Impact of Emerging Scientific Methods on Food Safety Policy: Issues and Examples

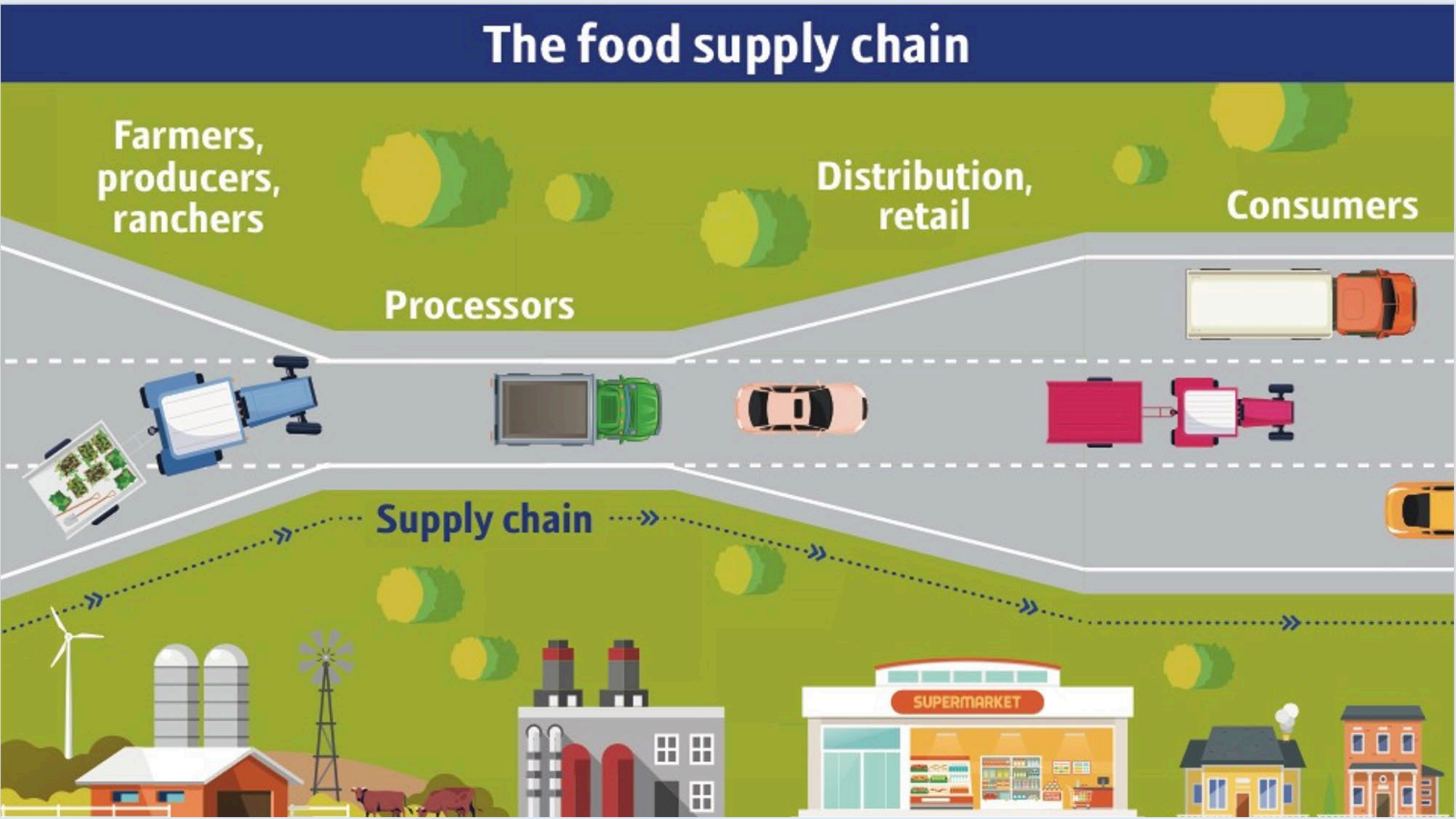
Lee-Ann Jaykus, Ph.D.

William Neal Reynolds Distinguished Professor

Dept. of Food, Bioprocessing and Nutrition Sciences

NC STATE UNIVERSITY

The food supply chain



Adulteration (Relevant Clauses), 21 U.S.C. §342

- Food that contains any poisonous or deleterious (injurious to health; harmful) substance,
  - *Exception: "but in case the substance is not an added substance, such food shall not be considered adulterated ... if the quantity of such substance in such food does not ordinarily render it injurious to health."*
  - *Zero tolerance (presence/absence) vs. tolerance (allowable amount)*
- Food that has been prepared, packed, or held under insanitary conditions whereby it **may have become** contaminated with filth, or whereby it may have been rendered injurious to health
  - *Note, the food does not have to be unsafe in order for it to be adulterated; even food that is safe to consume will be considered adulterated if it passed through an area that is insanitary and that **could** have contaminated the food.*
- Additives, microbes, and conditions that could lead to contamination; 21 CFR 110.5; must not have conditions that can lead to contamination
  - *Note that this prohibition does not address only the food product, but the conditions under which the food product is manufactured; thus this prohibition extends the reach of the regulatory agency.*
- The prohibitions against adulterated foods under U.S. federal law primarily apply to processing. State law also generally prohibits adulterated and misbranded foods; these laws usually apply to the processing and retail/food service sectors
- Food processors are required to establish that their food product meets legal requirements/standards; that is, establish that the food is not adulterated or misbranded. This is not the government's responsibility

How is Adulterated Food Identified?

- Pathogens enter the food chain mostly unintentionally
- Identification by:
  - Epidemiological investigation (illnesses, i.e., outbreak)
  - **Routine microbiological testing**
    - Industry
    - Regulatory
  - Both?
- What is the general source of contamination?
  - Environmental (e.g., *Listeria monocytogenes*)
  - Animal feces (e.g., *Salmonella, Campylobacter*)
  - Human feces (e.g., *Shigella*, norovirus, hepatitis A virus)
- Where does the contamination occur?
  - Farm, processing plant, restaurant, etc.
- What are the extenuating factors?

Emerging Scientific Methods and Implications

- Epidemiological surveillance (FoodNet, PulseNet, Genometrakr)
- Molecular-based detection methods—Polymerase chain reaction (PCR)**
- Next generation sequencing (NGS), metagenomics, and bioinformatics**
- Mathematical modeling and risk assessment**
- Social media and crowd sourcing (e.g., iwaspoisoned.com)**
- Gene editing (CRISPR-Cas9)/genetic engineering

Example #1: Non-Cultivable Foodborne Pathogens

Conventional Culture Methods--GOLD STANDARD

Pre-Enrichment

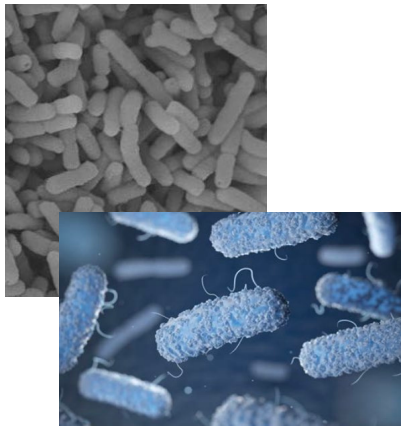
Selective Enrichment

(Rapid Methods) Selective Plating (PCR, ELISA)

