Fluorescent Residues on Food-Processing Surfaces (UV-A 365 nm)

Introduction: Ultraviolet (UV-A, ~365 nm) lighting is often used in food industries to inspect cleanliness and detect contamination. Many food-derived compounds **fluoresce** (absorb UV and re-emit visible light) due to their chemical structures (aromatic rings, conjugated double bonds, etc.). Below we highlight common **fluorescent substances** found as residues on equipment surfaces – including proteins, fats, vitamins, microbial metabolites, and mycotoxins – explaining why they glow and where they originate.

Proteins and Aromatic Amino Acids

- **Tryptophan, Tyrosine (Proteins):** Proteins containing aromatic amino acids (tryptophan, tyrosine, phenylalanine) exhibit intrinsic UV fluorescence (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC). The indole ring of tryptophan (and phenolic ring of tyrosine) absorbs UV (~280 nm) and emits in near-UV or violet wavelengths (~300–350 nm). Under a 365 nm lamp, protein residues can still produce a faint bluish-white glow due to these aromatic rings. **Examples:** Milk or egg residues (rich in protein) left on stainless steel will fluoresce, helping sanitation crews spot unclean spots (Handheld Multispectral Fluorescence Imaging System to Detect and Disinfect Surface Contamination - PMC). Meat juices or biofilms on equipment similarly show fluorescence from bacterial and tissue proteins. In meat products, such protein-derived fluorescence is observed in the UV/blue range (Handheld Multispectral Fluorescence Imaging System to Detect and Disinfect Surface Contamination - PMC), aiding detection of invisible films.

- **Crosslinked Proteins:** Food processing (heating or drying) can create *dityrosine* crosslinks (two tyrosines bonded), which have extended conjugation. Dityrosine emits blue fluorescence, further enhancing the glow of cooked-on protein residues. **Example:** Baked-on dairy or soy residues containing dityrosine will fluoresce blue under UV, signaling areas requiring re-cleaning (Fluorescence of Intrinsic Milk Chromophores as a Novel Verification Method of UV-C Treatment of Milk - PMC) (Fluorescence of Intrinsic Milk Chromophores as a Novel Verification

Method of UV-C Treatment of Milk - PMC).

Lipids and Fats (Conjugated Double Bonds)

- **Oxidized Fats and Grease:** Pure fresh fats (e.g. oil, lard) are weakly fluorescent, but **unsaturated fats** form *conjugated double bonds* when oxidized (rancidity) that fluoresce under UV (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC). Conjugated lipid peroxides absorb UV and emit blue light (~420–450 nm). **Examples:** A thin film of used cooking oil or meat grease on equipment will glow blue-white under 365 nm, indicating incomplete cleaning (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC). Research shows oxidized vegetable oils have a strong fluorescence band around 430–450 nm (Evaluation of the overall quality of olive oil using fluorescence spectroscopy - PubMed), correlating with rancidity. Thus, a greasy residue on a machine (even if invisible) can be revealed by its blue fluorescence from these conjugated dienes.

- **Chlorophyll in Oils:** Unrefined oils (like extra virgin olive oil) contain minor pigments that fluoresce. **Chlorophyll** (a porphyrin) in olive oil emits red (~680 nm) under UV (Evaluation of the overall quality of olive oil using fluorescence spectroscopy - PubMed). **Example:** A droplet of olive oil on a countertop will glow dull red under 365 nm due to residual chlorophyll. This red fluorescence is a telltale sign of plant-based oil residues.

- **Carotenoids and Oxidation Products:** Carotenoids (vitamin A precursors) have extended conjugation but typically quench fluorescence (emitting little light). However, thermal or oxidative breakdown can produce fluorescent polyenes. **Example:** Overheated palm oil (rich in carotenes) may show a faint greenish fluorescence from degraded carotenoids. In general, bright fluorescence from fat residues usually signifies oxidation or the presence of other fluorophores (like vitamins) rather than the fat itself (Evaluation of the overall quality of olive oil using fluorescence spectroscopy - PubMed).

Vitamins (Intrinsic Food Fluorophores)

- **Riboflavin (Vitamin B_2):** Riboflavin's isoalloxazine ring system is a potent fluorophore. It absorbs UVA and emits bright green-yellow light. In milk and dairy, riboflavin fluoresces with a peak around 520–535 nm (Fluorescence of Intrinsic Milk Chromophores as a Novel Verification Method of UV-C Treatment of Milk -

PMC). **Examples:** Dried milk or whey on surfaces will glow yellow-green under 365 nm due to riboflavin (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC). Dairy processors even use a “riboflavin test” by coating equipment with riboflavin solution and checking UV fluorescence to validate cleaning coverage (since any residual riboflavin glows visibly) ([PDF] Riboflavin Coverage Test | Labconco). This vitamin’s fluorescence stems from its conjugated isoalloxazine ring.

