

FOOD INTEGRITY PROJECT - (WP10 BARILLA Coordinated Activity)

“Industry-perspective-matrix”

combined selection/risk assessment of strategic food chains vulnerabilities vs current methods and technologies adopted

- * Look for the most used/suggested (within the food chain) analytical techniques for assessing the required characteristics or discovering potential issues
- * Knowledge base of current trade standard methods and novel approaches for solving specific raw materials/finished products issues
- * Reorganize existing information from different sources, suppliers and copackers network, to identify problems and potential solutions.

Work Package 10



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RAW MATERIAL	TYPE OF RISK (fraud, adulteration, contamination)	TYPE OF ANALYSIS	COMPANIES WITH RELEVANT EXPERTISE TO BE CONTACTED/ENQUIRED	COMMENTS
Cocoa powder	Cross contamination with not declared allergens (e.g. nuts)	ELISA; LC-MS; PCR	Barry Callebaut, Belgium	High risks may be present only if cocoa powder is purchased from "non-producers". Big producers in this particular food chain, generally, do not have interest/cannot make frauds.
	Adulteration with cocoa of different geographical origin	Isotopic ratios of Strontium and Bioelements (H, C, O, N, S)		
	Partial addition of fibers of different origins and low value to increase dry matter levels	HPLC; Ashes content		
	Partial substitution of cocoa powder with carub tree powder	HPLC, Electrophoresis (protein fractions research); FTIR?		
Chocolate/Chocolate compounds	Adulteration with vegetable fats different from cocoa butter or hydrogenated fats	Trans fatty acids profile/presence. Fatty acids and tryglicerides composition.	Barry Callebaut, Belgium	High risks may be present only if cocoa powder is purchased from "non-producers". Big producers in this particular food chain, generally, do not have interest/cannot make frauds.
	Cross contamination with milk derivatives or dried fruit (e.g. nuts) allergens	ELISA; LC-MS; PCR		
	Addition of ingredients without quality and food safety characteristics requested by law (e.g. milk for feed, emulsifiers different from soy-lecithin, vegetable fats more than 5%, hazelnut paste cut with other components,...)	Different kinds of chemical analysis (GC-MS, LC-MS, UV, FTIR,...)		

	Possible problems with GMO material in soy-lecithin	GM analysis (SoyScreen); PCR real time		
Grana Padano PDO cheese (& Other PDO cheeses - e.g. Pecorino Romano, PDO Idiazabal Cheese, PDO Manchego Cheese, PDO Zamorano Cheese, PDO Serena Cheese, PDO Roncal Cheese, etc...)	Total or partial substitution of original PDO cheese in grated or shredded form with product deriving from foreign countries, without traceability	Stable isotope ratios of C, H, N, S; Trace element profile; Free aminoacids and pepetide profile; Volatile fraction profile	Grana Padano Cheese Consortium DOP, Italy PDO Cheeses Consortiums	Expensive raw materials, frauds do not depend on variability of price, it can be always possible
	Total or partial substitution of original PDO cheese in grated or shredded form with declassified PDO Grana Padano cheese or with processing scraps with following alteration of rind content	Free aminoacids and pepetide profile; Volatile fraction profile		
	Addition of foreign (not declared geographical origin) milk for the production of cheese	Stable isotope ratios of C, H, N, S ; Trace element profile; Genotyping (SNPs)		
	adulteration with milk of different animal species (e.g. sheep's milk instead of cow milk, etc...)	ELISA; LC-MS; qPCR (DNA probes), Metabolomic		
	Addition of milk ingredients (casein, milk powder)	Furosin content; Lyso-alanin content; Stable isotope ratios of C, H, N, S ; Trace element profile		
	Use in animal feed, of forages or raw material not allowed in the production specification of the cheese	Stable isotope ratios of C, H, N, S ; Trace element profile; Volatile fraction profile		
	Addition of B-Carotene in order to increase color	HPLC; LC-MS		
	Production not in compliance with the requirements of PDO protocol (e.g. milk must be exclusively from specific ovine breeds; possible inclusion of other breeds,...)	Different kinds of chemical analysis (GC-MS, LC-MS, UV, FTIR,...); Allele Specific Genotyping (real type PCR)		
Total or partial substitution of original PDO cheese in grated or shredded form with product deriving from foreign countries, without traceability	Stable isotope ratios of C, H, N, S; Trace element profile;			