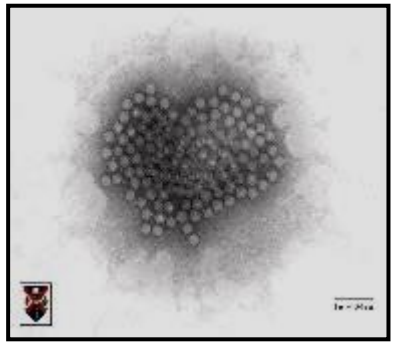
Confirmatory Assays

PCR/Sequencing | Biochemical | Serological

It's ALIVE!!
There is a culture to prove it



Non-Cultivable Pathogens--Enteric Viruses

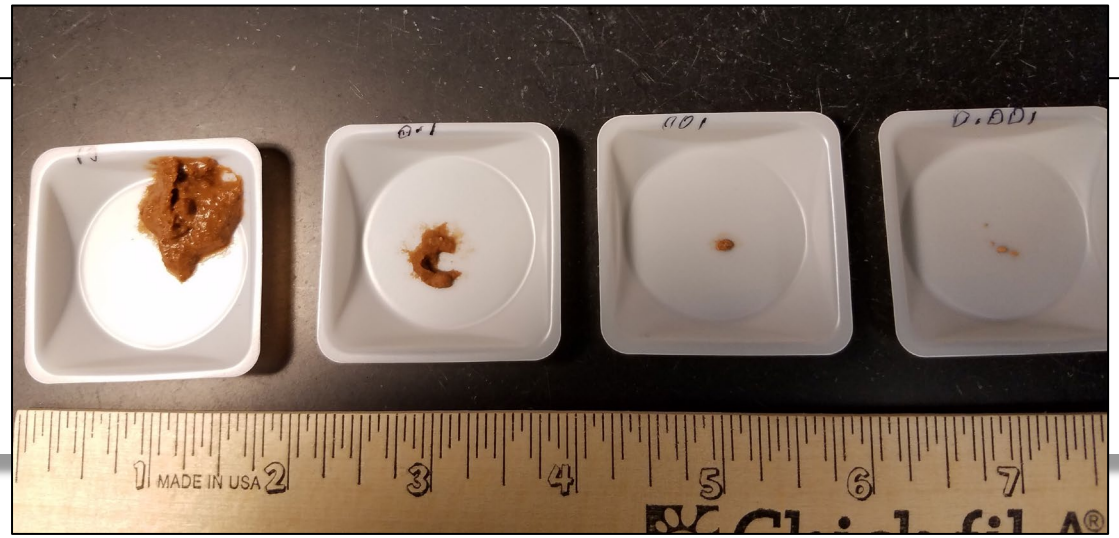


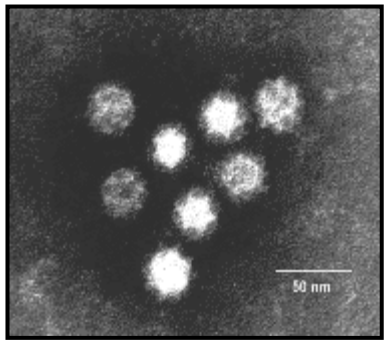
Hepatitis A virus

-Picornaviridae

- Self-limiting hepatitis syndrome
- Most severe of the foodborne viral diseases
- Reportable disease

- Simple structure
  - Protein coat (capsid)
  - Nucleic acid core (RNA)
- Very small (20-50 nm diameter)
- Fecal-oral transmission: ***Human*** fecal material
- RTE foods (restaurants) and produce



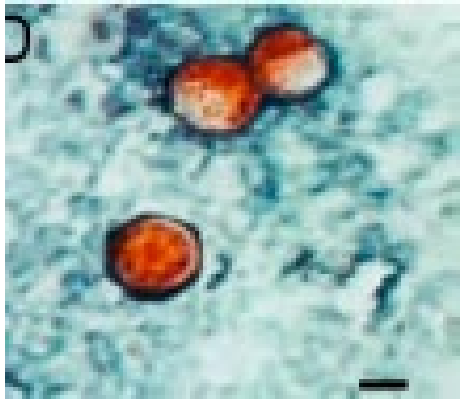


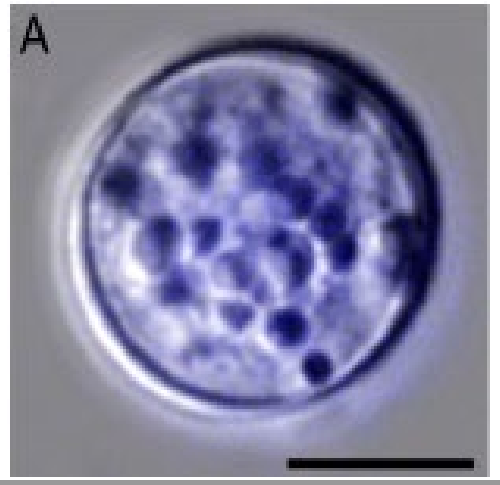
Human Norovirus

-*Caliciviridae*

- Self-limiting vomiting and diarrhea in adults and children
- Leading cause of viral gastroenteritis and foodborne disease
- Not a reportable disease

Non-Cultivable Pathogens--Parasitic Protozoa





- Much larger than bacteria (eukaryotes)
- Complex life cycle
- Of greatest foodborne significance
  - ***Cyclospora cayetanensis***
- Gastrointestinal illness
- Fecal-oral transmission (***humans***)
  - Water
  - Feces
  - Soil
  - Other sources?
- Fresh produce

Detection of Non-Cultivable Pathogens in Foods

Pre-Enrichment

Selective Enrichment

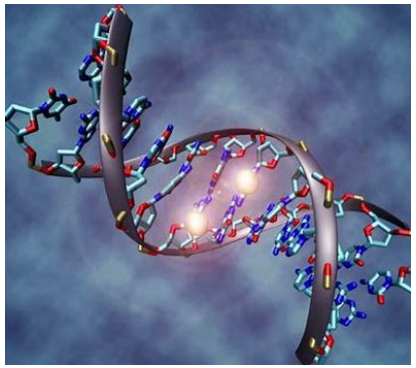
Selective Plating

Detection Assay

PCR

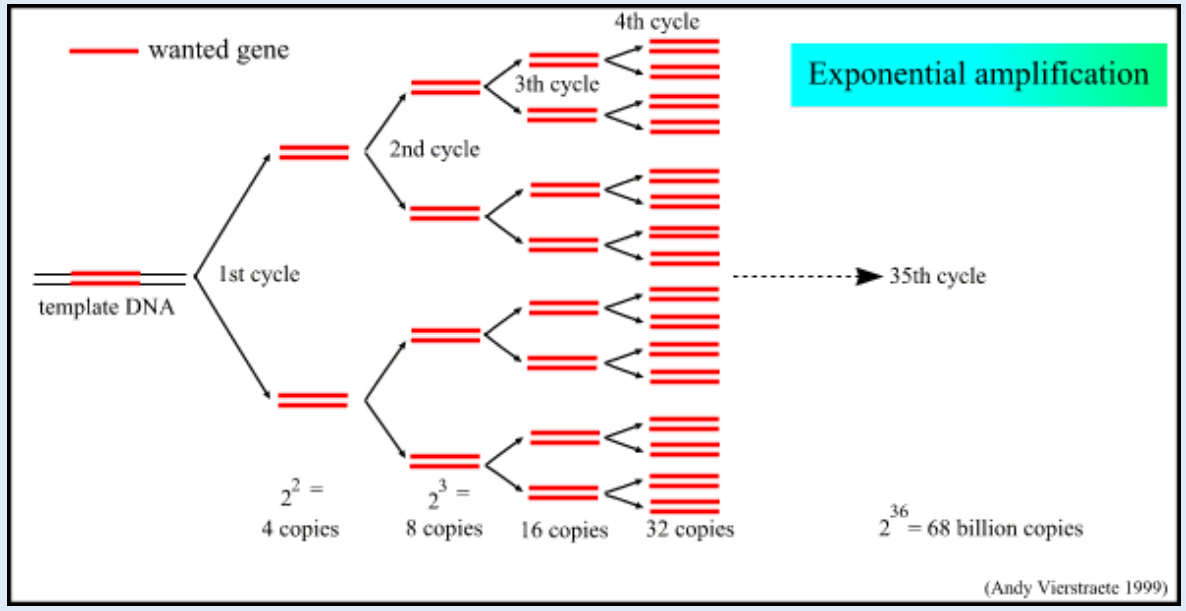
CONCENTRATION
PURIFICATION
NA EXTRACTION

End product is an amplified piece of DNA
Is/was it ALIVE????