- **Vitamin A (Retinol/Retinyl esters):** Vitamin A has a long conjugated polyene tail and a β-ionone ring, giving it fluorescence in the blue-green range. Retinol absorbs near 330–350 nm and can emit greenish light. **Examples:** Grease from animal fat or dairy cream contains retinyl esters that fluoresce. Butter or cheese smears (rich in vitamin A) on equipment may show a weak green-blue glow under UVA. Vitamin A was identified among key natural food fluorophores (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC), and its polyene structure underlies this fluorescence.

- **Vitamin E (α-Tocopherol):** Tocopherols (vitamin E) are phenolic compounds in fats that fluoresce green. In olive oil, for instance, a strong fluorescence band at ~525 nm is attributed to α-tocopherol (Evaluation of the overall quality of olive oil using fluorescence spectroscopy - PubMed). **Example:** Vegetable oil residues on a surface emit green light under UV, partly due to vitamin E content. The aromatic chromanol ring of tocopherol and its conjugated tail enable this fluorescence. This trait has been used to assess olive oil quality – fresh extra-virgin oil shows a distinct 525 nm (green) fluorescence from vitamin E (Evaluation of the overall quality of olive oil using fluorescence spectroscopy - PubMed).

- **Other B Vitamins:** Certain B vitamins or derivatives fluoresce as well. **Pyridoxine (B_6)** derivatives (e.g. pyridoxal-5-phosphate) are fluorescent (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC), and **folic acid** (B_9) exhibits blue fluorescence in solution. While less prominent on surfaces, residues of vitamin-fortified foods or supplements could be detected by their characteristic emissions.

Microbial Metabolites and Biomarkers

- **NADH/NADPH (Reduced Nicotinamide Adenine Dinucleotide):** This coenzyme, present in all living cells (including bacteria), strongly fluoresces under UV excitation. NADH has an aromatic nicotinamide ring; excited at ~340 nm, it emits blue (~450 nm) light. **Examples:** A high bioburden (microbial load) on a surface – say a biofilm or residue of bacterial growth – will show increased blue fluorescence thanks to cellular NADH (Understanding Real-Time Fluorescence Signals from Bacteria and Wound Tissues Observed with the MolecuLight i:XTM - PMC). Laboratory studies confirm bacteria emit a blue autofluorescence (400–450 nm) proportional to their concentration (Detecting Bacterial Biofilms Using Fluorescence Hyperspectral Imaging and Various Discriminant Analyses - PMC), largely from NADH. Thus, ATP-bioluminescence tests aside, UV inspection can reveal microbial contamination by this blue glow.

- **FAD and FMN (Flavins in Metabolism):** Flavin adenine dinucleotide (FAD) and flavin mononucleotide (FMN) are coenzymes (derived from riboflavin) that fluoresce green. In microbes, FAD (oxidized form) emits green (~520 nm) when excited by UVA (Understanding Real-Time Fluorescence Signals from Bacteria and Wound Tissues Observed with the MolecuLight i:XTM - PMC). **Example:** Bacterial films often contain fluorescent flavins; under 365 nm light, colonies can exhibit greenish fluorescence from FAD/FMN. Indeed, combined blue (NADH) and green (FAD) emissions form the basis of some optical bacterial detection devices (Understanding Real-Time Fluorescence Signals from Bacteria and Wound Tissues Observed with the MolecuLight i:XTM - PMC).

- **Bacterial Pigments (Siderophores):** Certain microbes produce fluorescent pigments. A notable example is **pyoverdine**, a yellow-green siderophore secreted by *Pseudomonas* species. Pyoverdine (also called fluorescein in older literature) is a dihydroxyquinoline derivative that glows green under UV (Pyoverdine - an overview | ScienceDirect Topics). **Examples:** *Pseudomonas fluorescens*, a common spoilage bacterium in meats and dairy, often deposits a fluorescent pigment on surfaces. Poultry processors noted that spoiled chicken skin or eggs with *Pseudomonas* can be checked with UV – colonies of *Pseudomonas* emit a greenish fluorescence, revealing contamination ((PDF) Determination of Pyoverdine on the Surface of Chicken Meat). Pyoverdine's conjugated ring system is responsible for its bright fluorescence, which has even been used in research as a marker for spoilage progression.