Parmigiano Reggiano PDO cheese	Unpermitted use in animal feed of ensilage	Cyclopropilic acid content; Lysozyme content	Parmigiano Reggiano Cheese Consortium DOP, Italy	Expensive raw materials, frauds do not depend on variability of price, it can be always possible Multivariate statistical analysis: it must be robust and recognised in legal debates
	Use of not raw milk	Specific markers (eg. alkaline phosphatase activity)		
	Unpermitted refrigeration of milk	Specific markers (to be found)		
	Use in animal feed, of forages or raw material not allowed in the technical specifications of the cheese	Stable isotope ratios of C, H, N, S; Trace element profile; Volatile fraction profile; Other specific markers		
	Addition of B-Carotene in order to increase color	HPLC; LC-MS		
	Production not in compliance with the requirements of PDO protocol (http://www.parmigianoreggiano.com/consortium/rules_regulation_2/default.aspx)	Different kinds of chemical analysis (GC-MS, LC-MS, UV, FTIR,...); Allele Specific Genotyping (real time PCR)		
Spices & Aromatic Herbs	adulteration with other vegetables.	Genotyping by SSR to check adulteration with other aromatic plants. GC-MS Aromatic profile of the volatile oil to check adulteration with other aromatic plants (such as marjoram); Electronic nose? HPTLC analysis to check contamination of other botanicals. Visual, MIR and NIR microscopy, botanical identification for herbs.	Drogheria & Alimentari, Italy	Normally produced in non-advanced countries where controls are more difficult. Many buyers do not check quality but only the price
Wheat	adulteration, purity of the raw material or variety	Genotyping by SSR. Electrophoresis HPLC ELISA kits GMO analysis: TripleScreen	Relevant Grain Cooperatives in France or Italy; Cargill + Collaboration CRA-W with PROVIMI (Fr): NIR on-line measurement NUTRECO (NL): demonstration (NIR HIS) BIOWANZE (B) : assessment (NIR HIS)	Mainly due to lack of infrastructures
	Impurities, other varieties, toxic contaminants (Ergot, datura, ...)	NIR spectroscopy NIR hyperspectral imaging system		- Organic cereal production lead to a reemerging of some impurities (datura, wild seeds, ...) - In case of supplying problem or depending of the prices, mixtures common wheat/durum wheat, common wheat/spelt, farro/spelt can occur

Flours	Presence of added enzymes	Residual enzymatic activity	GMI (Grandi Mulini Italiani SpA, Italy); Soufflet Group (France)	<p>Linked to quality of wheat, some millers can see opportunities</p> <p>Note about "Whole-grain": <i>Almost universally, the term whole grain indicates inclusion of all three components of the cereal grain kernel – endosperm (this is the largest part of the grain and provides mostly starch), germ (comprises only a small part of the grain; this is where sprouting begins) and bran (the grain's protective outer layer; it is rich in dietary fibre). Variances, however, arise around the particular grains considered "whole", precise combination of the three components once processed, and processing practices which can affect the resulting flour's nutritional value. The HEALTHGRAIN definition addresses all three of these issues detailing a permitted list of grains and "pseudo grains" (such as quinoa and amaranth) and processing guidelines that take into account current milling practices.</i></p>
	Radioactive substances contamination	Radioactivity.		
	Cross contamination with toxic plants (datura stramonium)	Visual observation, possibility of setting up other chemical methods...		
	Partial substitution with other cereals/GMO	<p>Research of soy, corn, etc.. rice, potatoes. Species Ident. In the case of corn flour GMO analysis may be necessary (CornScreen)</p> <p>PCR Real Time</p> <p>FTIR</p> <p>NIRMicroscopy et MIRMicroscopy</p>		
	Not declared added enzymes to modify rheological properties of flours	Spectrophotometric/Fluorimetric methods;		
	Adulteration with flours of different geographical origin	Stable isotopes analysis.		
	Illegal use of ozone to treat flour	Analytical methods devoted to detect oxidation markers (LC-MS,...)		
	Potential allergens contamination	ELISA; LC-MS; PCR		
	Melamine addition to apparently increase the protein content/dry matter	LC-MS Analysis; FTIR		
	Whole grain composition in relation to industrial processes	Chemical analysis		
	Mushrooms from areas with high radiation levels	Liquid Scintillation Analyzers		