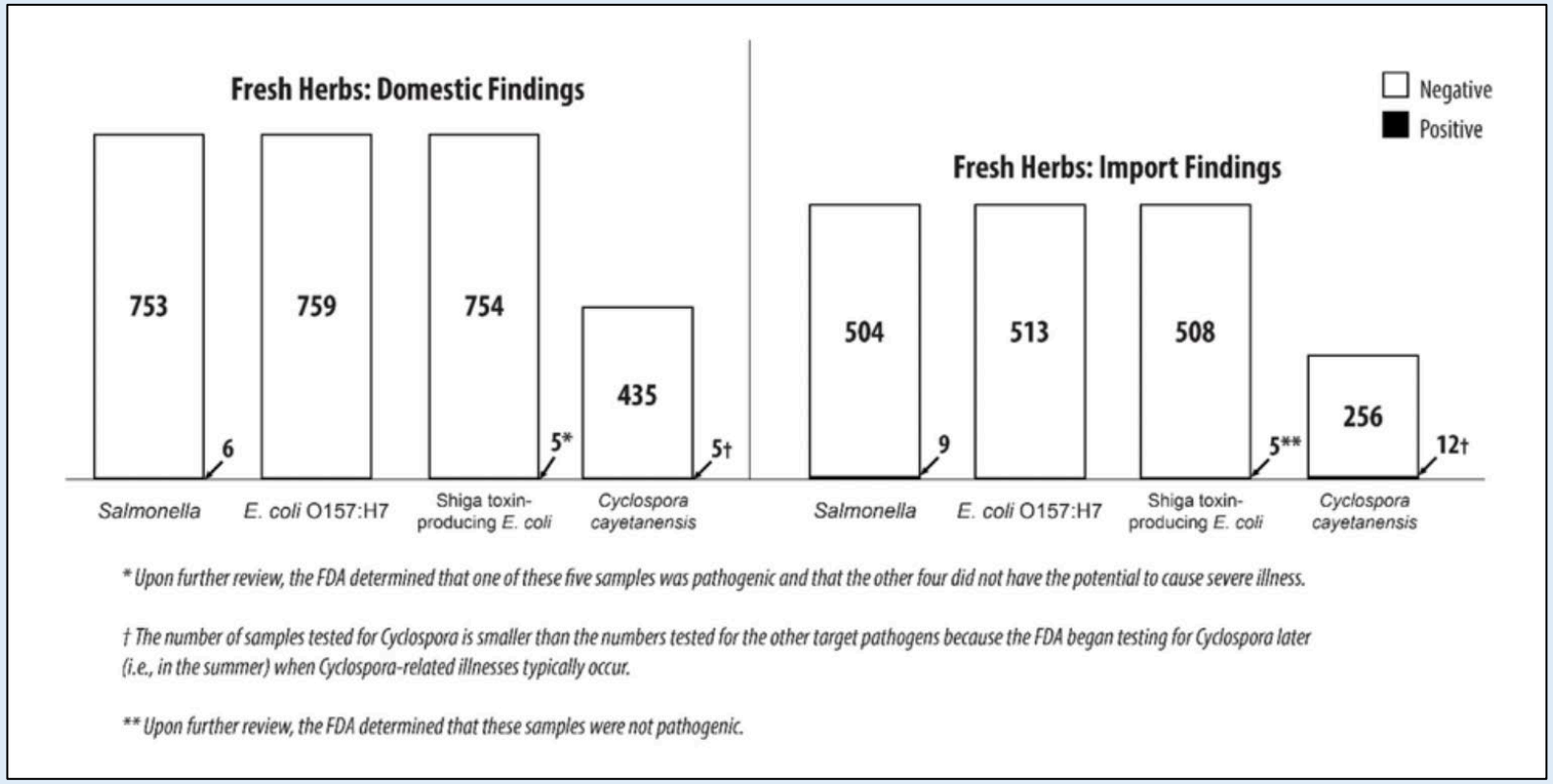
PCR Exponential Amplification

For Viruses, Preceded by Reverse Transcription
Step (RT), RNA ⟶ cDNA



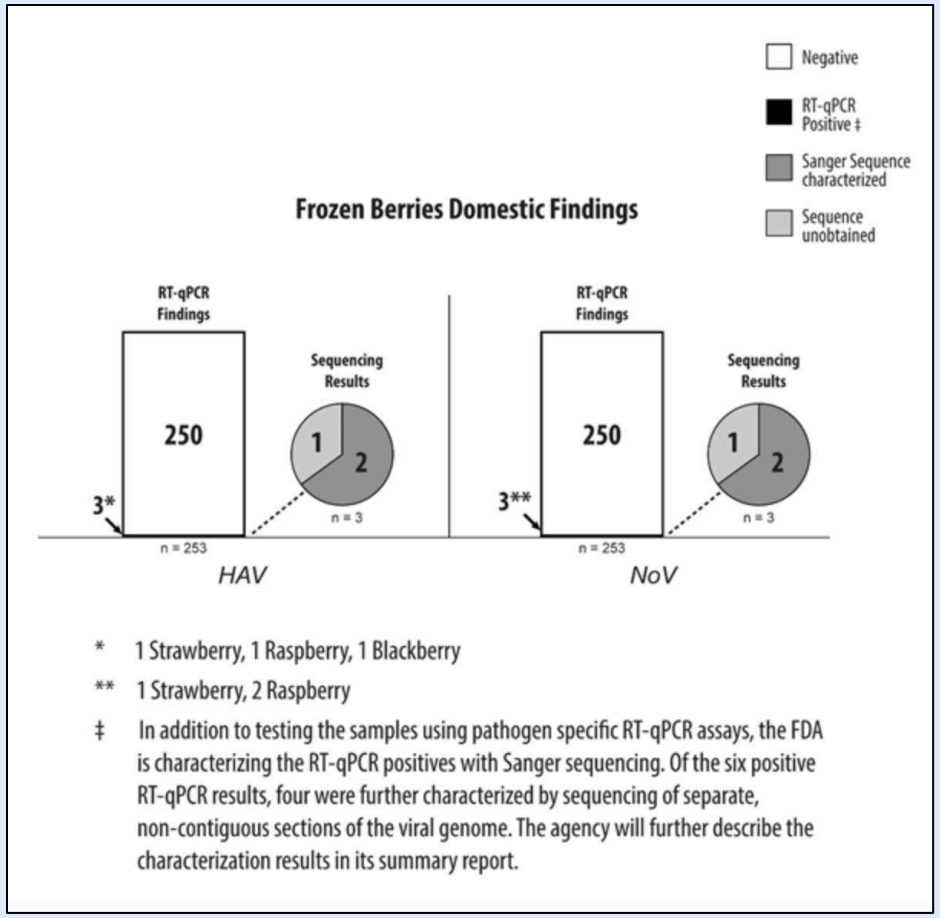
- Detects nucleic acid (DNA or RNA), not live virus or protozoa
- Nucleic acid can have be very stable, even after the microorganism is no longer infectious
- ***Detection of nucleic acid by RT-qPCR or PCR is not assurance that there is a "live" pathogen present—INFECTIVITY DILEMMA***
- ***We don't have a culture in-hand by which we can "prove" infectivity or do further characterization***
- There is a need to confirm (sequence)

