencing	96
	Marie-Alice Fraiture, Nina Papazova, Kevin Vanneste,	
	Sigrid C. J. de Keersmaecker and Nancy H. Roosens	
	8.1 The Current Landscape of GMO Detection	96
	8.2 Applying NGS to GMO Detection: Current	~-
	Approaches 8.3 Challenges for the Detection and Characterization	97
	of GMOs Using NGS Related to the Host	102
	8.4 Conclusion and Perspectives	103
	References	103
Chapter 9	A Perspective on Quantitative DNA Approaches	107
7	Malcolm Burns	
	9.1 Introduction: The Requirement for Accurate	
	Quantitation of Food Samples	107
	9.2 Recommendations for Methods for Accurate	107
	Quantitation of Food Samples	109

9.3 Approaches for Quantitative Estimation of Food Samples Using DNA

	9.3.1 Choice of Target for Quantitative Estimation 9.3.2 Choice of Method for Quantitative	111
	Estimation	111
	9.3.3 Expression Units and Measurement Scales	113
	9.4 Ensuring Fitness for Purpose for Accurate	
	Quantitation of Food Samples	115
	9.5 Summary and Future Perspectives on	
	Quantitative DNA Approaches	116
	Acknowledgements	117
	References	117
Chapter 10	DNA in Food and Feed Law Nigel Payne	120
	Nigel Payne	
	10.1 Introduction	120
	10.2 Food	121
	10.2.1 Framework Law	121
	10.2.2 Food – Subordinate Law	124
	10.2.3 Protected Designations	124
	10.3 Other Authenticity Issues	125
	10.3.1 Honey	125
	10.3.2 Herbs and Spices	125
	10.4 Allergens in Food	126
	10.5 Microbiological Safety of Food	127
	10.6 Genetically Modified Organisms – GMOs	128
	10.6.1 GM Rice and Rice Products	129
	10.7 Organic Food and Feed	130
	10.8 Protection of Vegan, Halal and Kosher Food	130
	10.9 Investigation of Food "Complaints"	130
	10.10 Animal Feeding Stuffs	131
	10.11 Legal Provisions for Official Controls of	
	Food and Feed	131
	10.12 Taking of Samples	132
	10.13 Interpreting Results in a Legal Context	133
	10.14 EU Exit	134
	References	134
Case Studie	References , notatorquanti A.2.71	139
Chapter 11	Harmonising DNA Methods – The GMO Story	141
	Hendrik Emons	
	11.1 Introduction	141
	11.2 The Harmonisation Challenge	142
	11.3 The Network Approach	143
	11.4 Achievements and Current Status	144
	Acknowledgements	145
	References	145

xxiii

xxiv		Contents
Chapter 12	Metrology of DNA Approaches	147
	Mojca Milavec, David Dobnik, Alexandra Bogožalec	
	Košir and Jana Žel	
	9.3.3 Expression Units and Mehabimman S	
	12.1 Introduction	147
	12.2 Key Comparison Studies	148
	12.3 Partition Volume Variability – a Critical Factor	
	Influencing the Accuracy of Absolute	4.50
	Quantification by Digital PCR	150
	12.4 Conclusions References	151 152
	References	152
Chapter 13	The Almond and Mahaleb Allergen Story -	
	PCR Resolution of Live Incident Investigations	154
	Michael Walker and Malcolm Burns	
	10.2 Food	
	13.1 Introduction	154
	13.2 Almond and the Prunus Family	155
	13.3 Sample Authentication	155
	13.3.1 Real Time PCR Assay Development	156
	13.4 Results	157
	13.4.1 Referred Cumin Sample	157
	13.4.2 Referred Paprika Sample	157
	13.5 Conclusions	158
	Acknowledgements	159
	References	159
Chapter 14	Food Fraud Prevention - Selecting the Right Test,	
	Method, and Sampling Plan	162
	John Spink	
	R. to Alentho Districtions for Contracts (Contracts of R	
	14.1 Introduction	162
	14.2 Food Fraud Overview	163
	14.3 Criminology and Food Fraud Prevention	163
	14.4 Case Study: Horsemeat Incident	164
	14.5 Conclusion	166
	References	166
Chapter 15	Meat Speciation	169
omptor 10	Maria Karczmarczyk	ndeur
		100
	15.1 Comparison Between PCR and ELISA	169
	15.2 Qualitative PCR Testing	170
	15.3 Quantitative PCR Testing	170
	15.3.1 Low DNA Content Samples – Dairy, Gelatin	e 171 171
	15.3.2 Effects of Food Processing 15.3.3 Interference – Inhibition	171
	15.5.5 Interference – Infilbition	1/1