- **Indoles and Metabolites:** Bacteria that metabolize tryptophan can excrete indole or other aromatic metabolites that fluoresce. For instance, some *Bacillus* species produce *para*-hydroxyphenylpyruvate (a tyrosine breakdown product) that emits blue light. **Example:** High levels of microbial spoilage in protein-rich residues might generate additional fluorescent compounds (e.g. kynurenine, a tryptophan metabolite with strong fluorescence (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC)). While not specific monitors, these contribute to the overall fluorescence signal of microbial soils.

Plant & Animal Pigments (Contamination Indicators)

- **Chlorophyll and Derivatives:** Chlorophyll **a** in plant matter is a classic fluorophore – its porphyrin ring (a large conjugated system) emits deep red light. Intact chlorophyll fluoresces red (~670–685 nm) under UV or blue excitation (Handheld Multispectral Fluorescence Imaging System to Detect and Disinfect Surface Contamination - PMC). On food surfaces, chlorophyll usually appears via contaminants like feces or field dirt. **Examples:** Fecal contamination from herbivorous animals on produce or carcasses is detected by red fluorescence of chlorophyll breakdown products (Handheld Multispectral Fluorescence Imaging System to Detect and Disinfect Surface Contamination - PMC). Dairy cow manure on lettuce, or ingesta on meat, shows a red glow (~680 nm) under 365 nm excitation, due to pheophorbide and related chlorophyll metabolites in digested grass (Handheld Multispectral Fluorescence Imaging System to Detect and Disinfect Surface Contamination - PMC). Inspectors use multi-spectral UV lamps to spot these red-fluorescent specks, improving food safety by removing fecal matter. (Notably, this method exploits chlorophyll's resonance; the conjugated tetrapyrolle structure yields far-red emission.)

- **Porphyrins (Heme Derivatives):** Heme itself (with iron) is non-fluorescent, but if blood residues age and the iron is lost, free porphyrins (e.g. protoporphyrin IX) can fluoresce red. **Example:** Old blood or meat juice residues might show a dull red-brown fluorescence under UV if porphyrin has been liberated. Similarly, *Coprophilous* molds or bacteria can produce porphyrin pigments on surfaces. While not a routine monitoring target in sanitation, porphyrin fluorescence can occasionally indicate remnants of blood or bile on equipment.

- **Plant Polyphenols:** Some plant-derived contaminants (fruit juices, polyphenol-rich residues) exhibit fluorescence due to aromatic rings. For instance, **aflatoxin precursors** in moldy grains come from polyketide pathways that include fluorescent intermediates. Also, spilled beer or wine (rich in polyphenols) can show faint blue-green fluorescence when dried. These cases are less common but contribute to the spectrum of UV-detectable residues.

Mycotoxins (Fluorescent Fungal Metabolites)

- **Aflatoxins:** Aflatoxins are notorious carcinogenic mycotoxins produced by *Aspergillus* molds on foods like peanuts, corn, and spices. Chemically, they are **bifuranocoumarin** compounds with highly conjugated structures, making them strongly fluorescent. Under 365 nm UV, contaminated grains often show a distinctive glow. **Aflatoxin B_1/B_2** emit **blue** fluorescence (peak ~425–450 nm), whereas **aflatoxin G_1/G_2** emit **green** (peak ~540 nm) (Aflatoxin Contamination, Its Impact and Management Strategies: An Updated Review - PMC). This color difference is so consistent that the toxins were named "B" (blue) and "G" (green) based on their fluorescence (Different types of aflatoxin - Groundnut Academy). **Examples:** Peanut kernels with *Aspergillus* infection will fluoresce bright greenish-yellow if G-type aflatoxins are present, or blue if B-type are present. Inspectors often use 365 nm "black lights" to screen crops – aflatoxin-contaminated nuts show a **bright greenish-yellow fluorescence (BGYF)** on the surface (Different types of aflatoxin - Groundnut Academy). The coumarin-like rings in aflatoxins are responsible for this glow. (Note: UV screening is a preliminary test; positive fluorescence indicates possible aflatoxin, which is then confirmed by analytical chemistry (Use of cyclodextrins as modifiers of fluorescence in the detection of ...).)

