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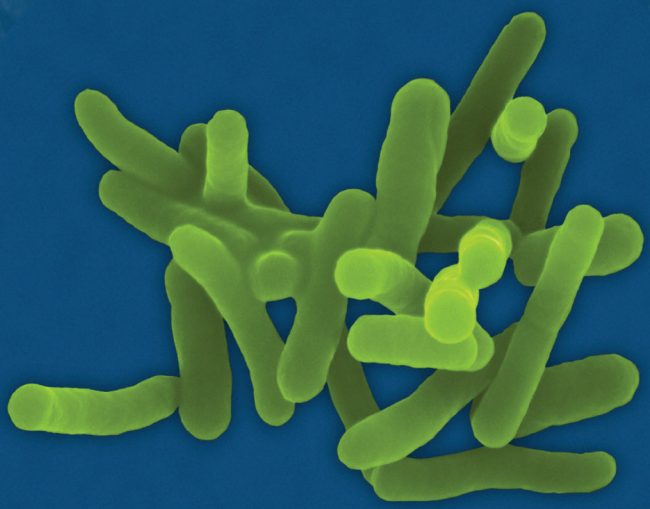
World Health Organization

Attributing illness caused by Shiga toxin-producing *Escherichia coli* (STEC) to specific foods

REPORT



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MICROBIOLOGICAL RISK ASSESSMENT SERIES

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Contents

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Contributors

Dr Shannon E. Majowicz, Associate Professor, School of Public Health and Health Systems, University of Waterloo, Canada

Dr Sara Monteiro Pires, Senior Researcher, Risk Benefit Research Group Division of Diet, Disease Prevention and Toxicology, National Food Institute, Technical University of Denmark, Denmark

Dr Brecht Devleesschauwer, Department of Epidemiology and Public Health, Sciensano, Brussels, Belgium

RESOURCE PERSONS

Dr Hiroshi Asakura, Director, Division of Biomedical Food Research, National Institute of Health Sciences, Japan

Dr Patricia Desmarchelier, Director, Food Safety Principles, Australia

Dr Roger Cook, Manager, Food Risk Assessment and Principal Microbiologist, Ministry for Primary Industries, New Zealand

Dr Eelco Franz, National Institute for Public Health and the Environment (RIVM), Centre for Infectious Disease Control, The Netherlands

Dr Pina Fratamico, Agricultural Research Service, Eastern Regional Research Center (ERRC), United States Department of Agriculture, United States of America

Dr Alex Gill, Principal Investigator, Verotoxigenic *Escherichia coli* Laboratory, Bureau of Microbial Hazards, Health Canada, Canada

Dr Patricia Griffin, Chief, Enteric Diseases Epidemiology Branch, Division of Foodborne, Waterborne, and Environmental Diseases, National Center for Emerging and Zoonotic Infectious Diseases, Centers for Disease Control and Prevention, United States of America

Dr Weidong Gu, Mathematical Statistician, Enteric Diseases Epidemiology Branch, Division of Foodborne, Waterborne, and Environmental Diseases, National Center for Emerging and Zoonotic Infectious Diseases, Centers for Disease Control and Prevention, United States of America

Dr Roberto Vidal Alvarez, Professor, Head Laboratory of Pathogenic Escherichia coli (LPE), Institute of Biomedical Sciences, Faculty of Medicine, Universidad de Chile, Santiago de Chile

JEMRA SECRETARIAT

Sarah Cahill, Food Safety Officer, Food Safety and Quality Unit, Agriculture and Consumer Protection Department, Food and Agriculture Organization of the United Nations, Italy

Jeffrey LeJeune, Food Safety Officer, Food Safety and Quality Unit, Agriculture and Consumer Protection Department, Food and Agriculture Organization of the United Nations, Italy

Blaise Ouattara, Food Safety and Quality Officer, Food Safety and Quality Unit, Agriculture and Consumer Protection Department, Food and Agriculture Organization of the United Nations, Italy

Kang Zhou, Food Safety and Quality Officer, Food Safety and Quality Unit, Agriculture and Consumer Protection Department, Food and Agriculture Organization of the United Nations, Italy

Rei Nakagawa, Technical Officer, Department of Food Safety and Zoonoses, World Health Organization, Switzerland

Satoko Murakami, Technical Officer, Department of Food Safety and Zoonoses, World Health Organization, Switzerland

Declarations of interest

All participants completed a Declaration of Interests form in advance of the meeting, based on the information provided.

All the declarations, together with any updates, were made known and available to all the participants at the beginning of the meeting. All the experts participated in their individual capacities and not as representatives of their country, government or organization.

Abbreviations

AFR	African Region [WHO classification]
AMR	Region of the Americas [WHO classification]
DALY	Disability-Adjusted Life Year
EMR	Eastern Mediterranean Region [WHO classification]
EUR	European Region [WHO classification]
FAO	The Food and Agriculture Organization of the United Nations
FERG	Foodborne Disease Burden Epidemiology Reference Group
HUS	Haemolytic uraemic syndrome
IFSAC	Interagency Food Safety Analytics Collaboration
OR	odds ratio
PECOS	Population Exposure Comparator Outcome Study Design
ROBINS-I	Risk Of Bias In Non-randomised Studies - of Interventions
SE	standard error
SEAR	South-East Asian Region [WHO classification]
STEC	Shiga toxin-producing *Escherichia coli*
UI	uncertainty interval
VTEC	Verocytotoxigenic *Escherichia coli*
WHO	World Health Organization
WPR	Western Pacific Region [WHO classification]

Executive summary

Shiga toxin-producing *Escherichia coli* (STEC) infections are a substantial public health issue worldwide, causing more than 1 million illnesses, 128 deaths and nearly 13 000 Disability-Adjusted Life Years (DALYs) annually. To appropriately target interventions to prevent STEC infections transmitted through food, it is important to determine the specific types of foods leading to these illnesses. An analysis of data from STEC foodborne outbreak investigations reported globally, and a systematic review and meta-analysis of case-control studies of sporadic STEC infections published for all dates and locations, were conducted.

A total of 957 STEC outbreaks from 27 different countries were included in the analysis. Overall, outbreak data identified that 16% (95% UI, 2-17%) of outbreaks were attributed to beef, 15% (95% UI, 2-15%) to produce (fruits and vegetables) and 6% (95% UI, 1-6%) to dairy products. The food sources involved in 57% of all outbreaks could not be identified. The attribution proportions were calculated by WHO region and the attribution of specific food commodities varied between geographic regions. In the European and American sub-regions of the WHO, the primary sources of outbreaks were beef and produce (fruits and vegetables). In contrast, produce (fruits and vegetables) and dairy were identified as the primary sources of STEC outbreaks in the WHO Western Pacific sub-region.

The systematic search of the literature identified useable data from 21 publications of case-control studies of sporadic STEC infections. The results of the meta-analysis identified, overall, beef and meat-unspecified as significant risk factors for STEC infection. Geographic region contributed to significant sources of heterogeneity.

Generally, empirical data were particularly sparse for certain regions. Care must be taken in extrapolating data from these regions to other regions for which there are no data. Nevertheless, results from both approaches are complementary, and support the conclusion of beef products being an important source of STEC infections. Prioritizing interventions for control on beef supply chains may provide the largest return on investment when implementing strategies for STEC control.

1

Introduction

Shiga toxin-producing *Escherichia coli* (STEC) infections are a significant public health issue worldwide. Circa 2010, the Foodborne Disease Burden Epidemiology Reference Group (FERG) of the World Health Organization (WHO) estimated that every year foodborne STEC (i.e. STEC infections transmitted via food, as opposed to water, person-to-person contact, or other routes of transmission) caused more than 1 million illnesses (95% uncertainty interval [UI]: 754 000 to 2.5 million), 128 deaths (95% UI: 55 to 374) and nearly 13 000 Disability Adjusted Life Years (DALYs; 95% UI: 5951 to 33 664; Kirk *et al*., 2015). To appropriately target interventions to prevent these foodborne infections, it is important to determine the specific food types leading to these illnesses. Source attribution is a methodology for identification of the food types that are important sources of exposure, which allows such estimates to be generated.

2

Source attribution of STEC illnesses

2.1 OVERVIEW OF SOURCE ATTRIBUTION CONCEPTS

Human foodborne illness *source attribution* is defined as attribution of the human disease burden of one or more foodborne illnesses to specific sources, where the term *source* can include reservoirs or vehicles. To this end, source attribution methods are used to analyse data from food/animal monitoring and/or public health registries to estimate the relative contribution of different sources to disease.

A variety of approaches for attributing foodborne diseases to specific sources are available, including hazard occurrence analysis (sub-typing and comparative exposure assessment methods); epidemiological methods (analysis of data from outbreak investigations and studies of sporadic infections); intervention studies; and expert elicitation (Pires *et al*., 2009). Each of these methods has advantages and limitations, and the usefulness of each depends on the questions being addressed and on the characteristics and distribution of the hazard. The choice of the method to be used should be guided by these factors. As well, source attribution can take place at different points along the food chain (points of attribution) – most often at the point of reservoir (e.g. animal production stage, environmental emissions) or point of exposure (i.e. end of the transmission chain). The point of attribution depends on the method chosen, which will depend on the risk management question being addressed and on the availability of data.