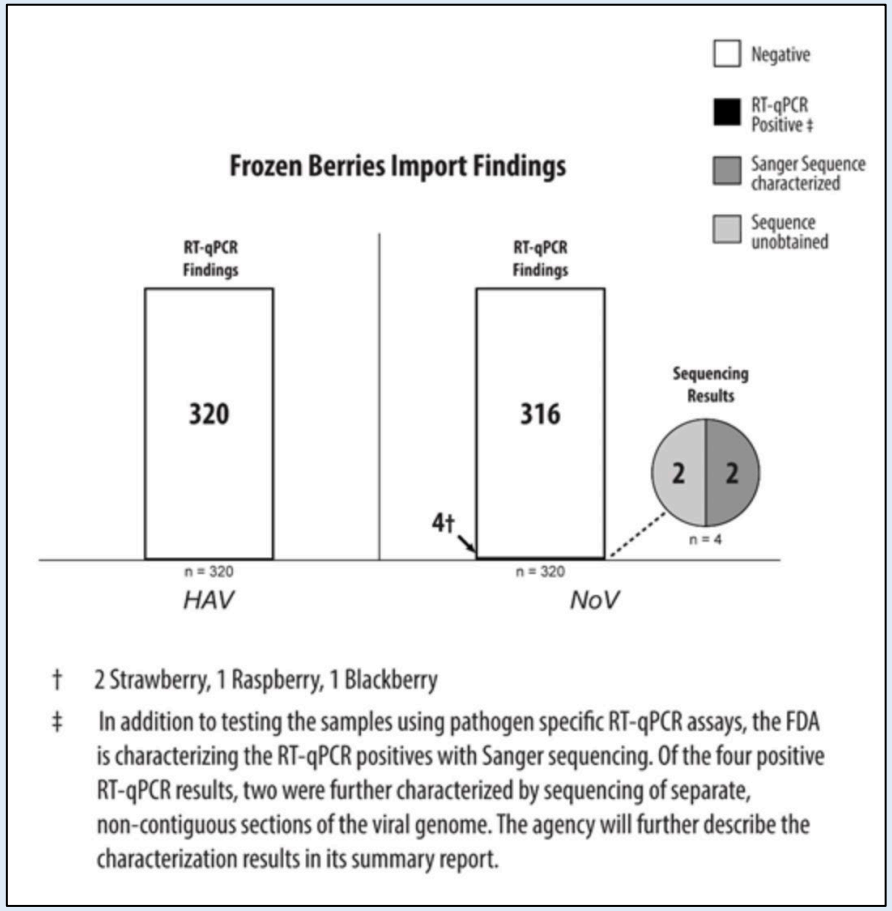
Microbiological Surveillance Sampling: FY18-21 Fresh Herbs (Cilantro, Basil & Parsley) and Processed Avocado and Guacamole Assignments



Available at: https://www.fda.gov/food/sampling-protect-food-supply/microbiological-surveillance-sampling-fy18-21-fresh-herbs-cilantro-basil-parsley-and-processed#Results

Microbiological Surveillance Sampling: FY 19-20 Frozen Berries (Strawberries, Raspberries and Blackberries)





Available at: https://www.fda.gov/food/sampling-protect-food-supply/microbiological-surveillance-sampling-fy-19-20-frozen-berries-strawberries-raspberries-and#sampling

Alma Pak Voluntarily Recalls Frozen Blackberries Due to Possible Health Risk of Norovirus

Kroger Recalls Select Frozen Private Selection Berries for Possible Health Risk

Shenandoah Growers, Inc Issues a Limited, Voluntary Recall of Specific Imported Organic Basil Because of Potential Health Risk

But there are are identifiable cases of illness

- What is the public health significance of a positive finding in the case of no associated illness?
- Interpretive criteria for RT-qPCR
  - Small proportion of the genome sequenced (<250 bp vs. 7.5 kb)
  - Interpretation of high Ct values (what constitutes a positive?)
  - Criteria for successful amplification, sequencing, sample positivity
  - Absence of a live culture upon which additional characterization can be done
- Expense to producers and processors
- Balance of economic vs. public health risks
- Surveillance studies for the pathogens will only be increasing

PCR Positivity Criteria used Internationally

- Move toward a Ct cut-off value
  - Ct<40 considered positive
  - Supported by clinical diagnostics (COVID-19, norovirus)
- Establishment of genome copy "tolerance"
  - Molluscan shellfish in Europe, in negotiation
  - 100-1,000 GEC (38-40, supported by clinical medicine, shellfish sanitation in UK/Europe
- Establish criteria for number of replicates positive and/or repeat testing (Eastern Europe)
- Risk-based (Canadian approach)

Use of PCR-Positive as an Indicator of the Potential for Product Adulteration

- Microbiological indicator: an organism, metabolite, or molecular signature whose presence and/or concentration in a sample is used as proxy for the potential presence of a pathogen or other harmful substance
- Examples
  - Shellfish sanitation (generic *E. coli*)
  - ATP bioluminescence to monitor sanitation
- Is the presence of a positive PCR signal for enteric viruses be an indicator of potential human fecal contamination?
  - Infectivity dilemma
  - Persistence of nucleic acids in the environment
  - Needs to be scientifically vetted

Example #2: Managing SARS CoV-2 Transmission in the Essential Workforce

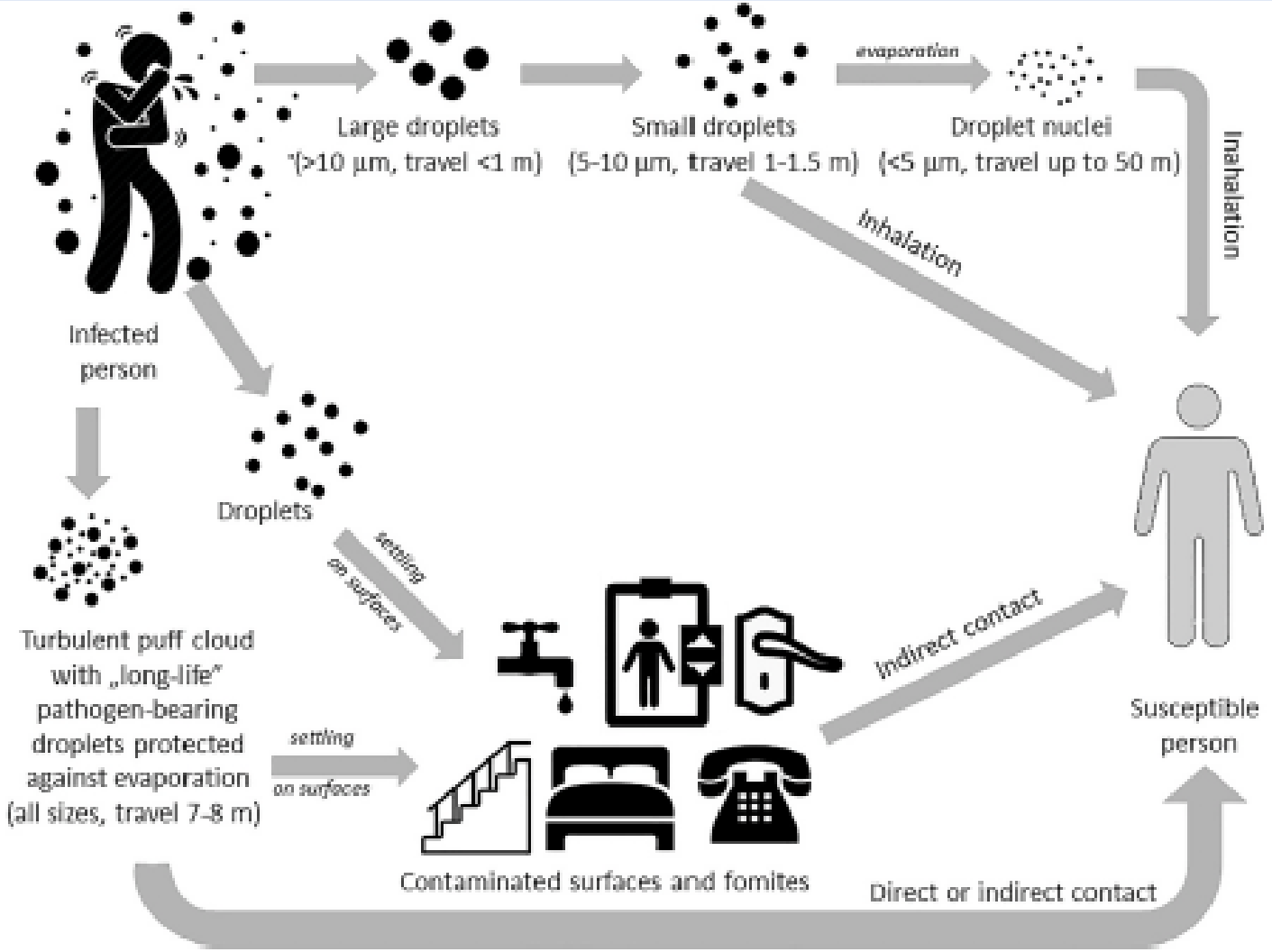
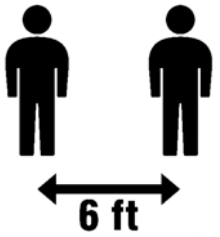