Contents		XXV
	15.3.4 Limit of Detection (LOD)	172
	15.4 Proprietary Methods - Considerations and	
	Validation Approaches	173
	15.5 Reporting	174
	15.6 New Developments	174
	References	175
Chapter 16	The Horse Meat Scandal – The European Analytical	
NOS	Response	177
	Marien Aline, Fumière Olivier, Debode Frédéric,	
	Hulin Julie and Berben Gilbert	
	16.1 Introduction	177
	16.2 Material and Methods	178
	16.2.1 Real-time PCR Analysis	178
	16.2.2 Specificity of the Horse PCR Method	179
	16.2.3 Sensitivity of the Horse PCR Method	180
	16.2.4 Robustness of the Horse PCR Method	180
	16.2.5 Development of a Semi-quantitative	
	Method for the Detection of Horse Meat	181
	16.3 Results	181
	16.3.1 Evaluation of the Performance of the	
	Horse PCR Method	181
	16.3.2 Development of a Semi-quantitative	
	Method for the Detection of Horse Meat	182
	Acknowledgements	185
	References	185
Chapter 17	Horse Meat: Technical Appeals and Court Action	189
	Michael Walker	
	17.1 Introduction	
		189
	17.2 Technical Appeals 17.2.1 Technical Approach	190
	17.2.2 DNA	190
	17.2.2 DNA 17.2.3 ELISA	191
	17.2.4 Interpretation	192
	17.3 Prosecutions	
	17.3.1 Bulgarian Sausage	194
	17.3.2 Abattoir with no Horse Traceability	194
	17.3.3 Frozen Blocks of Meat and 'Bargain'	247
	Beef Burgers with Horse	195
	17.3.4 Heart in Burgers	196
	17.3.5 The Netherlands	196
	17.3.6 France	196
	17.4 Conclusions	197
	References	197

xxvi		Contents
Chapter 18	Durum Wheat and Pasta Authenticity	200
	Gordon Wiseman	
	Validation Approaches Ind another who	
	18.1 Introduction and Regulations	200
	18.2 Case Study	201
178	18.2.1 Is <i>T. aestivum</i> Present in the Pasta?	201
	18.2.2 qPCR Confirmation and Quantification of the <i>T. aestivum</i>	202
	18.2.3 Analytical Uncertainty	202
	18.2.5 Analytical Oricertainty 18.3 Conclusions	204
	Acknowledgement	205
	References	206
		200
Chapter 19	The Authenticity of Basmati Rice – A Case Study	207
178	Mark Woolfe and Katherine Steele	
	16.2.2 Specificity of the Horse PCR Method	
	19.1 Introduction	207
	19.2 Methodology Development to Authenticate	
	Basmati Rice	208
	19.2.1 Basmati Rice Variety Authentication and	
	Quantification	208
	19.3 Food Standards Agency's Basmati Rice	200
	Survey	209
	19.3.1 Basmati Rice Variety Identification	209
	19.3.2 Quantitative Measurement of	200
	Non-basmati Varieties	209 211
	19.3.3 Follow-up Action of the Survey	211 211
	19.4 Further Development of the Methodology 19.4.1 Basmati Variety Identification	211 211
	19.4.2 Quantitative Determination of	211
	Non-basmati Varieties in Basmati Rice	213
	19.4.3 Lab-on-a-chip Approach to Basmati Rice	210
	Authenticity	214
	19.5 Postscript to Basmati Rice Methodology	216
	Acknowledgements	216
	References	217
	. 17.2.4 Interpretation	
Chapter 20	Horse Meat: The International Collaborative Trial	
Watpres 15	of the Real-time PCR Method for the Quantitation	
	of Horse DNA	219
	Malcolm Burns and Lucy Foster	
	13.1 Comparison Balance States 154	
	20.1 Food Fraud	219
	20.2 The 2013 Horse Meat Issue	220
	20.3 Challenges in Meat Quantitation	220