2.2 SUMMARY OF THE FINDINGS FROM THE FERG EXPERT ELICITATION

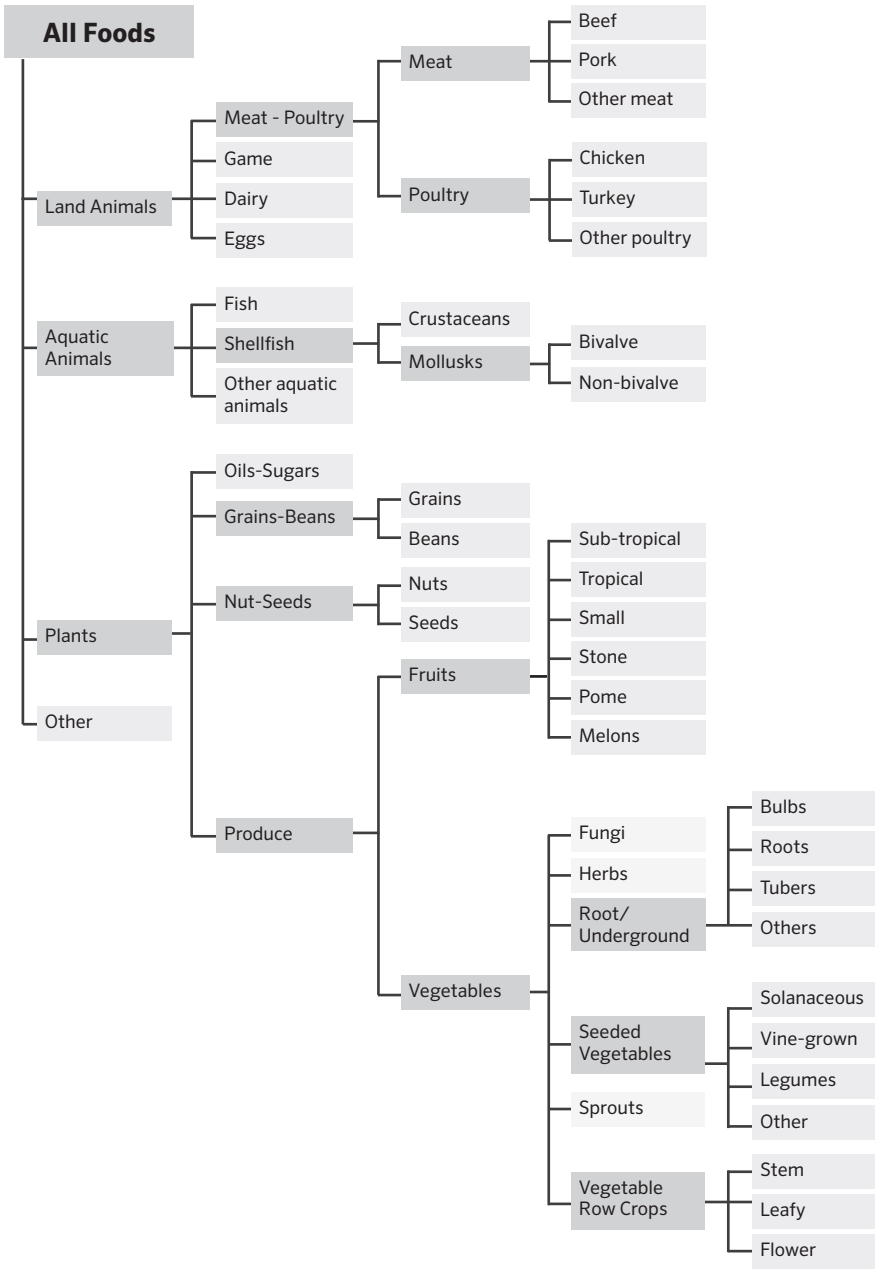
FERG estimated the proportion of foodborne disease burden of STEC that is attributable to specific foods (Hoffmann *et al*., 2017) as follows. In the absence of data-based evidence at regional or global level, FERG relied on expert elicitation to estimate the proportion of the foodborne disease burden of STEC due to specific foods (Hald *et al*., 2016; Havelaar *et al*., 2015; Hoffmann *et al*., 2017). Expert elicitations are particularly useful to attribute human illness to the main routes of transmission – i.e. foodborne, environmental, and direct contact with humans or animals. Another advantage of expert elicitation is that it enables the views of experts in all regions of the world to be used in making regional attribution estimates.

FERG's expert elicitation applied Cooke's classical model for structured expert elicitation, to provide a consistent set of estimates. The global expert elicitation study involved 73 experts and 11 elicitors, and was one of the largest, if not the largest, study of this kind ever undertaken (Hald *et al*., 2016; Hoffmann *et al*., 2017). Due to the study constraints (e.g. remote elicitation rather than face-to-face meetings), accuracies of individual experts – elicited based on calibration questions – were generally lower than in other structured expert judgment studies. However, performance-based weighting, a key characteristic of Cooke's classical model, increased informative usefulness, while retaining accuracy at acceptable levels (Aspinall *et al*., 2016).

The expert elicitation attributed the foodborne STEC burden to six food categories and "other foods"; the proportion of disease attributable to unknown categories was not estimated. Beef was estimated to be the major food source in most regions (~50%), except in the South-East Asian sub-region, where small ruminant meat was estimated to be the major source (~25%). In the medium-mortality Western Pacific sub-region (WPR B), beef and small ruminant meat were attributed equal contributions to disease burden (~25% each).

2.3 SOURCE ATTRIBUTION APPROACH USED IN THIS REPORT

The source attribution approach taken here was designed to supplement FERG's expert elicitation attribution estimates with data-driven attribution estimates.



NOTES: Food categories not shown can be included by further detailing the scheme.

FIGURE 1. Food categorization scheme, Interagency Food Safety Analytics Collaboration (IFSAC).

To produce data-driven source attribution estimates at the global and regional level, two methods were applied to attribute regional and global burden of STEC infections to specific foods:

- An analysis of data from outbreak investigations; and
- A systematic review of case-control studies of sporadic infections.

In consistency with the work of FERG, source attribution of the STEC disease burden was performed at the point of exposure.

Both of these methods used a harmonized source categorization scheme, which is necessary to compare and integrate results from various data sources, source attribution models, approaches or hazards. Specifically, the food categorization scheme produced by the United States' Interagency Food Safety Analytics Collaboration (IFSAC) was adopted (Figure 1).

3

An analysis of data from outbreak investigations

3.1 BACKGROUND AND OBJECTIVE

Epidemiological approaches for source attribution use public health surveillance data to estimate the relative contribution of different sources, routes of exposure or risk factors for disease (Pires *et al*., 2009). These include analyses of data from outbreak investigations, which have been used for source attribution of several pathogens at national or regional level (Greig and Ravel, 2009; Painter *et al*., 2013; Pires *et al*., 2012; Pires *et al*., 2010). A simple summarization of results of outbreak investigations can be useful for identifying the most common foods causing human illness by a pathogen. However, often the implicated food is a "complex" food – i.e. containing multiple food items and ingredients, where in principle any of them could be the specific source of the outbreak (Painter *et al*., 2013). The objective of this study was to estimate the relative contribution of different foods to STEC infections in WHO regions and globally, using data from outbreak investigations. We applied a method that is able to consider implicated complex foods to attribute human STEC infections to specific sources.

3.2 METHODS

3.2.1 Data

A call[1] for STEC outbreak surveillance data was sent by WHO to national Codex contact points and through other relevant channels to Member States in April

[1] http://www.fao.org/3/a-br569e.pdf

2016. The request aimed at collecting data on all STEC outbreaks reported globally and contained no restriction on time period. Collected data were harmonized and organized so that each reported outbreak corresponded to one observation in the final dataset. Each observation contained information on the year of occurrence, country, etiology, number of ill people and fatalities associated with the outbreak, location of the outbreak, and implicated source. For uncompleted fields, the parameter was included as *unknown.*

3.2.2 Food categorization

We applied the food categorization scheme produced by IFSAC (see Figure 1), allowing for potential adaptations to accommodate sub-categories that are common in different countries or regions. The level of sub-categorization within each main food category varied for different fields. For example, under "land animals", the lowest level of sub-categorization was used, while all fruits and all vegetables were grouped in the higher level categories, i.e. "fruits" and "vegetables", respectively.

3.2.3 Model overview

The method was based on the work of Pires *et al.* (2010), modified and applied to the STEC dataset. The principle is to attribute human illnesses to food sources on the basis of the number of outbreaks that were caused by each of these foods. For this purpose, implicated foods are classified by their ingredients as simple foods (i.e. belonging to one single food category), or complex foods (i.e. belonging to multiple food categories). The ingredients that constitute the complex foods are designated through defined criteria (Painter *et al.*, 2009). The proportion of disease that can be attributed to each food source was estimated in a two-step process based on: a) the number of simple-food outbreaks caused by that source; and b) the number of complex-food outbreaks, the ingredients (food categories) composing complex foods, and the probability that each of these categories was the cause of the complex-food outbreaks. The attributable proportions were calculated by WHO region (AFR: African Region; AMR: Region of the Americas; EUR: European Region; EMR: Eastern Mediterranean Region; SEAR: South-East Asia Region; WPR: Western Pacific Region).

In the first step, the number of simple-food outbreaks attributed to each single food category was calculated by WHO region. In the second step, we first calculated the probability P_j that an outbreak was caused by source j, by summarizing the data from simple-food outbreaks per source across all countries and across the whole study period. Specifically, P_j was defined as the proportion of single-food outbreaks caused by source j. The uncertainty in the probability vector P was quantified using a Dirichlet(S) distribution, with S the vector of components S_j corresponding to the number of single-food outbreaks caused by source j. Next,

complex-food outbreaks were partitioned according to each of the food categories in the implicated food proportionally to the probability P_j of causing a simple-food outbreak. We used a Monte Carlo simulation approach to propagate the uncertainty in P_j and the uncertain allocation of a complex-food outbreak to a specific food category. First, we simulated 10 000 values of P_j for each source *j*. Then we multiplied P_j with a dummy matrix F_{ij}, representing the implicated food categories *j* in outbreak *i*. As an example, outbreaks caused by chili con carne would be attributed to the categories 'beef', 'vegetables', 'grains and beans', and 'oils and sugar'; F_{ij} would thus contain the value 1 for sources 'beef', 'vegetables', 'grains and beans', and 'oils and sugar', and value 0 for other sources. By multiplying with P_j, outbreaks due to a complex food were only attributed to categories that had been implicated in at least one simple-food outbreak. In our example above, if 'grains and beans' and 'oils and sugars' were not implicated in any pathogen-specific outbreak caused by simple foods, these two categories would be excluded for the attribution of the chili con carne outbreak. In the second step of the Monte Carlo process, we accounted for the uncertain allocation of complex-food outbreaks to specific food categories. For each complex-food outbreak and per iteration of P_j, we simulated 20 realizations of a multinomial distribution with size 1 and probability vector $P_j F_j$. For each complex-food outbreak *i*, this then resulted in 200 000 random attributions to a single source *j*.

Finally, the results of the simple-food outbreaks were summed with the probabilistic attributions of the complex-food outbreaks to obtain the total number of outbreaks, by region, attributed to each source *j*. The proportion of disease attributed to each source, again by region, was then obtained by dividing the total number of attributed outbreaks to the total number of reported outbreaks.

The proportion of disease attributable to specific sources was estimated on the basis of the number of reported outbreaks. The number of ill people implicated in the outbreaks was not considered in the analysis to avoid potential overestimation of the importance of sources that caused large outbreaks, e.g. foods with larger distribution chains. To estimate the relative importance of the food sources implicated in cases of Haemolytic uraemic syndrome (HUS), we applied the same modelling approach to attribute the outbreaks that included HUS cases to food sources. In addition, to estimate relative importance of the food sources for severe cases of disease, we applied the same model to outbreaks associated with fatalities.