Fig. 5. Transmission routes involving direct contact, indirect contact by contaminated surfaces and fomites, as well as by droplets (short range) and droplet nuclei (long range) spreading [on the basis of (Tang et al., 2006)].

Understanding Transmission, the Food Industry is Applying Risk-Based Approaches to Protect Workers from SARS-CoV-2

- Temperature, Symptom Monitoring & Furlough
- Close Contact Documentation
- Universal Mask Usage
- Handwashing or Sanitizer Stations and Education
- Surface Disinfection: Increased frequency and rigor

- Physical Distancing Controls
  - Plexiglass Shields
  - Slower line speeds
  - Worker Shifts
  - No contact between Proc Workers and Visitors/Administrative Staff
  - Smaller Cohorts for breaks
  - Uni-directional traffic
  - Smaller cohorts for breaks



EMORY | ROLLINS SCHOOL OF PUBLIC HEALTH | NC STATE UNIVERSITY

Controlling risk of SARS-CoV-2 infection in essential workers of enclosed food manufacturing facilities

Julia S. Sobolik [a,*], Elizabeth T. Sajewski [a], Lee-Ann Jaykus [b], D. Kane Cooper [a], Ben A. Lopman [a], Alicia N.M. Kraay [a], P. Barry Ryan [a], Juan S. Leon [a]

[a] *Rollins School of Public Health, Emory University, Atlanta, GA, 30322, USA*
[b] *Food, Bioprocessing and Nutrition Sciences, North Carolina State University, Raleigh, NC, 27695, USA*

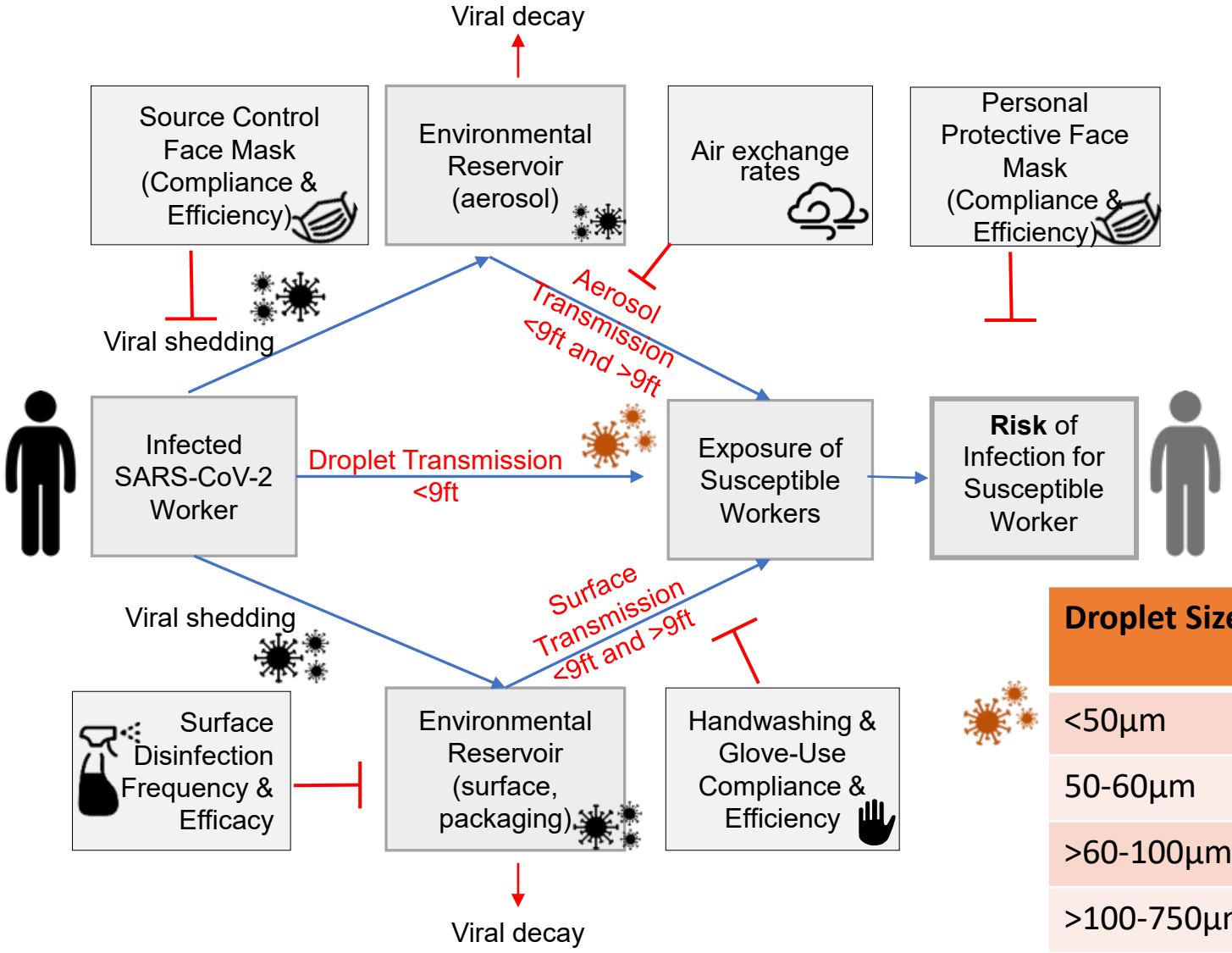
Among essential food workers in an enclosed facility,

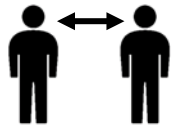
1. Which transmission pathway is most important?
   - Resiratory—Aerosol/Droplets
   - Fomites (Surfaces)
2. Are worker protection strategies enough?