- **Ochratoxin A (OTA):** Ochratoxin A, a toxin from *Aspergillus* and *Penicillium* species (common in cereals, coffee, grapes), is an **isocoumarin** derivative linked to phenylalanine. It is colorless but exhibits blue or green fluorescence under UV, depending on conditions (Low cost optical device for detection of fluorescence from Ochratoxin ...). Typically, OTA has an excitation around 333 nm and an emission ~460 nm (blue) in neutral/alkaline solution, shifting to greenish in acidic environment (Low cost optical device for detection of fluorescence from Ochratoxin ...). **Examples:** Moldy coffee beans or contaminated barley malt may show a blue-green fluorescence spot attributable to OTA. Thin-layer chromatography of OTA makes use of its native fluorescence for detection (Colorimetric Analysis of Ochratoxin A in Beverage Samples). The extended conjugation in the isocoumarin ring system is the basis for OTA's fluorescence. In sanitation contexts, OTA on surfaces isn't a usual concern (since it's a toxin in foods), but its presence can be monitored in analytical labs by UV fluorescence.

- **Other Mycotoxins:** Some other mycotoxins fluoresce as well. **Zearalenone** (from *Fusarium* on corn) is a resorcylic acid lactone with weak blue fluorescence. **Sterigmatocystin** (an *Aspergillus* toxin) is structurally similar to aflatoxin and has fluorescence under UV. However, many mycotoxins (e.g. trichothecenes like DON, fumonisins) lack inherent fluorescence due to less conjugation. Thus, UV inspection most notably highlights the aflatoxin family and OTA. In contaminated food facilities (e.g. peanut processing), UV lamps can pinpoint areas of mold growth by the telltale blue/green glow of these toxins, prompting thorough cleanup.

References (Selected Studies & Reviews)

- Simeone *et al.*, **Sensors (2018):** Lists intrinsic food fluorophores including aromatic amino acids in proteins, vitamins A & B_2, NAD(P)H, and chlorophyll (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC). Also notes UV fluorescence can detect grease (conjugated hydrocarbons) on equipment (Enhanced Clean-In-Place Monitoring Using Ultraviolet Induced Fluorescence and Neural Networks - PMC).
- Lepore *et al.*, **Foods (2021):** UV imaging for sanitation – proteins emit in UV, aromatic compounds emit blue/green (Handheld Multispectral Fluorescence Imaging System to Detect and Disinfect Surface Contamination - PMC). Demonstrated red 680 nm fluorescence from chlorophyll in feces under 360 nm excitation (Handheld Multispectral Fluorescence Imaging System to Detect and Disinfect Surface Contamination - PMC).
- Khanna *et al.*, **J. Dairy Sci. (2022):** Monitored milk oxidation by fluorescence – riboflavin emits ~534 nm in dairy (Fluorescence of Intrinsic Milk Chromophores as a Novel Verification Method of UV-C Treatment of Milk - PMC). Noted similar emission from porphyrin (heme) and chlorin compounds in milk (Fluorescence of Intrinsic Milk Chromophores as a Novel Verification Method of UV-C Treatment of Milk - PMC).
- Guzmán *et al.*, **Food Chem (2015):** Characterized olive oil by fluorescence – fresh extra virgin oil shows bands at 440–455 nm (polyphenols), a strong green band at 525 nm (vitamin E), and red at 681 nm (chlorophyll) (Evaluation of the overall quality of olive oil using fluorescence spectroscopy - PubMed). Oxidized oils had a prominent 430–450 nm fluorescence, correlating with peroxide value (Evaluation of the overall quality of olive oil using fluorescence spectroscopy - PubMed).
- MolecuLight Device Tutorial (2019): Describes bacterial autofluorescence – **blue** from NADH, **green** from flavins (Understanding Real-Time Fluorescence Signals from Bacteria and Wound Tissues Observed with the MolecuLight i:XTM - PMC). These endogenous fluorophores allow real-time imaging of bacterial load in wounds.
- Pseudomonas Spoilage Studies: Noted many *Pseudomonas* secrete a fluorescent yellow-green siderophore (pyoverdine) detectable under UV (Pyoverdine - an overview | ScienceDirect Topics). Cotterill (1956) first suggested UV lamps to detect *Pseudomonas* contamination on meat by this fluorescence ((PDF) Determination of Pyoverdine on the Surface of Chicken Meat).
- Iqbal *et al.*, **Toxins (2022):** Aflatoxin review – B_1/B_2 show blue fluorescence (~425 nm) and G_1/G_2 green (~540 nm) under 365 nm (Aflatoxin Contamination, Its Impact and Management Strategies: An Updated Review - PMC). This property is used in screening peanuts and corn for "bright greenish-yellow" glowing kernels (high aflatoxin risk).
- Bueno *et al.*, **Sensors (2016):** Ochratoxin A emits fluorescence under UV light (Colorimetric Analysis of Ochratoxin A in Beverage Samples). OTA's green or blue

fluorescence can be observed in different pH environments (Low cost optical device for detection of fluorescence from Ochratoxin ...), facilitating its detection in contaminated beverages.