3.3 RESULTS

3.3.1 Data used in the model

STEC outbreak surveillance data were received from 27 countries covering the period between 1998 and 2017 and spanning three WHO geographic regions: AMR, EUR and WPR (Annex 1). The oldest data were reported by the United States between 1998 and 2015; the remaining countries reported data corresponding to outbreaks that occurred between 2010 and 2017.

In total, the data set included 957 STEC outbreaks, the large majority (78%: 746/957) reported in the AMR. Of the 957 outbreaks, 345 (36%) were caused by a simple food, 80 (8%) by a complex food, and 532 (56%) were caused by an unknown source (Table 1).

TABLE 1. Number and proportion of outbreaks caused by simple, complex or unknown foods in WHO regions*

	Simple		Complex		Unknown		
Region	**Number**	**%**	**Number**	**%**	**Number**	**%**	**Total**
AMR	283	38	61	8	402	54	746
EUR	55	31	14	8	107	61	176
WPR	7	20	4	11	24	69	35
Total	**345**	**36**	**80**	**8**	**532**	**56**	**957**
Outbreaks associated with HUS cases							
AMR	92	53	15	7	121	40	228
EUR	1	100	0	0	0	0	1
WPR	3	43	0	0	4	57	7
Total	**96**		**15**		**125**		**236**
Outbreaks associated with deaths							
AMR	21	50	1	2	20	48	42
EUR	2	100	0	0	0	0	2
WPR	1	100	0	0	0	0	1
Total	**24**		**1**		**20**		**45**

*AMR: Region of the Americas; EUR: European Region; WPR: Western Pacific Region.

A total of 236 outbreaks that involved HUS cases were reported during the time period analyzed, nearly all (97%) in the AMR. Of these outbreaks reported in the AMR, 53% were caused by simple foods, 7% by complex foods and 40% by

an unknown source (Table 1). Twenty-nine percent (281/957) of all reported outbreaks were associated with either HUS or deaths.

Most of the 45 outbreaks that involved fatalities were also reported in the AMR, the large majority of them being caused by simple foods (50%) or unknown sources (48%) (Table 1).

3.3.2 Attribution to foods

The results of the overall analysis, including all countries and the entire time period, showed that the most frequently attributed sources of STEC globally were beef, with an attribution proportion of 16% (95% UI: 2-17%); produce (fruits and vegetables), at 15% (95% UI: 2-15%); and dairy products, at 6% (95% UI: 1-6%). More than half of the outbreaks globally (57%) could not be attributed to any source. These estimates are downward biased, as they do not include outbreaks attributable to complex or unknown food sources that may have contained one or more of these food commodities.

WHO regions differed in the relative contributions of different sources of STEC (Table 2, Figure 2). When outbreaks attributed to unknown source were excluded, beef and produce (fruits and vegetables) were responsible for the highest proportion of cases in the AMR, with source attribution estimates of 40% for beef (95% UI: 39.1-40.9%) and 35% for produce (fruits and vegetables) (95% UI: 34.1-36.2%) (Figure 2); all following estimates disregard outbreaks attributed to unknown source. Twelve percent (95% UI: 11.5-12.9%) of STEC cases in AMR could be attributed to dairy products. In the EUR, the ranking of the sources of cases was similar, though with less marked differences among sources, with an overall attribution proportion of 31% (95% UI 28.4-34.3%) for beef; 30% (95% UI 26.9-32.8%) for produce (fruits and vegetables); and 16.4% for dairy (Figure 2).

In contrast, the most common source of STEC in WPR was produce (fruits and vegetables) (43%; 95% UI 36.4-45.5%), followed by dairy (27%) and with game and beef third and fourth (9% and 8% [95% UI 0-9.1 %], respectively). It is important to note that in this region approximately 5% (95% UI 0-18.2%) of outbreaks with known source were attributed to another category "meat", which cannot distinguish between the relative contributions of different animal sources. Among all other meat categories, pork played a minor role, with an attribution proportion between 3 to 5% across regions. The general term "poultry", turkey, or ducks was never cited as a source of any outbreaks in any region; however, chicken was mentioned as a source in a very few outbreaks in the AMR (0.3%, 95% UI 0.3-0.6%) and the EUR (0.1 %, 95% UI 0-1.5%). The proportion of STEC outbreaks attributed to an unknown source varied between 55% in AMR and 69% in WPR. Because data were only available from three out of six WHO regions, it was not possible to estimate global STEC source attribution proportions.

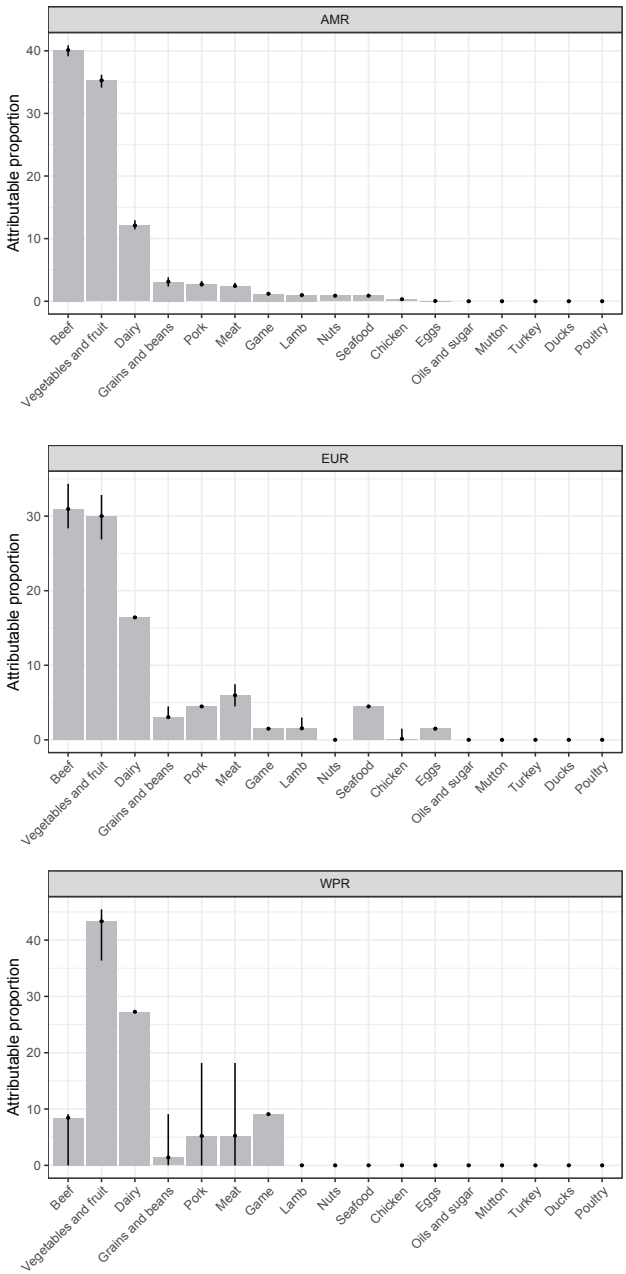


FIGURE 2. Relative contribution of food categories to STEC cases in WHO regions (mean %). Estimates exclude proportion of unknown-source outbreaks, i.e. were normalized to fit to 100%. *AMR: Region of the Americas; EUR: European Region; WPR: Western Pacific Region.

The estimates of the probability that an outbreak was caused by a given source are plotted in Figure 3. Results show that beef, produce (fruits and vegetables) and dairy were the sources with highest probability of being the cause of an STEC outbreak caused by a complex food. For example, if a complex food containing beef, grains, dairy and eggs was implicated in an outbreak, the probability that it was caused by beef was 40% (95% UI: 35-45%); by grains, 3% (95% UI: 1-5%); by eggs, 0.005% (95% UI: 0-0.01%); and by dairy, 14% (95% UI: 11-17%).

TABLE 2. Proportion of STEC cases attributed to foods in WHO regions (%, mean and 95% uncertainty interval [UI])

	AMR			EUR			WPR		
	Mean	95% UI		Mean	95% UI		Mean	95% UI	
Beef	18.3	17.8	18.6	11.8	10.8	13.1	2.7	0	2.9
Produce (fruits and vegetables)	16.1	15.5	16.5	11.4	10.2	12.5	13.6	11.4	14.3
Dairy	5.5	5.2	5.9	6.2	6.2	6.2	8.6	8.6	8.6
Grains and beans	1.4	1.1	1.7	1.2	1.1	1.7	0.4	0	2.9
Pork	1.2	1.1	1.5	1.7	1.7	1.7	1.6	0	5.7
Meat	1.1	1.1	1.3	2.3	1.7	2.8	1.7	0	5.7
Game	0.5	0.5	0.7	0.6	0.6	0.6	2.9	2.9	2.9
Lamb	0.4	0.4	0.5	0.6	0.6	1.1	0	0	0
Seafood	0.4	0.4	0.4	1.7	1.7	1.7	0	0	0
Nuts	0.4	0.4	0.4	0	0	0	0	0	0
Chicken	0.1	0.1	0.3	0	0	0.6	0	0	0
Eggs	0	0	0.1	0.6	0.6	0.6	0	0	0
Poultry	0	0	0	0	0	0	0	0	0
Ducks	0	0	0	0	0	0	0	0	0
Turkey	0	0	0	0	0	0	0	0	0
Mutton	0	0	0	0	0	0	0	0	0
Oils and sugar	0	0	0	0	0	0	0	0	0
Unknown	54.4	54.4	54.4	61.9	61.9	61.9	68.6	68.6	68.6

*AMR: Region of the Americas; EUR: European Region; WPR: Western Pacific Region.

To estimate the relative contribution of different food sources for severe STEC cases, we restricted the analysis to data from AMR, where most of the outbreaks involving HUS cases or deaths were reported. We found no significant differences between attribution proportions for mild and severe disease (results not shown).

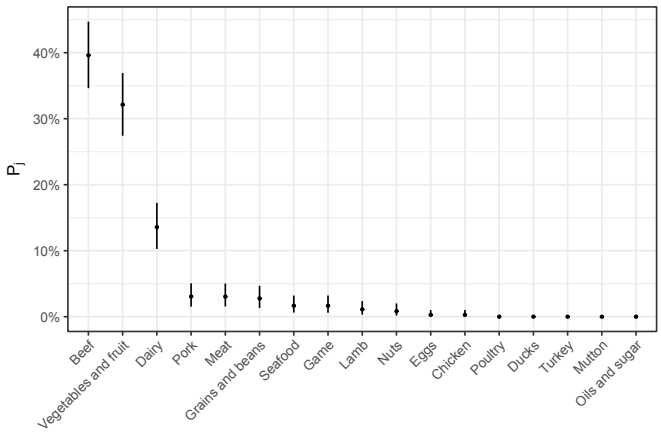


FIGURE 3. Estimates for P_j for food sources (median and 95% uncertainty interval)

4

A systematic review of case-control studies of sporadic infections

4.1 BACKGROUND AND OBJECTIVE

When investigating the specific food types associated with illnesses, different methods are used to investigate outbreaks versus sporadic illnesses. In outbreak situations, the goal is to identify the specific food exposure common across the cases, and both retrospective cohort and case-control studies are used to meet this objective. For sporadic cases of illness, case-control studies are the most common study design used to identify food types associated with illness. In these studies, the association of cases with various food exposures (typically through odds ratios [ORs]) can be quantified, and meta-analyses of these studies may be able to yield summary estimates for the range of food exposures of interest (Pires *et al.*, 2009). The objective of this work was to attribute sporadic illness caused by STEC to specific foods. The specific question addressed by this systematic review and meta-analysis was: what is the relative contribution of different foods to sporadic illnesses caused by STEC?