Worker Protection Strategies

- Physical Distance
- Mask Use
- Air Exchange
- Hand-Hygiene
- Surface Disinfection

SARS-CoV-2 QMRA Exposure Asssessment for Respiratory Event (Coughing vs Breathing)





@ 3, 6, 9, >9ft from Infected Worker *(3, 6, 9, >9 ft)*

1-8h Exposure

Droplet Size	Distance Traveled	% of Fluid Expelled (Cough)
<50µm	>9ft (aerosol)	0.06%
50-60µm	0-9ft (close contact)	0.07%
>60-100µm	0-6ft (close contact)	0.12%
>100-750µm	<3ft (close contact)	99.75%

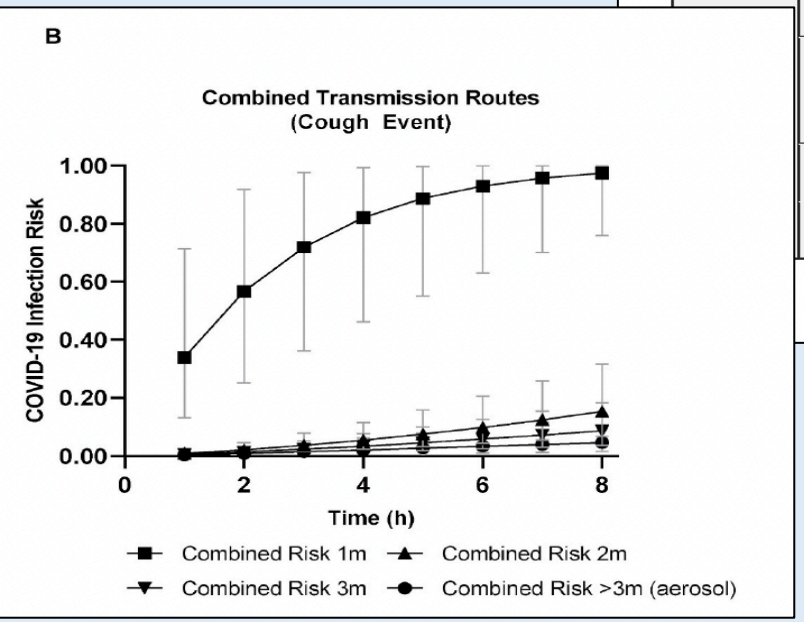
EMORY | ROLLINS SCHOOL OF PUBLIC HEALTH | NC STATE UNIVERSITY

Parameter	Units	Description	Input Values (a, b) *	Distribution
VIRUS				
$Log_{10}(C_{virus})$	PFU/mL	Concentration of virus in saliva	6.1 (5.2, 7.0)	Uniform
$d_{p,c}$	cm	Diameter of respiratory particles for coughing event	6E-4 (2E-4, 4.9E-3)	Triangular
$V_{F,c}$	mL/Cough	Fraction of volume associated with droplet diameters 100µm–750µm	6.8E-3 (4.0E-3, 7.6E-3)	Triangular
λ_{virus}	hr	Viral decay of SARS-CoV-2 per relative humidity environment	HH: (0.0466, 0.0911) LH: (0.0676, 0.1527)	Uniform
STRATEGY				
S_{mask}	Log reduction	Surgical mask efficacy	0.1549 (0.140, 0.222)	Triangular
C_{mask}	Log reduction	Cloth mask efficacy	(0.0458, 0.7100)	Uniform
SC_{eff}	Log reduction	Surface cleaner percent reduction in virus	(2.0, 6.0)	Uniform
HW_{freq}	Washes / hr	Frequency of handwashing per hour	1	Point
RISK				
F_{sa}	m^2	Surface area of fomite	(0.74, 2.20)	Uniform
F_{12}	Proportion	Proportion of virus transferred from fomite to hand per relative humidity	HH: 0.374 (0.160) LH: 0.069 (0, 0.158)	Normal Triangular
L_{dep}	Proportion	Deposition fraction of infectious virus into the lungs	0.210 (0.006, 0.320)	Triangular
k_{risk}	Unitless	Dose-response parameter	2.46E-3 (1.35E-3, 4.59E-3)	Triangular

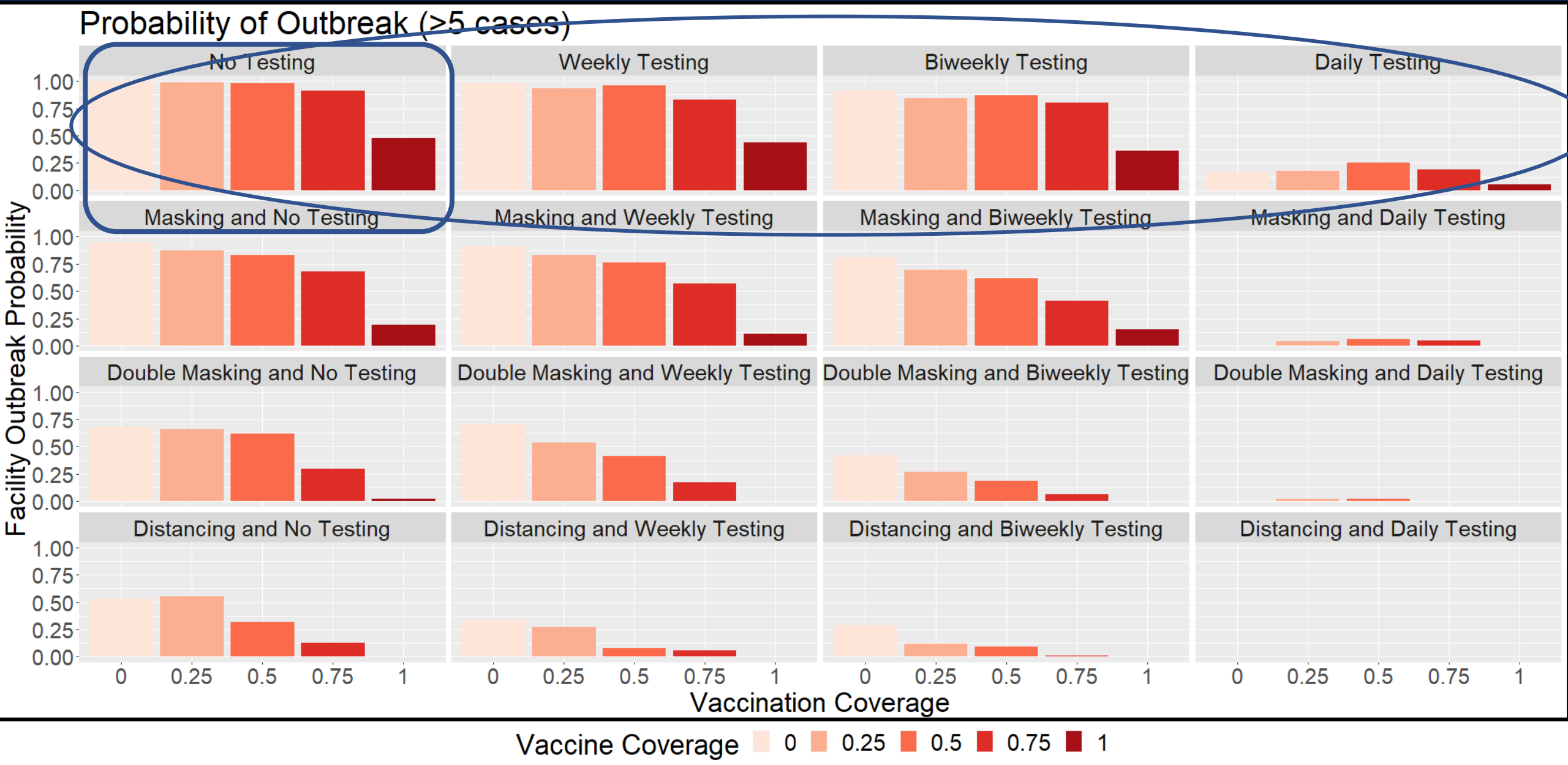
Table 3

Impact of bundled interventions (mask use, ventilation, hourly handwashing and surface disinfection twice per shift [4 h and 8 h]) on median infection risk (5th – 95th percentile) and percent risk reduction (%) following 8 h cumulative exposure to an infected worker (cough event as a function of distance). Colors indicate risk level from each bundled package. Dark purple indicates a high relative level of risk (>0.25–1.0), medium purple indicates a moderate relative level of risk (0.01–0.25), and light purple indicates a low relative level of risk (<0.01).