Dried mushrooms	Adulteration, purity of the raw material	<p>Botanical Analysis</p> <p>HPLTLC fingerprint for fungi spp identification.</p> <p>Genotyping by SSR to check the mushrooms species.</p> <p>Electronic nose? FTIR?</p>	Merlini, Italy	Many buyers look only for the prices and do not check quality
	Whitening treatments - to improve appearance	To be defined		
	Treatments with addition of starch to change the weight / firmness.	Enzyme methods, Colorimetric iodine method, HPLC-DAD		
	<p>1) ADDITION OF FOREIGN OIL TO EXTRA VIRGIN OLIVE OIL</p> <p>2) ADDITION OF REFINED OLIVE OIL TO EXTRA VIRGIN OIL</p> <p>3) ADDITION (OR COMPLETE SUBSTITUTION) OF NOT DECLARED GEOGRAPHICAL ORIGIN OLIVE OIL</p> <p>4) ADDITION OF OLIVE OIL WITHOUT QUALITY CHARACTERISTICS REQUESTED BY LAW</p> <p>5) ADDITION OF OLIVE OIL WITHOUT FOOD SAFETY CHARACTERISTICS REQUESTED BY LAW</p>	<p>Conventional analysis already requested by law as fatty acids and sterols composition, tryglicerides, minor components, etc.</p> <p>Unconventional methods, including those devoted to the determination of compounds related to</p>	<p>COMPANIES/INSTITUTIONS WITH RELEVANT EXPERTISE AT EU LEVEL:</p> <p>International Olive Council (IOC)</p>	

Vegetable Oils and Fats (SPECIAL FOCUS ON OLIVE OIL)	<p>6) RESEARCH OF NOT "CONVENTIONAL" OR "EMERGING" CONTAMINANTS OF OLIVE OIL</p> <p>7) ADULTERATION OF OLIVE OIL IN PREPARED FOOD (E.G. CANNED FOODS)</p> <p>8) ADULTERATION THROUGH ADDITION OF SEED OIL, HAZELNUT OIL, REFINED OR DEODORIZED OILS</p> <p>9) ADDITION OF CRUDE OILS (E.G. AVOCADO OIL) OR REFINED OILS (E.G. SEED OIL) TO VIRGIN OLIVE OIL</p> <p>10) MISLABELLING IN GEOGRAPHICAL DECLARATION (E.G. FALSE INFORMATION IN GEOGRAPHICAL PROVENANCE)</p> <p>11) AUTHENTICATION OF PDO VIRGIN OLIVE OILS (E.G. PDO VS NON PDO OILS)</p>	<p>to the determination of compounds related to quality (e.g. research of chlorophyll degradation products, pyropheophitin, pheophitin, other pigments & their derivatives, diacylglycerol isomers, volatile profiling, cold index, etc...)</p> <p>Research of markers by separate and non-separative techniques (e.g. NMR, isotopic spectroscopy, metabolomics, etc...)</p> <p>All methods mentioned above adapted, if possible, to prepared food in which olive oil is an ingredient</p>	<p>CRA, Italy Salov, Italy Deoleo, Spain Elourgiki, Greece Instituto de la Grasa, Spain ITERG, France SSOG, Italy Oleoestepa, Spain</p>	<p>MARKET AND COMMERCIAL ASPECTS ARE ABLE TO INFLUENCE THE INTERESTS FOR EVENTUAL ADULTERATIONS ALONG THE WHOLE SUPPLY CHAIN. ADULTERATION COULD OCCUR ALSO BY NON-VOLUNTARY ADDITION BUT ACCIDENTAL CROSS-CONTAMINATION (VESSELS, TANKS, OTHER TRANSPORTS OR STORAGES).</p> <p>MANUFACTURING INDUSTRY IS INTERESTED MAINLY IN THE FULL COMPLIANCE OF PURCHASED GOODS TO LAW QUALITY AND FOOD SAFETY PARAMETERS AND COMMERCIAL SPECIFICATIONS, IN ORDER TO HAVE GUARANTEE OF FINISHED PRODUCTS BY WITH THE LAW AND WITH CONSTANT QUALITY CHARACTERISTICS</p> <p>MANUFACTURING INDUSTRY NEEDS TO LINK WITH RELIABLE PARTNERS OF THE SUPPLY CHAIN BECAUSE THE CHAIN IS COMPLEX, WIDE, CROSSING WITH MANY SMALL ACTORS (I.E. FARMERS OR COOPERATIVES, TRADERS, ETC.)</p>
Butter	<p>Blending with bad quality or expired or fractionated butter</p> <p>adulteration with fats of different origin, as vegetable</p>	<p>Conventional analysis already requested by law as fatty acids and sterols composition, tryglicerides, minor components, etc.</p> <p>Unconventional methods, including those devoted to the determination of compounds related to quality (e.g. research of chlorophyll degradation products, pyropheophitin, pheophitin, other pigments & their derivatives, diacylglycerol isomers, volatile profiling, cold index, etc...)</p>	to be evaluated	<p>Some big industrial players usually directly sourcing from traders. Despite this, they traces the supply chain up to the milk fat producer and requires and verifies that the butter derives from milk cream</p>
	Intentional addition of incubated eggs	<p>Analysis of chemical markers such as organic acids, 3-OH-Butyrric Acid, Uracile only for shell eggs...</p> <p>Electronic nose</p>		<p>Market with high volatility and often not linked to feed prices. When feed price is too high any frauds can happen. Sometimes the market is short of eggs and at least 6 months are needed before having enough eggs.</p>
	Use of mineral oils instead of vegetable oils in hens feed / eggproducts to apparently increase the fat content	GC-FID, GC-HRMS, GC-MSMS		