4.2 METHODS

The protocol for this review can be found in the PROSPERO Registry (Majowicz *et al.*, 2017; # CRD42017074239).

4.2.1 Assessment of existing systematic reviews

Existing or currently ongoing systematic reviews related to the research question were searched via the PROSPERO Registry and the Cochrane Library, as well as PubMed. Only one potentially relevant systematic review and meta-analysis was found, which examined the relative contribution of routes of exposure to STEC infection (Kintz *et al.*, 2017). Because this review did not assess specific foods (except for raw/undercooked meat), but rather broader routes of transmission (e.g. food, person-to-person) and only included larger (n≥20) studies of multiple designs (including but not limited to case-control studies), a new systematic review with a more in-depth analysis of different food categories was conducted here.

4.2.2 Population Exposure Comparator Outcome Study Design (PECOS)

The PECOS for this review (defined as per Sargeant and O'Connor, 2014) was as follows. The *population* was all human populations, with no limitations by age or other participant characteristics, location or context/settings. The *exposures* were all foods (e.g. hamburger, leafy greens); we did not consider drinking water (tap, bottled or other) as a food. The *comparator* group was individuals who are not ill with STEC infection (i.e. controls) and the *outcome* was sporadic illness caused by laboratory-confirmed STEC infection. The *study design* was case-control studies, and thus the effect measure of interest was the odds ratio (OR) – i.e. the relative odds of exposure to a given food for cases as compared to controls.

4.2.3 Search strategy

The search strategy was developed in consultation with a medical librarian and was reviewed by an expert in systematic reviews of foodborne disease who was not involved in the original strategy development. The search terms were developed through Medline Ovid and then adjusted as needed for each database searched.

PROSPERO was searched for studies on STEC to determine search terms commonly included in systematic reviews on STEC. Synonyms of STEC (e.g. verotoxigenic *E. coli*, Shiga toxigenic *E. coli*) were identified via relevant literature, expert consultation and via a PubMed literature search on STEC. To identify and include all potentially relevant STEC serogroups as search terms, the Joint FAO/WHO Core Expert Group on STEC/VTEC was consulted and a list of serogroups that have been found in humans was compiled (Annex 2); serogroups "O", "OR", "ON" and "OUT" were excluded from the list of search terms because – as stand-alone words – their inclusion generated a substantial number of irrelevant results. These serogroups were combined with the term "coli" during the search, to ensure results were relevant to *Escherichia coli*. Serogroups were also searched with "*Escherichia coli*" as a prefix (e.g. "*Escherichia coli* O157") to ensure sensitivity in the

search. Search filters provided by the University of Texas informed development of the study design terms for case-control studies (University of Texas, 2017). When possible, the case-control term in each database's thesaurus was expanded and combined with other chosen study design terms using the Boolean term "OR". Where possible, a population term for "humans" was included, specific to the database. For example, in Medline, articles with the tag "animals" were excluded to ensure that studies conducted solely in animal populations were excluded. A list of the final search terms is available on request.

As described above, the search was limited to human populations where possible. Otherwise, the search was not limited by language, location, study period or any other characteristics. Searches were conducted from 1 August to 30 September 2017, across the resources described below.

Seven databases of peer-reviewed literature (Medline [OVID], EMBASE, Scopus, CAB Direct, African Journals Online, Asia Journals Online and Latin America Journals Online), as well as the European Food Safety Authority (EFSA) journal and five databases of grey literature (ProQuest, E-Theses Online Services [ETHOS], OpenGrey, Agricultural Research Service and Current Research Information System) were searched. The main WHO website, as well as the six WHO regional websites, the FAO website and the Africa Centers for Disease Control and Prevention website were searched. Regional experts were consulted to identify any unpublished or pre-publication studies, specifically authors from Hooman *et al.* (2016) and Paudyal *et al.* (2017), WHO advisors from any identified STEC-related consultation reports on WHO websites, WHO regional public health contacts and members of the Joint FAO/WHO Core Expert Group on STEC/VTEC. Finally, citation reference lists of review articles on STEC (narrative, systematic or other) identified during the search were also reviewed for relevant articles, as well as the reference lists of the final set of references.

4.2.4 Citation collection, deduplication and screening

Citations were collected, managed, deduplicated and screened in RefWorks (ProQuest LLC, 2017). Attempts were made to translate articles in foreign languages; if suitable translation could not be obtained, the title and abstract were put through Google Translate for relevance screening and, if relevant, the entire article was reviewed and extracted by a native speaker of the article's original language. Cohen's kappa coefficient was used to measure interrater agreement between reviewers.

For the first stage (i.e. relevance screening), titles and abstracts were screened by two independent reviewers per reference, with a third reviewer to resolve any conflicts. First, the two reviewers screened 25 references together, to establish

agreement on how to apply the screening criteria; additional subsets were then independently screened and then discussed, until Cohen's kappa was 0.75 or greater. Then reviewers independently screened the remaining references using standardized instructions. Citations that fulfilled the inclusion criteria were advanced to full-text screening (i.e. the second stage of screening); advancement also occurred if the reviewer did not find sufficient information to determine whether the citation fulfilled the inclusion criteria. Inclusion criteria for relevance screening of the titles and abstracts were: the study is about STEC; the study is a case-control study; the study is done in humans; and the study is not an outbreak investigation.

For the second stage of screening (i.e. full-text screening), each complete article was screened by two independent reviewers per reference, with a third reviewer to resolve any conflicts, using standardized instructions. Studies that passed the second stage of screening advanced to data extraction. Inclusion criteria for this stage were: the study is done in humans; the study investigates the exposures (or risk factors) experienced by a series of cases, compared to the exposures (or risk factors) experienced by a series of controls; the controls are not cases of some other disease (e.g. another enteric infection, called a "case-case" study, or diarrhoeal controls); cases are individuals with illness caused by STEC; cases are sporadic (i.e. not from an outbreak); and the study assessed food exposures (even if no food exposures were statistically significant). Drinking water, breastfeeding and nasogastric feeding were excluded as foods, as were studies that assessed general nutrition (including malnutrition) as a risk factor for STEC infection.

4.2.5 Data extraction and quality assessment

Data were extracted by two reviewers, with a third reviewer to resolve any conflicts, using standardized forms and instructions. Authors of a convenience sample of eligible studies were contacted to provide the original questionnaires used in the case-control studies (if not available online). Extracted variables were: author; year published; study country and timeframe; characteristics of the study population (age, type); the cases (case definition, laboratory confirmation, how cases were identified) and the controls (how controls were identified and whether controls were matched to cases); case and control exclusion criteria (prior international travel, co-infection, secondary cases or individuals with ill family members); the type of STEC (e.g. O157, non-O157) and laboratory methods used; the sample size of cases and controls; the statistical methods used (including those employed to control confounding); and the specific food exposures assessed, including the numbers of cases and controls exposed (and unexposed) and the measure of association with 95% confidence interval (CI) and p-value (if reported). For papers in which the description of the laboratory methods used (as extracted from the methods section) was insufficient to determine appropriateness for identifying STEC, an expert in STEC laboratory identification assessed the full-text article,

and used information implicit in other parts of the paper (e.g. in the presentation of the results), as well as historical knowledge of the standard laboratory methods used by authoring institutions at different times, to assess the adequacy of laboratory methods used to identify cases.

Risk of bias was assessed using the Newcastle-Ottawa Quality Assessment Scale (http://www.ohri.ca/programs/clinical_epidemiology/nosgen.pdf), modified to address critical items for case-control studies using relevant questions from the RTI International–University of North Carolina at Chapel Hill Evidence-based Practice Center Item Bank and its modifications (Viswanathan and Berkman, 2012; Viswanathan *et al.*, 2013). Overall study quality was captured using the RTI Overall Assessment question ("are the results of the study believable taking study limitations into consideration?"), as well as a modified version of ROBINS-I, relevant to case-control studies (Sterne *et al.*, 2016). Finally, because the most important confounder given the study design and the topic (STEC infection related to food) was age, an assessment of whether age was adequately controlled was included as part of the quality assessment by assessing the combined impact of the study's design and analyses.

4.2.6 Analysis

Study countries were classified into WHO sub-regions (e.g. as in Kirk *et al.*, 2015). Food items were categorized using IFSAC's food categorization scheme (Richardson *et al.*, 2017), a hierarchical scheme of mutually exclusive food categories. Foods belonging to the same category but that were described as raw or undercooked, versus cooked, were also classified by this status. Within a single study, if more than one food fell within a particular category, a combined effect was calculated, to ensure that a study with several food exposures in the same food category did not have inflated influence on the summary/pooled estimate, as per Domingues *et al.* (2012a and 2012b). In our registered protocol, we stated that raw/undercooked foods would be treated as separate items than cooked foods. However, given that many of the food items were reported with unknown raw/cooked status, we chose instead to group raw and cooked food items (e.g. categorize raw beef, cooked beef, and beef of unknown status all as 'beef'), and explore the impact of raw/cooked/unknown status in the meta-regression.

Descriptive analyses were conducted to summarize study characteristics. To calculate the individual, study-specific ORs for each food for which results were reported, the following process was used. For all instances where the number of cases and controls who were either exposed or unexposed to a given food were reported in the paper directly (such as the four cells of a 2x2 table), these exact values were used to calculate the OR and standard error (SE). For the remaining

instances where ORs were reported, the reported univariate OR and 95% UI were used to back-calculate the OR and SE. To this end, an optimization process was designed in which the log-transformed OR was fitted to a normal distribution and the sum of squared differences between the observed and the fitted 95% UI minimized. For comparison purposes, an alternate approach was also explored, in which the reported univariate OR was used for all instances where such values were reported, and then the reported number of cases and controls who were either exposed or unexposed to a given food category were used for instances where ORs were not given.