Contents		xxvii
	20.4 Development of a Real-time PCR Method for the	
	Quantitation of Horse DNA	221
	20.5 International Collaborative Trial of the Real-time	
	PCR Method for the Quantitation of Horse DNA	222
	20.6 Summary	224
	Acknowledgements	224
	References	225
Chapter 21	Standardization of DNA-based Methods for Food	
	Authenticity Testing	227
	L. Grohmann and C. Seiler	
	23.4 Discussion sampor bus anoiseursid 3.65	300
	21.1 Introduction	227
	21.2 CEN and ISO – Activities and Projects	228
	21.2.1 CEN WS/86 – Authenticity in the Feed and	
	Food Chain – General Principles and Basic	10
	Requirements	228
	21.2.2 CEN Food Authenticity Coordination Group	
	(FACG) and a New Technical Committee	302
	on Food Authenticity	229
	21.2.3 CEN/TC 275/WG 11 'Genetically Modified	
	Organisms and Species Analysis'	229
	21.2.4 ISO/TC 34/SC 16 'Horizontal Biomarker	
	Analysis'	232
	21.3 Discussion and Outlook	233
	21.4 Conclusion	233
	Acknowledgement	234
	References	234
Chapter 22	Authentication of Chinese Traditional Medicine by DNA	
chapter ==	Analysis	235
	Foo Wing Lee, Olive Tin Wai Li and Winnie Wing Yan Chum	
	and the state way was been a second state and the	
	22.1 Introduction	235
	22.2 Reliability of DNA Methods for Species	
	Identification	236
	22.3 Selection of DNA Markers	237
	22.4 Bioinformatics Analysis	238
	22.5 Demo Analysis	243
	22.6 Example Cases	245
	22.6.1 Authentication of Cordyceps	247
	22.6.2 Authentication of Oviductus Ranae	250
	22.7 Conclusion	251
	Acknowledgements	251
	References OSOS nosinolis 8.8.82	251

xxviii		Contents
Chapter 23	DNA Point of Use Applications	255
221	Michael Walker, Malcolm Burns and Lucy Foster	
	20.5 International Collaborative Trial of the Real-th	
	23.1 Introduction	255
	23.2 PoU in Criminal DNA Profiling	256
	23.2.1 ParaDNA™	256
	23.2.2 RapidHIT by IntegenX	257
	23.3 PoU in Point of Care Diagnostics	257
	23.3.1 Genie II/III™ by OptiGene	257
	23.3.2 QuantuMDx Q-POC [™]	258
	23.3.3 Biocartis Idylla™	258
	23.4 Discussion	259
	23.5 Conclusions	261
	Acknowledgements	261
	References	261
Chapter 24	Commercial DNA Testing	264
ompter 21	Barbara Hirst, Lourdes Fernandez-Calvino and	204
	Thomas Weiss	
	24.1 Introduction	264
	24.2 Expectations from Commercial Clients	265
	24.3 Are Expectations Always Deliverable?	266
	24.4 DNA Approaches	267
	24.5 Overview of Commercial DNA Tests	268
	24.5.1 DNA Detection Methods	268
	24.6 Comparison of Commercial DNA Tests and	
	In-house Methods	276
	24.7 Challenges in the Development of In-house	2.0
	DNA Test Methods	277
	24.8 Reporting PCR Results	278
	References	280
	A reasonable sets the sets of	200
Chapter 25	EU Food Integrity and Joining up the Landscape	
	(EU Perspective)	283
	Elena Maestri and Nelson Marmiroli	
	25.1 The Turning Point in EU Food Studies	283
	25.2 Research on Food in the EU	284
	25.2.1 The Beginning	285
	25.2.2 Research at the Turn of	
	the Century	289
	25.2.3 The Advent of Food Integrity	292
	25.2.4 Research in FP7	293
	25.2.5 Horizon 2020	293
	Acknowledgements	294
	References	294
		474

Contents		xxix
Chapter 26	The Food Authenticity Network	296
	Selvarani Elahi, Stephen Ellison and Mark Woolfe	
	26.1 Background	296
	26.2 Creation of the Food Authenticity	
	Network	297
	26.2.1 Network Structure	298
	26.3 Centres of Expertise (CoE) for Food	
	Authenticity Testing	299
	26.4 Authenticity Research and Methods	300
	26.5 Training	300
	26.6 Discussions and Forums	300
	26.7 Policy and Law	301
	26.8 Quality	301
	26.9 Relevant Organisations	301
	26.10 Latest News and Events	301
	26.11 Food Fraud Mitigation	302
	26.12 Social Media Platforms	302
	26.12.1 Twitter	302
	26.12.2 LinkedIn	303
	26.13 Growth	303
	26.14 Impact	303
	26.14.1 Membership	303
	26.14.2 Member Surveys	305
	26.14.3 Analytics Data	307
	26.15 International Recognition	307
	26.16 Global Network	309
	26.17 Conclusions	309
	References	310
Chapter 27	A Vision for the Future	311
	L. H. Foster, M. Burns and M. Walker	
	27.1 Introduction	311
	27.2 Changing World	312
	27.3 Food Fraud Prevention – Technological	
	Opportunities	312
	27.4 Analytical Developments and Future Needs	313
	27.5 Labelling and the Consumer	315
	27.6 Conclusions	315
	References	316
Subject Inde	ex	318