Relative level of risk:			**High (>0.25-1.0)**	**Moderate (0.01-0.25)**	**Low (<0.01)**
			1m	**2m**	**3m**	**Aerosols**
No ACH	No mask	**Risk** (5^{th} – 95^{th})	**0.98** (0.76–0.99)	**0.15** (0.07–0.32)	**0.09** (0.04–0.18)	**0.05** (0.01–0.13)
			---	---	---	---
2 ACH	Cloth mask	**Risk** (5^{th} – 95^{th})	**0.22** (0.06–0.61)	**0.005** (0.002–0.01)	**0.004** (0.001–0.01)	**0.002** (0.0005–0.009)
		% Reduction	77.6%	96.6%	95.9%	95.5%
	Surgical mask	**Risk** (5^{th} – 95^{th})	**0.14** (0.02–0.47)	**0.003** (0.0009–0.01)	**0.002** (0.0005–0.007)	**0.001** (0.0001–0.006)
		% Reduction	86.0%	97.8%	97.4%	97.1%
	Double mask	**Risk** (5^{th} – 95^{th})	**0.04** (0.005–0.30)	**0.001** (0.0002–0.005)	**0.0007** (0.0001–0.004)	**0.0003** (0.00003–0.003)
		% Reduction	96.2%	99.3%	99.2%	99.3%
6 ACH	Cloth mask	**Risk** (5^{th} – 95^{th})	**0.10** (0.02–0.33)	**0.002** (0.0007–0.006)	**0.001** (0.0005–0.004)	**0.001** (0.0002–0.003)
		% Reduction	89.8%	98.6%	98.3%	98.2%
	Surgical mask	**Risk** (5^{th} – 95^{th})	**0.06** (0.009–0.24)	**0.001** (0.0004–0.004)	**0.001** (0.0002–0.003)	**0.001** (0.00005–0.002)
		% Reduction	93.8%	99.1%	98.9%	98.8%
	Double mask	**Risk** (5^{th} – 95^{th})	**0.02** (0.002–0.14)	**0.0004** (0.00007–0.002)	**0.0003** (0.00004–0.002)	**0.0001** (0.00001–0.001)
		% Reduction	98.4%	99.7%	99.7%	99.7%



Outbreak probability is most reduced by daily testing or high vaccination coverage combined with masking or distancing.



Title: Low risk of SARS-CoV-2 transmission via fomite, even in cold-chain

Author names and affiliations: Julia S. Sobolik, MS[a1], Elizabeth T. Sajewski, MS[a], Lee-Ann Jaykus, PhD[b], D. Kane Cooper, MSPH[a], Ben A. Lopman, PhD[a], Alicia NM. Kraay, PhD[a], P. Barry Ryan, PhD[a], Jodie L. Guest, PhD[a], Amy Webb-Girard, PhD[a], Juan S. Leon, PhD[a]

[a]Rollins School of Public Health, Emory University, Atlanta, GA, USA, 30322

[b]Food, Bioprocessing and Nutrition Sciences, North Carolina State University, Raleigh, NC, USA, 27695

Findings: In a representative facility with no specific interventions, SARS-CoV-2 infection risk to a susceptible worker from contact with contaminated packaging was $2{\cdot}8 \times 10^{-3}$ per 1h-period (95%CI: $6{\cdot}9 \times 10^{-6}$, $2{\cdot}4 \times 10^{-2}$). Implementation of standard infection control measures, handwashing and masks ($9{\cdot}4 \times 10^{-6}$ risk per 1h-period, 95%CI: $2{\cdot}3 \times 10^{-8}$, $8{\cdot}1 \times 10^{-5}$), substantially reduced risk (99·7%). Vaccination of the susceptible worker (two doses Pfizer/Moderna, vaccine effectiveness: 86-99%) combined with handwashing and masking reduced risk to less than $1{\cdot}0 \times 10^{-6}$. Simulating increased infectiousness/transmissibility of new variants (2-, 10-fold viral shedding) among a fully vaccinated workforce, handwashing and masks continued to mitigate risk ($2{\cdot}0 \times 10^{-6}$ -$1{\cdot}1 \times 10^{-5}$ risk per 1h-period). Decontamination of packaging in addition to these interventions reduced infection risks to below the $1{\cdot}0 \times 10^{-6}$ risk threshold.

Interpretation: Fomite-mediated SARS-CoV-2 infection risks were very low under cold-chain conditions. Handwashing and masking provide significant protection to workers, especially when paired with vaccination.

medRxiv preprint doi: https://doi.org/10.1101/2021.08.23.21262477; this version posted August 26, 2021. The copyright holder for this preprint **(which was not certified by peer review)** is the author/funder, who has granted medRxiv a license to display the preprint in perpetuity.
It is made available under a CC-BY-NC-ND 4.0 International license .

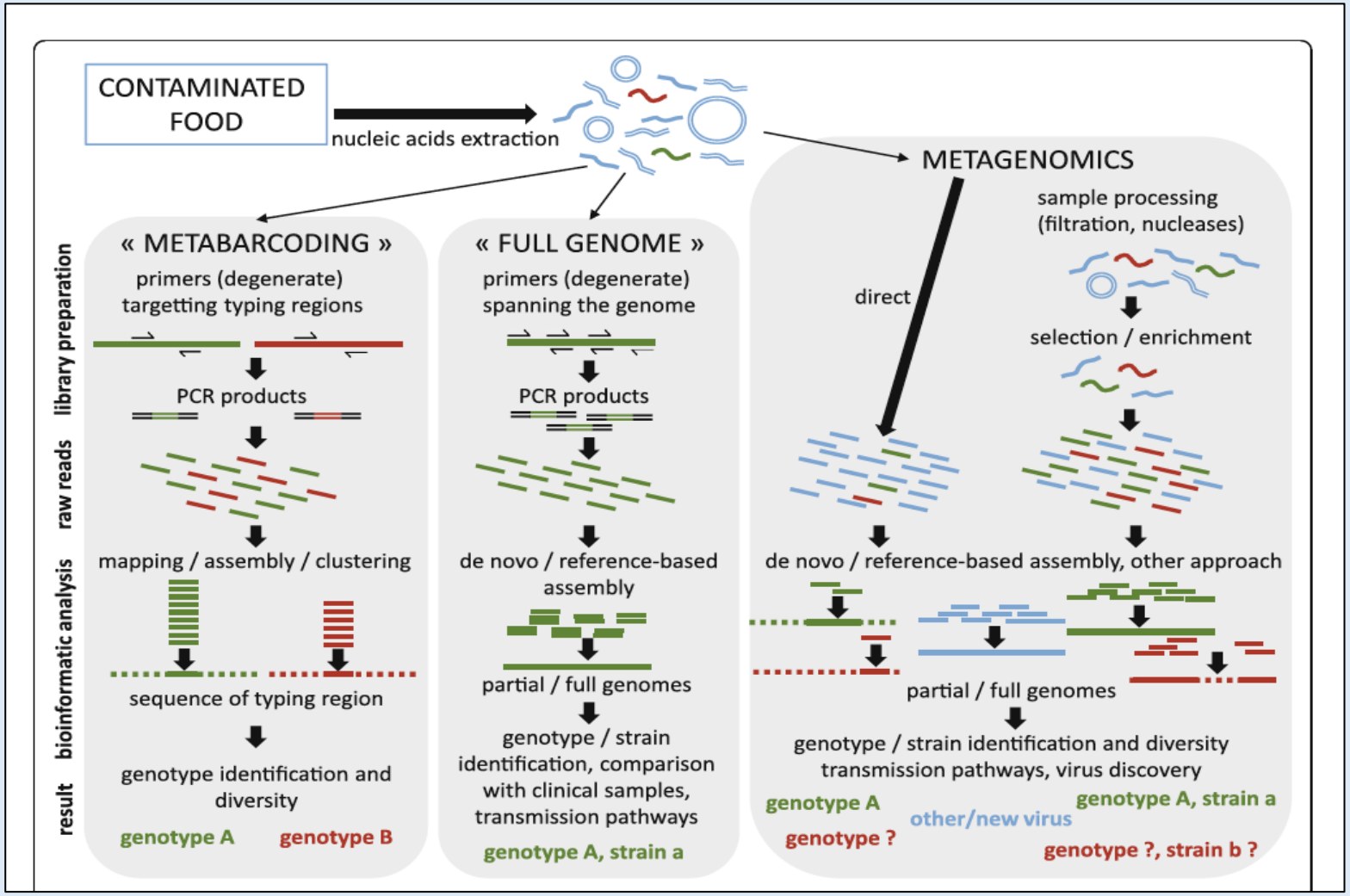
Example #3: Next Generation Sequencing, Metagenomics, and Bioinformatics