Eggs/Eggproducts	Commercialization of eggproducts obtained during the quarantine period after hens medical treatments (vet drugs)	LC-MS	Eurovo, Italy	<p>Uracil in the eggproduct can not be considered necessary as an indicator of fraud; this is because its potential presence could be also connected only to general deterioration of the product (which can also be caused by poor storage after production)</p> <p>The addition of artificial colorants is due only to the egg itself: the presence in the egg shell is a result of the addition in the feed of laying hens</p> <p>Eggs classified into Category "B" are more related to an overall aspect of traceability. In fact, eggs, even if classified into Category "A" can quickly deteriorate if not properly preserved</p>
	Melamine addition to apparently increase the protein content/dry matter in eggproducts (egg white mainly)	LC-MS Analysis, FTIR,...		
	Addition of artificially coloured eggs	Research in HPLC of extraneous pigments molecules		
	Partial or complete substitution with eggs without quality and food safety characteristics requested by law and by the contract (as "fresh eggs" or A category)	Conventional chemical and microbiological markers able to detect and classify the level of freshness and quality Electronic nose		
Tomatoes and Derivatives	Partial or complete substitution with GM tomatoes	ELISA; PCR	Consorzio Casalasco, Italy	
	Adulteration with tomatoes of different geographical origin [e.g.: tomato concentrate imported from China, reworked in Italy and sold as Italian origin; geographic origin,...]	Stable isotopes analysis.		
	Melamine addition to apparently increase the protein content in powdered milk	LC-MS Analysis, FTIR,...	To be evaluated	<p>High request of milk powders ww; at each stage of the food chain operators can do frauds to increase productions or to earn more money</p>
	Adulteration with milk of not declared geographical origin	Stable isotopes analysis.		
	Type of animal, if declared (cow, sheep, bufalo, etc...). Adulteration with milk of different animal species (e.g. sheep's milk instead of cow milk, etc...)	ELISA; LC-MS; qPCR (DNA probes), Metabolomic		
	Addition of non/milk fat&oil into dairy products	Fatty acids and tryglicerides composition&profiles (e.g. by GC-MS, DART-HRMS, MALDI,...)		

Milk and Derivatives	Addition of water	Crioscopic point; Vapor Pressure Osmometry; Conductivity		
Enzymes	Formulation often not completely clear/declared. Risks also associated to the cross contamination during the preparation phase (e.g. chloramphenicol presence). Further issue: enzyme genetically modified instead to be obtained from GM microorganisms	ELISA; LC-MS; PCR	DSM; Danisco	It should be noted that the enzymes are not declared on the label by law (there is no requirement in Europe)
Meat	use of adulterated feed (e.g. dioxin case in 2011 because of mixing with dioxin-containing vegetable fats for animal feed,...)	GC-HRMS, GC-MSMS	Unipeg, Italy Italcarni, Italy	Different meats vary a lot in term of price per kg. In some period there can be lack of availability of some type of meat. There is often on the market low quality meat (also out of law) sold at very low prices. Horse scandal, substitution of calf with pork, halal food etc...this misdescription is generally frequent in ready to eat food like sausages, burgers etc...
	Addition of blood proteins, non/meat proteins	Immunoassays, Electrophoresis		
	Addition of mechanically recovered meat	ELISA; Microscopy		
	Frauds related to differentiation between fresh and frozen/thawed meat	Enzymatic methods; NIR		
	Irradiated meat	ESR; Microbiological analysis		
	Meat declared 100% "Hormones free"	ELISA; LC-MS; Biosensors		
	adulteration/fraud; adulteration with meat of different animal species (e.g. Horse-meat scandal 2012,...)	ELISA; LC-MS; qPCR (DNA probes), Metabolomic		
Coffee	Geographical origin	HSGC Volatile Profiling, HPLC (chlorogenic acids determination), SNIF-NMR, IRMS	Illy SpA, Italy	
	Sugars addition (soluble coffee)	HPAE-PAD		
	Species mixtures (Arabica vs robusta varieties)	qPCR (DNA probes), FTIR, NIR		