Summary univariate ORs (i.e. pooled ORs) and their corresponding 95% UIs were calculated for each food category, both overall and by WHO sub-region, using a random effects model meta-analysis. Publication bias was assessed using Begg and Mazumdar's rank correlation test (1994) and Egger's regression test (1997). When significant publication bias was present, Duval and Tweedie's trim-and-fill method (2000) was used to explore the impact on model estimates.

For the food categories with significant overall associations, meta-regressions were conducted to explore the relationship between single study characteristics (i.e. WHO sub-region, publication year, study population age and cooking status) and the ORs for food exposures. All meta-analyses were carried out in R using the "metafor" package (Viechtbauer, 2010).

4.3 RESULTS

4.3.1 Numbers of citations identified

Results from the search, including the number of citations identified, are shown in Figure 4.

The majority of the 411 full-text articles screened were in English, but several were in other languages, specifically Japanese (n=9); Spanish (n=7); Portuguese (n=3); French (n=2); and Czech, Chinese, Dutch, German, Hungarian, Italian, Romanian, Slovenian and Thai (all n=1). From these 411 articles, 22 case-control studies of sporadic STEC infection in humans were identified, from ten countries within four WHO sub-regions (AMR A, AMR B, EUR A, WPR A), conducted from 1985 to 2012 (Table 3); study locations and timeframes are also shown in Figure 5. All 22 studies were published in English and were from the peer-reviewed, indexed literature.

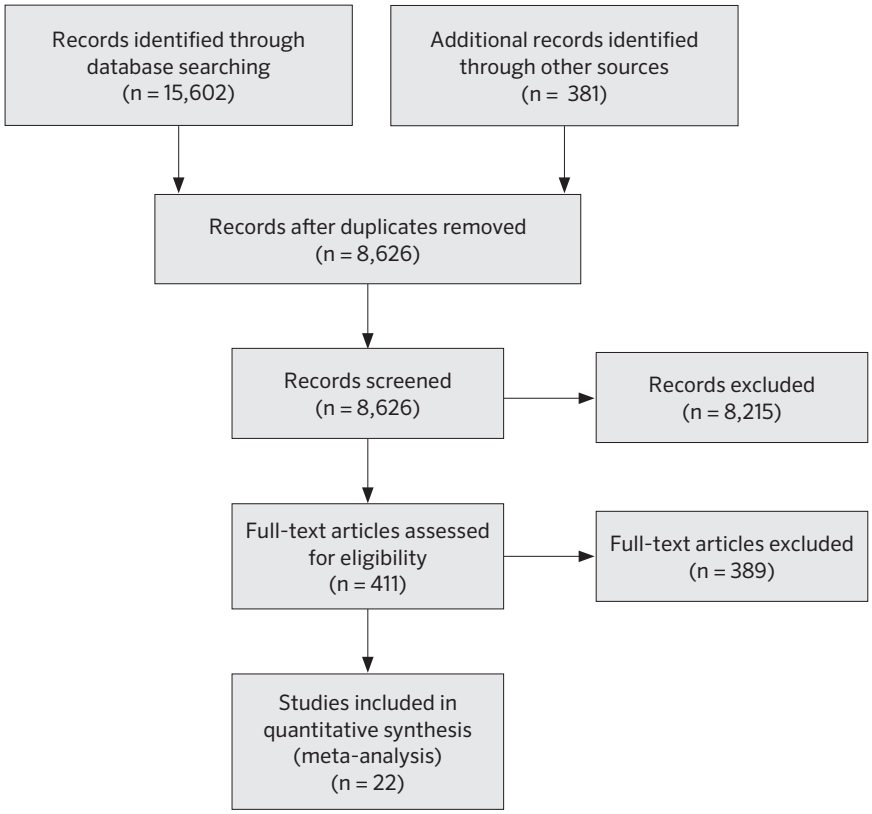


FIGURE 4. PRISMA diagram showing the results of the search for case-control studies of sporadic STEC infections in humans (all dates and locations)

TABLE 3. Characteristics of the 22 case-control studies of non-outbreak (i.e. sporadic) STEC infection in humans, ordered by study timeframe (oldest to newest)

Lead Author (Year Published)	Country (WHO sub-region)	Study Timeframe	Study Pop. Age	Study Pop. Type	Types of Cases (all lab. confirmed?)	Case Finding Method	Control Type	STEC Category	Lab. Methods Adequate to identify STEC?
MacDonald (1988)	USA (AMR A)	1985-1986	All	Patients of specific facilities	Non-outbreak GI illness with positive lab. result (yes)	Active case finding (at laboratory level)	Facility/ practice	O157	Yes (confirmed STEC O157:H7)
Bryant (1989)	Canada (AMR A)	1986-1987	All	Patients of specific facilities	Non-outbreak GI illness with positive lab. result (yes)	Active case finding (in ER)	Friends	O157	Yes (confirmed STEC O157)
Le Saux (1993)	Canada (AMR A)	1990	All	General pop.	Non-outbreak positive lab. Result (yes)	Active case finding (at laboratory level)	Neighbours	O157	Yes (confirmed STEC O157:H7)
Rowe (1993)	Canada (AMR A)	1990	Children (0-14 years)	Patients of specific facilities	Post-diarrhoea cases of HUS (no; 88% were + for VTEC)	Active case finding (by physicians)	Facility/ practice	STEC	Yes (confirmed STEC)
Slutsker (1998)	USA (AMR A)	1990-1992	All	Patients of specific facilities	Non-outbreak GI illness with positive lab. result (yes)	Active case finding (at laboratory level)	Facility/ practice	O157	Yes (confirmed STEC O157)
Holton (1999)	Canada (AMR A)	1991	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Lab.-based surveillance with public health notification	Neighbours	O157	Yes (confirmed STEC O157:H7)
CDC (1995)	USA (AMR A)	1994	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Lab.-based surveillance with public health notification	Inadequately described	O157	Inadequately described

(cont.)

Lead Author (Year Published)	Country (WHO sub-region)	Study Timeframe	Study Pop. Age	Study Pop. Type	Types of Cases (all lab. confirmed?)	Case Finding Method	Control Type	STEC Category	Lab. Methods Adequate to identify STEC?
Mead (1997)	USA (AMR A)	1994	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Active case finding (at laboratory level)	Population	O157	Yes (confirmed STEC O157:H7)
Parry (1998)	UK (EUR A)	1994-1996	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Active case finding (at laboratory level)	Facility/ practice	O157	Yes (confirmed STEC O157)
O'Brien (2001)	UK (EUR A)	1996-1997	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Active case finding (at laboratory level)	Facility/ practice	O157	Yes (confirmed STEC O157)
Kassenborg (2004)	USA (AMR A)	1996-1997	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Active case finding (at laboratory level)	Population	O157	Yes (confirmed STEC O157)
Pierard (1999)	Belgium (EUR A)	Inadequately described; 1990's	All	Patients of specific facilities	Non-outbreak GI illness, or HUS, with positive lab result (yes)	Active case finding (at laboratory level)	Facility/ practice	STEC	Yes (confirmed STEC)
Locking (2001)	UK (EUR A)	1996-1999	All	General pop.	Non-outbreak GI illness, or HUS, with positive lab result (yes)	Lab.-based surveillance with public health notification	Facility/ practice	O157	Yes (confirmed STEC O157)
Voestch (2007)	USA (AMR A)	1999-2000	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Active case finding (at laboratory level)	Population	O157	Yes (confirmed STEC O157)
Vaillant (2009)	France (EUR A)	2000-2001	Children (0-15 years)	General pop.	Post-diarrhoeal HUS with confirmation of STEC (yes)	HUS surveillance with public health notification	Facility/ practice	STEC	Yes (confirmed STEC)

(cont.)

Lead Author (Year Published)	Country (WHO sub-region)	Study Timeframe	Study Pop. Age	Study Pop. Type	Types of Cases (all lab. confirmed?)	Case Finding Method	Control Type	STEC Category	Lab. Methods Adequate to identify STEC?
Rivas (2008)	Argentina (AMR B)	2001-2002	Children (0-15 years)	Patients of specific facilities	Non-outbreak GI illness, or HUS, with positive lab result (yes); also post-diarrhoeal HUS (no)	Health record review	Neighbours	O157	Yes (confirmed STEC O157)
Werber (2007)	Germany (EUR A)	2001-2003	All	General pop.	Non-outbreak GI illness, or HUS, with positive lab result (yes)	Lab.-based surveillance with public health notification	Population	STEC	Yes (confirmed STEC)
Hundy (2004)	Australia (WPR A)	2002	All	General pop.	Non-outbreak illness/blood in stool with positive lab result (no)	Lab.-based surveillance with public health notification	Population	STEC	Yes (presumptive STEC)
Denno (2009)	USA (AMR A)	2003-2005	Children (0-19 years)	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Lab.-based surveillance with public health notification	Practice/ facility	O157	Yes (confirmed STEC O157)
McPherson (2009)	Australia (WPR A)	2003-2007	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Lab.-based surveillance with public health notification	Population	STEC	Yes (confirmed STEC)
Friesema (2015)	The Netherlands (EUR A)	2008-2012	All	General pop.	Non-outbreak GI illness with positive lab. result (yes)	Lab.-based surveillance with public health notification	Population	STEC	Yes (confirmed STEC)
Jaros (2013)	New Zealand (WPR A)	2011-2012	All	General pop.	Non-outbreak GI illness, or HUS, with positive lab result (yes)	Lab.-based surveillance with public health notification	Population	STEC O157	Yes (confirmed STEC)

Lead author	Country	Study timeframe																												
		1985	1986	1987	1988	1989	1990	1991	1992	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015
AMR A																																
Bryant	Canada					x																										
Le Saux	Canada									x																						
Rowe	Canada									x																						
Holton	Canada															x																
MacDonald	USA				x																											
Slutsker	USA														x																	
Mead	USA													x																		
CDC	USA											x																				
Kassenborg	USA																				x											
Voestch	USA																							x								
Denno	USA																									x						
AMR B																																
Rivas	Argentina																								x							
EUR A																																
Parry	United Kingdom														x																	
O'Brien	United Kingdom																	x														
Locking	United Kingdom																	x														
Vaillant	France																									x						
Pierrard	Belgium*							*	*	*	*	*	*	*	*	*x																
Friesema	Netherlands																															x
Werber	Germany																							x								
WPR A																																
Hundy	Australia																				x											
McPherson	Australia																									x						
Jaros	New Zealand																													x		

* specific years not reported

FIGURE 5. Study locations and timeframes for the 22 identified case-control studies of sporadic STEC infections in humans

Although website searches identified experts who were contacted, neither websites nor citation reference lists identified new studies beyond those found within the peer-reviewed literature databases. The grey literature search did identify two potentially relevant doctoral theses, one from United Kingdom (Kemp, 2005) and one

from France (Espie, 2007), but the full-text documents were unable to be accessed and thus were not assessed. Expert consultation identified a case-control study of sporadic human non-O157 STEC infections in the the United States of America but the results were unavailable at the time of the analysis.