Novel opportunities for NGS-based one health surveillance of foodborne viruses

Marion Desdouits[1], Miranda de Graaf[2], Sofia Strubbia[1], Bas B. Oude Munnink[2], Annelies Kroneman[3], Françoise S. Le Guyader[1] and Marion P. G. Koopmans[2*]

Desdouits *et al. One Health Outlook* (2020) 2:14
https://doi.org/10.1186/s42522-020-00015-6

One Health Outlook



January 24, 2020

Petition Asks FSIS to Declare 31 Salmonella Strains as Adulterants

Marler Clark LLP filed a petition on behalf of several individuals and consumer groups on January 19, asking USDA's Food Safety and Inspection Service (FSIS) to issue an interpretive rule declaring 31 salmonella strains as per se adulterants in meat and poultry products. These strains, which the petition refers to as the "*Salmonella* Outbreak Serotypes," include four antibiotic-resistant strains—*Salmonella hadar*, *heidelberg*, *newport*, and *typhimurium*—as well as *Salmonella dublin*, *enteritidis*, and *infantis*. The petition seeks expedited review of its request on the grounds that these strains have been shown to be linked to foodborne illness outbreaks and/or product recalls and, as such, constitute an imminent threat to public health.

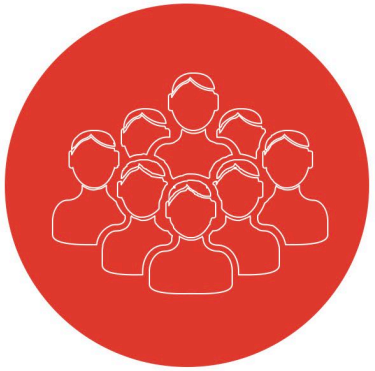
According to the petition, declaring these 31 strains as adulterants would promote the goals of the Federal Meat Inspection Act (FMIA) and the Poultry Products Inspection Act to protect public health by encouraging the meat and poultry industry to implement more effective safeguards and oversight measures. The petition relies heavily on its interpretation of precedent established following the 1993 *Escherichia coli*, when USDA declared *Escherichia coli O157:H7* a per se adulterant in raw ground beef through interpretive rulemaking.

https://www.jdsupra.com/legalnews/petition-asks-fsis-to-declare-31-35558/

CONFIDENTIAL INTERNAL USE ONLY - NOT FOR REDISTRIBUTION

Example #4: Crowd Sourcing

1. iwapoisoned.com Data

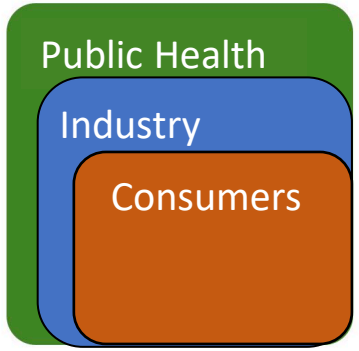


Online reporting platform allows consumers to easily search, find the site, and file reports

2. Manage Data

Data analytics team curates and moderates the data and extracts usable intelligence.

3. Provide Services



Early warning signals are provided to subscribers, including the public health community, industry and consumers

CONFIDENTIAL INTERNAL USE ONLY - NOT FOR REDISTRIBUTION

Sample Report

Local Restaurant a New Jersey Town

Reported: Jan 14th 2019
Symptoms: Diarrhea, Nausea

I ate the meal on Sunday January 13th, at around 12:00. At 1:00am, approximately 11 hours subsequent, I had violent diharreah and stomach pain. This is the point I'm at rn, writing this.

I ate:
-lobster bisque
-house salad with iceberg lettuce and the champagne vinegarette
-the platter with lobster, crab legs, and two types of shrimp. I barely ate shrimp or lobster though (besides in the bisque), mostly just the crab, so that is my suspect

I just had tap water to drink, and haven't eaten anything else all day.

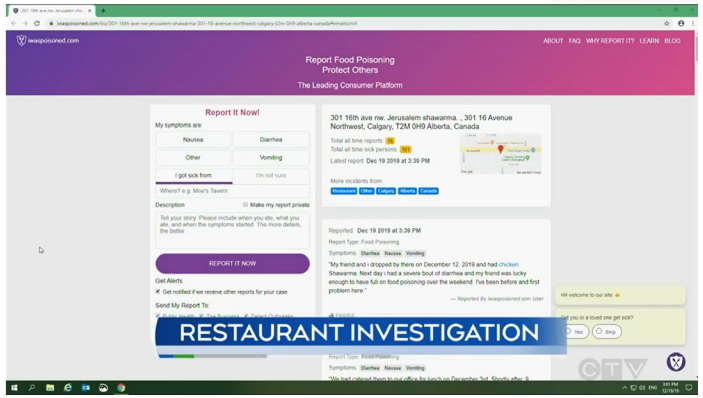
Outbreak Case Study

Calgary, CA December 2019

We first began receiving reports relating to ********** in Calgary on Dec 5th, 2019. By Dec 19th, we had received reports of over 100 sick, and local news coverage commenced. By the 23rd: 123 sickened, and Alberta Health Services confirmed a Norovirus outbreak linked to the local chain. We found out about this over 3 weeks in advance. This is the type of outcome we specialize in preventing.

CTV NEWS

Click here to watch the news story



Conclusions

- As scientific and analytical methods become more sophisticated and sensitive, there will be downstream management and regulatory actions that have policy and legal implications
- For example:
  - Identification of outbreaks in near real-time
  - Identification of new or emerging pathogens (e.g., STEC, *Cyclospora cayetanensis*)
  - ***Detection advances-"chasing zero" and finding "needles in haystacks"***
  - Regulatory actions in food production (e.g., FSMA Water Rule in fresh produce)
- It is important to carefully consider the strengths and weaknesses of these techniques to prevent unintended consequences that can manifest as over-protection of public health and have financial implications, among other consequences