Fruit-based preparations (juices, jam, dietary products originated from); Pickles	Use of ascorbic acid of industrial source to substitute natural vitamin C present in a fruit juice or in a dietary product originated from the claimed fruit	Isotope Ratio MS (13C-IRMS)	AEAZN, Asozumos, SGF, Qualijus, Ponti.	An example of fraud: substitution with mandarin which is, generally, cheaper than orange and lemon
	Juice adulteration; species mixtures with cheaper fruits	qPCR (DNA probes)		
	botanical and/or varietal origin of raw material	Isotope Ratios of bioelements (H, C, O) Elemental profile (ICP-MS, OES) NMR, MS profiling		
	Geographical origin of vinegar and must, if declared	Isotope Ratios of bioelements (H, C, O) and Sr Elemental profile (ICP-MS, OES) NMR profiling		
	Presence of jelling agents or additives, not declared	Specific markers (to be found)		
	Artificial flavours addition	Chiral-GC, HPLC		
	Thermic treatments not declared	Specific markers (to be found)		
	Use of undeclared sweeteners, addition of citric-malic-tartaric acids	IRMS, SNIF-NMR, IC, HPAEC-PAD		
Seafood; fresh/canned fish; Surimi	Seafood from areas with high radiation levels	Liquid Scintillation Analyzers	Marengo, Italy AIPCE-CEP, the EU Fish Processors and Traders Association.	Some kind of fish are very cheap and can be used to increase earnings lack of transparency in raw material, illegal fishing, sometimes one species (i.e. albacore) is replaced by cheaper species (i.e. bigeye, yellowfin)
	Substitution/mislabelling frauds; Species mixtures with cheaper varieties	ELISA; LC-MS; qPCR /DNA probes)		
	Adulteration with fish of not declared geographical origin	Genotyping (SNPs), IRMS		
Honey	Country of origin issues. Authenticity. Risks also associated with contamination during the preparation phase (e.g. veterinary drugs presence).	IRMS, LC-IRMS, HR-NMR, NMR Profiling	European Federation of Honey Packers & Distributors	Honey used as ingredient by many food producers and processors. Certifying Mauka Honey (e.g. 10 times more sold than produced).

Vinegar/balsamic vinegar of Modena (ABM)	Ethanol in ABM must and acetic acid from beet and cane sugar	Isotope Ratios of H and C of ethanol and acetic acid; Sugar profile by HPLC in ABM must	Ponti, Italy	<p>According to EC regulations, wine vinegar cannot contain acetic acids obtained from either petroleum derivatives or pyrolysis of wood (synthetic acetic acid) or from the fermentation of non-grape sugars (e.g. from beet or cane).</p> <p>Moreover, wine and wine vinegar cannot be produced from dried grapes diluted with water.</p> <p>This also applies to 'Aceto Balsamico di Modena', a PGI (Protected Geographical Indication) salad dressing ingredient now renowned throughout the world, obtained from cooked and/or concentrated grape must (at least 20% of the volume), with the addition of at least 10 % of wine vinegar and a maximum 2% of caramel for color stability (EU Reg. 583/2009).</p>
	Dilution of dried grapes	Isotope ratio of O in vinegar water		
	Geographical origin of vinegar and must, if declared	Isotope Ratios of bioelements (H, C, O) and Sr; Elemental profile (ICP-MS, OES); NMR profiling		
	Presence of caramel when not declared	Specific markers (to be found); Detection of some characteristics of the product due to the presence of caramel (e.g colour)		
	Synthetic acetic acid	C14 determination; Stable isotope ratio of C in acetic acid		
	Presence of mannoproteins and xanthan (not allowed), too high content of gum arabic (o acacia gum), galactomannans, tannins to improve structure and flavour of ABM	Specific markers (to be found)		
	Addition of flavour	Specific markers (to be found); Compound specific stable isotope ratios; Analysis by GC-IRMS		
Beverages e.g. brandy, wine, whisky	Country of origin issues. Authenticity. Adulteration through addition of cheaper sugar/alcohol/water	IRMS; Spectroscopic techniques (e.g. RAMAN); LC-MS/MS, QToF-MS; SNIF-NMR; GC-FID, GC-MS; LC-UV, LC-IEC	SpiritsEUROPE (EU spirit drink producers association) The Scotch Whisky Research Institute, UK	Beverages e.g. wine, brandy, whisky used as ingredient by many food producers and processors

Further Aspects of potential interest to be considered along some of the food chains cited above:

Organic vs Conventional raw materials/finished products distinctions

Animal welfare demonstration (e.g. declaration of poultry farming in cage systems or not...)