4.3.2 Description of the identified studies

In terms of study quality, all studies used an adequate case definition, but for three of the 22 studies (14%; Rowe *et al.*, 1993; Pierard *et al.*, 1999; Rivas *et al.*, 2008), it was difficult to determine whether the cases were representative based on the information provided. Of the 22 studies, 20 (91%) contained enough detail to demonstrate that the laboratory methods used were adequate to identify STEC, one had a description that supported identification of presumptive STEC (Hundy *et al.*, 2004), and one did not provide adequate information within the text of the paper to assess laboratory methods (CDC, 1995; Table 3).

Only one study (CDC, 1995), published as a short report, did not provide an adequate description of control selection nor definition of controls as "healthy" or without gastrointestinal infection or other similar characteristics. All other studies used controls that were without symptoms of current gastrointestinal infection. Thirteen of the studies (59%) used different methods to identify cases versus controls (Table 3); in all these studies, cases were identified via existing health system mechanisms, including laboratory-based surveillance, whereas controls were predominantly identified via random or semi-random sampling from the population. However, when considering feasibility, validity, ethical, and other issues, the selection of controls was considered appropriate in 21 of the 22 studies (96%) – i.e. controls represent the population from which the cases arose and, if the controls had acquired STEC infection, they would have been included as cases in the study – and was unable to be assessed for only one study (CDC, 1995).

Assessing how studies controlled for age in the study design, analysis, or both, two of the 22 studies (9%; MacDonald *et al.*, 1988; Rowe *et al.*, 1990) did not appear to fully and adequately control for age. Both these studies matched on age during control selection, but they did not account for this in their analysis (i.e. calculated unmatched ORs). Of the 20 studies that adequately controlled for age, two (10%) did not match on age during control selection, but adequately adjusted for age by including it in their regression models (Friesema *et al.*, 2015; Jaros *et al.*, 2013), and the remaining 18 (90%) matched on age during control selection, as well as conducted analysis that accounted for matching on age.

In all studies, exposures were ascertained via interview using comparable questions for cases versus controls; however, in only one study (Bryant *et al.*, 1989) did the interviewers appear to be blinded to the case or control status of the participant

when assessing participant exposures. Case and control exposure assessment appeared to vary mainly by the exposure window applied. All studies assessed case exposures during the incubation period prior to illness, whereas for controls, half the studies assessed control exposure during the same calendar period as the cases, while the other half assessed control exposure during the window prior to control interview (Table 4). Descriptions of non-response rates for cases and controls, and descriptions of non-respondents were infrequent, in that in 17 of the 22 studies (77%), these details were not provided (Table 4). In 20 of the 22 studies (91%), the statistical methods applied were considered adequate to determine ORs for food exposures; in one study, there was insufficient information to make this assessment (CDC, 1995) and in one study the statistical methods were considered inadequate (Rowe *et al.*, 1990).

TABLE 4. Selected quality assessment indicators for the 22 case-control studies of non-outbreak (i.e. sporadic) STEC infection in humans, ordered by study timeframe (oldest to newest)

Lead Author (Year Published)	Country (WHO sub-region)	Study Timeframe	Exposure Window: Cases	Exposure Window: Controls	Non-Response Rate and Non-Respondents	ROBINS-I*
MacDonald (1988)	USA (AMR A)	1985-1986	7 d prior to illness	7 d prior to interview	Not described	4
Bryant (1989)	Canada (AMR A)	1986-1987	7 d prior to illness	Same calendar dates as case	Same non-response rate for cases and controls	4
Le Saux (1993)	Canada (AMR A)	1990	10 d prior to illness	Same calendar dates as case	Not described	2
Rowe (1993)	Canada (AMR A)	1990	14 d prior to illness	14 d prior to interview	Not described	4
Slutsker (1998)	USA (AMR A)	1990-1992	7 d prior to illness	7 d prior to interview	Not described	2
Holton (1999)	Canada (AMR A)	1991	7 d prior to illness	Same calendar dates as case	Not described	2
CDC (1995)	USA (AMR A)	1994	7 d prior to illness	Not reported	Not described	6
Mead (1997)	USA (AMR A)	1994	7 d prior to illness	Same calendar dates as case	Different non-response rates for cases versus controls, with non-respondents described	2

(cont.)

Lead Author (Year Published)	Country (WHO sub-region)	Study Timeframe	Exposure Window: Cases	Exposure Window: Controls	Non-Response Rate and Non-Respondents	ROBINS-I*
Parry (1998)	UK (EUR A)	1994-1996	7 d prior to illness	Same calendar dates as case	Not described	1
O'Brien (2001)	UK (EUR A)	1996-1997	5 d prior to illness	Same calendar dates as case	Different non-response rates for cases versus controls, with non-respondents not described	1
Kassenborg (2004)	USA (AMR A)	1996-1997	5 d prior to illness	5 d prior to interview	Not described	1
Pierard (1999)	Belgium (EUR A)	Inadequately described; 1990s	14 d prior to illness	14 d prior to interview	Not described	2
Locking (2001)	UK (EUR A)	1996-1999	14 d prior to illness	Same calendar dates as case	Not described	2
Voestch (2007)	USA (AMR A)	1999-2000	7 d prior to illness	Same calendar dates as case	Not described	1
Valliant (2009)	France (EUR A)	2000-2001	7 d prior to illness	Same calendar dates as case	Not described	2
Rivas (2008)	Argentina (AMR B)	2001-2002	7 d prior to illness	Same calendar dates as case	Not described	2
Werber (2007)	Germany (EUR A)	2001-2003	10 d prior to illness	10 d prior to interview	Different non-response rates for cases versus controls, with non-respondents not described	1
Hundy (2004)	Australia (WPR A)	2002	10 d prior to illness	10 d prior to interview	Not described	2
Denno (2009)	USA (AMR A)	2003-2005	2-8 d prior to illness	2-8 d prior to interview	Not described	1
McPherson (2009)	Australia (WPR A)	2003-2007	10 d prior to illness	10 d prior to interview	Not described	2

(cont.)

Lead Author (Year Published)	Country (WHO sub-region)	Study Timeframe	Exposure Window: Cases	Exposure Window: Controls	Non-Response Rate and Non-Respondents	ROBINS-I*
Friesema (2015)	The Netherlands (EUR A)	2008-2012	7 d prior to illness	7 d prior to interview	Not described	2
Jaros (2013)	New Zealand (WPR A)	2011-2012	14 d prior to illness	14 d prior to interview	Different non-response rates for cases versus controls, with non-respondents described	2

*Modified ROBINS-I categories:

1 – low risk of bias in the reported ORs for food exposures

2 – moderate risk of bias in the reported ORs for food exposures, with the bias likely towards the null (i.e. towards an OR=1)

3 – moderate risk of bias in the reported ORs for food exposures, with the bias likely away from the null (i.e. away from an OR=1)

4 – serious risk of bias (either towards or away from the null): the study has some important problems

5 – critical risk of bias (either towards or away from the null): the study is too problematic to provide useful evidence

6 – no information

In considering all quality assessment items together, 20 of the 22 studies (91%) were considered reliable, taking study limitations into consideration, whereas two studies (CDC, 1995; Rowe *et al.*, 1990) were not. Of the 22 studies, six (27%) were assessed to have a low risk of bias in the reported ORs for food exposures; 12 (55%) were assessed to have a moderate risk of bias in the reported ORs for food exposures, with the bias likely towards the null (i.e. towards an OR=1); three (14%) were assessed to have a serious risk of bias (either towards or away from the null); and one did not have adequate information to make an assessment (Table 4).

Of the 22 studies, 18 (82%; Table 3) included individuals of all ages, with 15 providing results for all age groups combined in their estimates, two providing results stratified by age (Werber *et al.*, 2007; Friesema *et al.*, 2015) and one providing results for both all participants combined and for the subset of children (Pierard *et al.*, 1999). Of the 22 studies, four (18%) included only children. Most studies (16/22; 73%) included cases and controls drawn from the general population, with cases identified either via existing laboratory surveillance with public health notification (10/16; 63%) or via active case ascertainment at the laboratory level (6/16; 38%), and controls identified via random/semi-random sampling of the general population – including via existing registries, control databases or random digit dialing – (8/16; 50%) or via the same facility or practice (5/16; 31%) or the same neighbourhood as the case (2/16; 13%). The remaining six of the 22 studies (27%) drew cases from specific facilities, via active case ascertainment within labora-

tories (3/6; 50%), emergency rooms (1/6; 17%) and by physicians (1/6; 17%), as well as via health record reviews (1/6; 17%). These six studies selected controls from the same facility as the cases (4/6; 66%), as well as from the case's friends (1/6; 17%) and neighbours (1/6; 17%). In 20 of the 22 studies (91%), cases were defined as symptomatic individuals with laboratory confirmation of STEC; in one study (Rowe *et al*., 1993) cases were those with post-diarrhoeal hemolytic-uremic syndrome (of whom 88% were positive for VTEC) and in one study (Rivas *et al*., 2008) cases were either symptomatic individuals with laboratory confirmation of STEC or those with post-diarrhoeal hemolytic-uremic syndrome.

4.3.3 Food items associated with STEC infection

Of the 22 papers, 21 provided extractable information on the relative odds of exposure to a given food for cases as compared to controls (Denno *et al*., 2009 did not report data we could extract). Thus, data were extracted from 21 papers, for 245 individual measures in 11 food categories and across three status types: raw or undercooked; not raw (i.e. adequately cooked, treated, pasteurized or other mechanism); and unknown (Table 5). The dairy category included cheeses and cheese products, cream and milk; only two of the dairy foods described in the identified papers provided information about the animal origin of the product (ewes' milk cheese and goats' milk cheese; Vaillant *et al*., 2009) so this category could not be further divided by animal source. Similarly, animal source was not provided for "eggs", which were reported in two studies from the UK and Australia (Locking *et al*., 2001; McPherson *et al*., 2009). The 62 items classified as "meat – unspecified" included items that could not be assigned to their animal food origin (e.g. beef, pork), such as sliced and minced meat, sausages, meat casseroles, hot dogs, kebabs, salami, as well as generic terms like "meat". The 38 items classified as "produce" included specific fruits and vegetables (including various stone fruits, berries, cantaloupe, watermelon, fresh fruit juice, various root vegetables, various leafy greens, tomatoes), as well as the generic terms "fruits" and "vegetables". Because there were very few results per specific produce item, we did not divide this category further. The eight items classified as "seafood" included the generic terms "fish", "shellfish", and "seafood".

Overall, beef and meat–unspecified were significantly associated with sporadic STEC infection, although meat-unspecified became non-significant when the trim-and-fill method was used (Table 6; Figure 6). Produce (fruits and vegetables), dairy, eggs and poultry/game-unspecified were also significant but had ORs of less than one. When the alternate approach (see methods) to determining the ORs for each study/food was applied, estimates of the summary ORs did not change in magnitude, nor direction, nor significance (Annex 3).

TABLE 5. Categories of the 245 food items extracted from the 21 case-control studies of non-outbreak (i.e. sporadic) STEC infection in humans that reported useable data, ranked in descending order by the number of food items per category

Food Category (no. items within category)	Number of items by cooked or processed status of the food item		
	Raw or undercooked	Not raw (i.e. cooked, treated, pasteurized)	Unknown/not reported
Beef (83)	35	1	47
Meat – unspecified (62)	14	10	38
Produce (fruits and vegetables) (38)	16	-	22
Dairy (25)	11	2	12
Chicken (10)	1	1	8
Seafood (8)	-	-	8
Pork (7)		-	7
Eggs (5)	-	-	5
Lamb (3)	-	-	3
Turkey (2)	-	-	2
Poultry/Game – unspecified (2)	-	-	2

TABLE 6. Results of the meta-analysis, showing pooled univariate odds ratios (ORs) per food category (significant values shown in **bold**), ranked in descending order by the number of food items in the category

Food Category (no. items within category)	Odds ratio (95% UI)	p-value	p-value Regression test	p-value Rank test	Trim-and-Fill Method – Odds ratio (95% UI)	p-value
Beef (80*)	**1.667 (1.408, 1.975)**	**<0.001**	**<0.001**	**0.008**	**1.437 (1.205, 1.713)**	**<0.001**
Meat - unspecified (60*)	**1.281 (1.090, 1.506)**	**0.003**	**<0.001**	**0.007**	1.069 (0.894, 1.279)	0.463
Produce (fruits and vegetables) (38)	**0.671 (0.534, 0.843)**	**<0.001**	**0.035**	0.119	**0.671 (0.534, 0.843)**	**<0.001**
Dairy (23*)	**0.734 (0.558, 0.966)**	**0.027**	**0.048**	0.319	**0.673 (0.500, 0.906)**	**0.009**
Chicken (9*)	0.827 (0.377, 1.814)	0.636	0.517	0.358		
Seafood (8)	0.758 (0.457, 1.256)	0.282	0.902	0.905		

(cont.)

Pork (7)	1.032 (0.632, 1.685)	0.900	0.201	0.239
Eggs (5)	**0.658 (0.515, 0.841)**	**<0.001**	0.504	0.483
Lamb (3)	1.936 (0.582, 6.441)	0.282	0.072	0.333
Turkey (2)	1.055 (0.085, 13.102)	0.967	N/A	1.000
Poultry/Game - unspecified (2)	**0.411 (0.228, 0.740)**	**0.003**	N/A	1.000

* This number is less than in Table 5 because some food items as reported did not have sufficient useable data

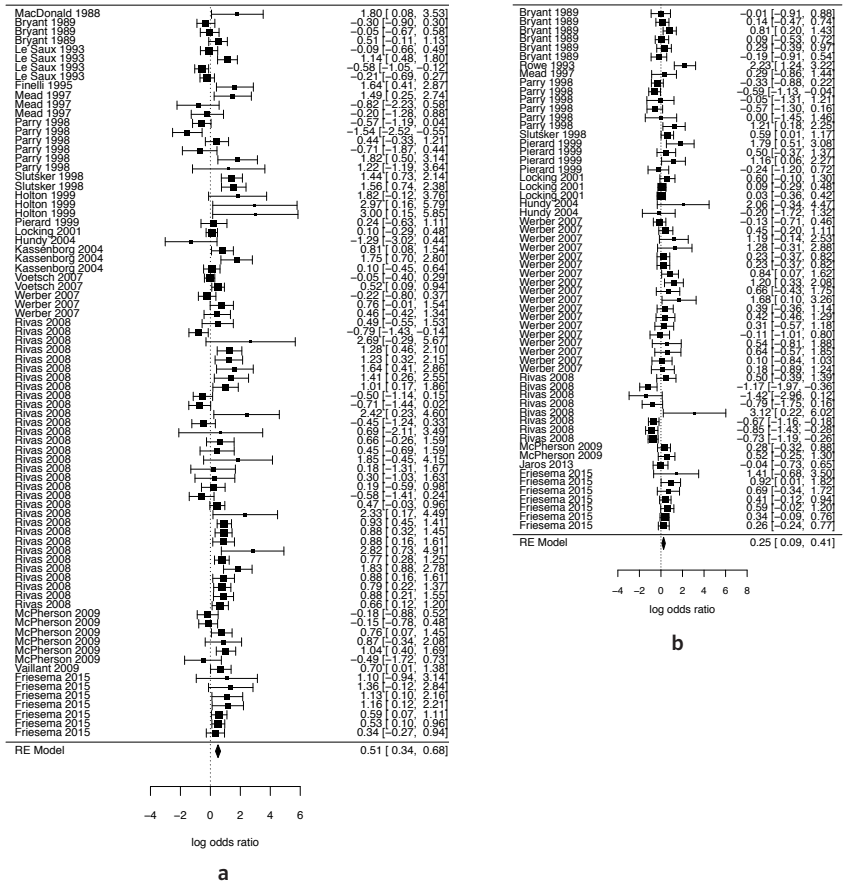


FIGURE 6. Forest plots of the log odds ratio (OR) of the risk of human STEC infection from beef (a) and meat-unspecified (b), showing the overall pooled OR together with the 95% UI; ordered from oldest (top) to newest (bottom) study.

Significant food categories varied moderately by WHO sub-region (Table 7). In AMR A, beef and meat–unspecified remained the significant risk factors for STEC, whereas in AMR B and EUR A the only significant risk factor was beef, and in WPR A the only significant risk factor was chicken. Under the alternate approach to determining the ORs for each study/food, our estimates of the summary ORs by WHO sub-region changed: in AMR A both beef and meat-unspecified became non-significant and in EUR A meat-unspecified became significant (Annex 3).

TABLE 7. Results of the meta-analysis for each World Health Organization (WHO) sub-region, showing pooled univariate odds ratios (ORs) per food category (significant values shown in **bold**)

Food Category	WHO Sub-Region AMR A[1]		WHO Sub-Region AMR B[2]		WHO Sub-Region EUR A[3]		WHO Sub-Region WPR A[4]	
	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)
Beef	22	**1.548 (1.086, 2.207)** †	32	**1.555 (1.173, 2.063)** †	19	**1.429 (1.044, 1.956)**	7	1.243 (0.730, 2.118)
Meat - unspecified	9	**1.545 (1.033, 2.310)**	8	**0.518 (0.380, 0.704)** †	38	1.172 (0.988, 1.391) †	5	1.295 (0.891, 1.882)
Produce (fruits and vegetables)	9	**0.520 (0.369, 0.734)**	0	N/A	17	0.872 (0.658, 1.158)	12	0.476 (0.188, 1.206) †
Dairy	1	9.774 (0.981, 97.360)	0	N/A	20	**0.670 (0.507, 0.886)**	2	1.209 (0.695, 2.101)
Chicken	4	**0.335 (0.221, 0.507)**	0	N/A	2	1.320 (0.170, 10.273)	3	**2.689 (1.357, 5.326)**
Seafood	2	0.683 (0.417, 1.118)	0	N/A	5	0.932 (0.385, 2.258)	1	**0.452 (0.295, 0.693)**
Pork	2	1.430 (0.841, 2.431)	2	1.107 (0.320, 3.830)	0	N/A	3	**0.527 (0.379, 0.733)** †
Eggs	0	N/A	0	N/A	1	**0.675 (0.477, 0.956)**	4	**0.642 (0.455, 0.907)**
Lamb	0	N/A	0	N/A	3	1.936 (0.582, 6.441)	0	N/A

(cont.)

Food Category	WHO Sub-Region AMR A[1]		WHO Sub-Region AMR B[2]		WHO Sub-Region EUR A[3]		WHO Sub-Region WPR A[4]	
	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)
Poultry/ Game - unspecified	0	N/A	0	N/A	2	**0.411 (0.228, 0.740)**	0	N/A

[1] AMR A includes the following countries: Canada; Cuba; United States of America.
[2] AMR B includes the following countries: Antigua and Barbuda; Argentina; Bahamas; Barbados; Belize; Brazil; Chile; Colombia; Costa Rica; Dominica; Dominican Republic; El Salvador; Grenada; Guyana; Honduras; Jamaica; Mexico; Panama; Paraguay; Saint Kitts and Nevis; Saint Lucia; Saint Vincent and the Grenadines; Suriname; Trinidad and Tobago; Uruguay; Venezuela (Bolivarian Republic of).
[3] EUR A includes the following countries: Andorra; Austria; Belgium; Croatia; Cyprus; Czech Republic; Denmark; Finland; France; Germany; Greece; Iceland; Ireland; Israel; Italy; Luxembourg; Malta; Monaco; Netherlands; Norway; Portugal; San Marino; Slovenia; Spain; Sweden; Switzerland; United Kingdom.
[4] WPR A includes the following countries: Australia; Brunei Darussalam; Japan; New Zealand; Singapore.

[†] Using trim-and-fill method

The exploratory analysis of the association between different study characteristics and ORs for food categories is shown in Table 8; note that results are shown only for food categories with greater than 20 food items; for those with fewer than 20 items (i.e. chicken, seafood, pork, eggs, lamb, turkey and poultry/game–unspecified) the findings may be spurious due to lack of data and consequently are not interpreted. Study population age was significant for dairy (with studies in children yielding lower ORs than studies of all ages). Study sub-region was significant for meat–unspecified (with studies from AMR B yielding lower ORs than studies from AMR A), produce (fruits and vegetables) (with studies from EUR A yielding higher ORs than studies from AMR A) and dairy (with studies from EUR A yielding lower ORs than studies from AMR A). Measures of study quality were significant for meat–unspecified and dairy (with studies whose findings were considered not reliable yielding higher ORs than studies whose findings were considered reliable). Publication year and whether the food item was raw/undercooked, not raw, or unknown were not significant moderating factors. Results under the alternate approach varied slightly and are given in Annex 4.

TABLE 8. Univariate odds ratios (ORs) of study characteristics, by food category for foods with ≥20 items, with significant values shown in **bold**

Study Characteristic	Characteristic level	OR	95% C.I.	p-value
Beef (n=80)				
WHO subregion	AMR A (reference)	—	—	—
	AMR B	1.121	0.732, 1.717	0.600
	EUR A	0.840	0.522, 1.351	0.473
	WPR A	0.714	0.368, 1.385	0.319

(cont.)

Publication year		1.019	0.995, 1.044	0.128
Study population age	All (reference)	—	—	—
Study Characteristic	**Characteristic level**	**OR**	**95% C.I.**	**p-value**
Beef (n=80)				
	Adults	1.327	0.689, 2.552	0.397
	Children	1.336	0.941, 1.898	0.105
Food item status	Not raw (reference)	—	—	—
	Raw or undercooked	2.404	0.892, 6.483	0.083
	Unknown	1.036	0.387, 2.776	0.944
Robin's I	1 (reference)	—	—	—
	2	1.389	0.903, 2.137	0.135
	4	1.009	0.448, 2.274	0.982
	6	4.002	0.702, 22.804	0.118
Believeable findings	Yes (reference)	—	—	—
	No	3.122	0.564, 17.270	0.192
Meat–unspecified (n=60)				
WHO subregion	AMR A (reference)	—	—	—
	AMR B	**0.345**	**0.216, 0.549**	**<0.001**
	EUR A	0.915	0.645, 1.298	0.619
	WPR A	0.866	0.488, 1.537	0.624
Publication year		0.999	0.978, 1.021	0.935
Study population age	All (reference)	—	—	—
	Adults	1.453	0.940, 2.246	0.093
	Children	0.921	0.651, 1.302	0.641
Food item status	Not raw (reference)	—	—	—
	Raw or undercooked	1.577	0.959, 2.595	0.073
	Unknown	1.266	0.825, 1.941	0.280
Robin's I	1 (reference)	—	—	—
	2	0.868	0.610, 1.235	0.431
	4	1.126	0.671, 1.891	0.654
	6	N/A	N/A	N/A
Believeable findings	Yes (reference)	—	—	—
	No	**7.523**	**2.073, 27.302**	**0.002**
Produce (fruits and vegetables) (n=38)				
WHO subregion	AMR A (reference)	—	—	—
	AMR B	N/A	N/A	N/A
	EUR A	**1.707**	**1.010, 2.884**	**0.046**

(cont.)

	WPR A	1.149	0.650, 2.029	0.633
Publication year		0.976	0.939, 1.014	0.212
Study Characteristic	**Characteristic level**	**OR**	**95% C.I.**	**p-value**
Produce (fruits and vegetables) (n=38)				
Study population age	All (reference)	—	—	—
	Adults	0.888	0.412, 1.916	0.762
	Children	1.077	0.585, 1.982	0.812
Food item status	Not raw	N/A	N/A	N/A
	Raw or undercooked (reference)	—	—	—
	Unknown	1.035	0.649, 1.650	0.885
Robin's I	1 (reference)	—	—	—
	2	0.841	0.523, 1.352	0.475
	4	N/A	N/A	N/A
	6	N/A	N/A	N/A
Believeable findings	Yes (reference)	—	—	—
	No	N/A	N/A	N/A
Dairy (n=23)				
WHO subregion	AMR A (reference)	—	—	—
	AMR B	N/A	N/A	N/A
	EUR A	**0.068**	**0.006, 0.807**	**0.033**
	WPR A	0.127	0.009, 1.695	0.118
Publication year		0.954	0.909, 1.000	0.051
Study population age	All (reference)	—	—	—
	Adults	0.887	0.374, 2.104	0.786
	Children	**0.580**	**0.349, 0.964**	**0.035**
Food item status	Not raw (reference)	N/A	N/A	N/A
	Raw or undercooked	1.183	0.485, 2.887	0.711
	Unknown	0.557	0.247, 1.254	0.157
Robin's I	1 (reference)	—	—	—
	2	0.638	0.350, 1.163	0.143
	4	10.040	0.789, 127.810	0.076
	6	N/A	N/A	N/A
Believeable findings	Yes (reference)	—	—	—
	No	**13.783**	**1.145, 165.894**	**0.039**

5

Discussion and conclusions

5.1 DISCUSSION OF THE MAIN FINDINGS

Beef, produce (fruits and vegetables), dairy products, other unspecified types of meat, and chicken all emerged as significant sources of STEC, depending on geographic region and whether the cases being considered were from outbreaks or sporadic.

The analysis of outbreak data showed that the most important sources of STEC globally were produce (fruits and vegetables), beef and dairy products. The ranking of the top three food categories varied between regions. Beef and produce (fruits and vegetables) were estimated to have the highest proportion of STEC cases attributed in the AMR and EUR regions. In WPR, dairy appeared to play a more important role, followed by produce (fruits and vegetables); beef ranked third. Possible explanations for regional variability include differences in the proportion of specific foods in the diet and how they are prepared for consumption, the level of STEC contamination of foods and live animals from which foods are derived, the virulence characteristics of regionally predominant STEC strains, or differences in how outbreaks are detected, investigated and reported. More than half of the outbreaks globally could not be attributed to any source.

The overall assumption of the outbreak analysis model is that the estimated attribution proportions based on outbreak data can be used to attribute the overall burden of STEC infections (i.e. the total incidence, including both outbreak-associated and sporadic cases) (Painter *et al.*, 2013; Pires *et al.*, 2010). However, a number of

uncertainties are linked to this assumption. Some foods are more likely to cause outbreaks than others, and some foods are associated with larger numbers of cases per outbreak; thus, the relative importance of sources of outbreak-associated cases may not be representative of the overall contribution of sources for the total burden of disease (Pires *et al.*, 2009). The estimated relative contribution of each food type depends on the types of foods and situations that result in an outbreak being identified and successfully investigated. For example, outbreaks in groups of children may be more frequently identified than outbreaks in young adults. Likewise, outbreaks in restaurants or large groups are more likely to be detected. Thus, certain food-risk groups and smaller outbreaks may be underrepresented in the available data and more data are required to improve estimates. Overall, estimates inevitably depend on the selection of sources to be examined in the event of an outbreak, as well as the reporting capacity of each country. To avoid potential overestimation of the importance of sources that have caused a small number of large outbreaks – e.g. foods with large production chains – the number of ill people implicated in the outbreaks was not considered in the analysis. To minimize potential bias introduced by large outbreaks, we chose not to adjust for outbreak size, but rather to disregard the number of reported cases in each outbreak and simply consider the number of outbreaks caused by each food. This means that each outbreak was considered as one single "case", which could be comparable to a sporadic case. Although our approach can also introduce bias and artificially reduce the relative importance of foods that frequently cause many illnesses, it provides confidence when extrapolating our attribution proportion results to all STEC cases (i.e. sporadic and outbreak cases). Foods identified in outbreak investigations may not well represent foods responsible for sporadic disease. Although a study found that outbreak and sporadic infections caused by four priority pathogens (*Salmonella, Campylobacter,* STEC O157 and *Listeria monocytogenes*) were similar in the United States, a number of published studies have noted that food sources for some pathogens can vary substantially (IFSAC, 2015; Painter *et al.*, 2013; Pires *et al.*, 2010). For STEC, potential differences are relevant for sources that are frequently involved in outbreaks (raw produce (fruits and vegetables), unpasteurized dairy products) but less likely to cause sporadic cases, either because contamination events are rare (even if they have a large impact) or because they are not frequently consumed by the general population but rather in specific risk groups. To assess these differences, comparing outbreak data-driven estimates with source attribution estimates obtained with analysis of data from sporadic infections is paramount.

Analysis of the data from case-control studies of sporadic infection shows that the most important source of STEC globally was beef. Beef was also a significant risk factor in the Americas and Europe, but not in the Western Pacific region, where chicken was the most significant risk factor. These findings for beef and chicken

were not significantly moderated by the raw or cooked status of the food item, nor by the publication year of the study. Some food items (e.g. produce (fruits and vegetables), dairy, eggs, poultry/game–unspecified) were significant but had ORs of less than one. For the purpose of source attribution, conclusions are not drawn for factors associated with a statistically significant reduced risk of disease. Reasons for this include the impact of bias inherent in individual case-control studies, and thus in the final meta-analysis. While this is true for all exposures and all data that originate from interviews with patients and controls, it is particularly important when making inferences on the protective effect of specific exposures, which may eventually also be routes for infection (Domingues *et al.*, 2012a, 2012b). Thus, any ORs of less than one are reported herein but are not interpreted further.

The applied source attribution methods attribute illness at the point of exposure/consumption, and do not address the point in the farm to fork continuum where contamination occurred or was amplified. Other source attribution methods attribute illnesses at the point of origin of the pathogen and/or investigate different transmission routes from the same origin (Pires *et al.*, 2009). Even though we acknowledge the advantages of such methods to estimate the relative importance of sources and exposure routes for foodborne infections, we concluded that attributing STEC illnesses at regional and global level was feasible by applying point of exposure methods.

5.2 COMPARISON WITH FERG ESTIMATES

FERG's expert elicitation was conducted to address knowledge gaps at that time and provide evidence on the relative contribution of specific foods to the burden of sporadic STEC infections at global and regional level. While expert elicitations should not replace use of "hard" data, they are useful where such data are unavailable or have significant limitations (Hoffmann *et al.*, 2017). In these situations, studies have conventionally relied on the judgments of study authors or modelers, whose uncertainty judgments may reflect specific experience or specialty bias. Formal structured elicitation of judgments from a panel of multiple experts provides a systematic, transparent and auditable alternative.

The data-driven source attribution estimates presented are based on data from outbreak surveillance and a systematic review of case-control studies. Both are epidemiological methods that build on public health surveillance data to attribute illness at the point of exposure.

In general, the results presented here and the estimates of the expert elicitation conducted by FERG were largely in agreement (Hoffmann *et al.*, 2017). Differences

between outbreak and expert elicitation estimates could be explained by the fact that the expert elicitation was not limited to outbreaks (i.e. experts were asked to estimate attribution proportions for all cases, sporadic and outbreak-associated), and because limited evidence on the relative contribution of different sources for STEC illness was available to inform the experts' estimations.

5.3 DATA LIMITATIONS AND THEIR IMPACTS

The results presented here are subject to several limitations which must be considered when interpreting these findings.

5.3.1 Lack of data

It is important to note that, with data-driven approaches such as those used here, the quality of the outcome depends on the availability and quality of the data. Neither outbreak data nor case-control studies were found from three WHO regions: African, South-East Asian, and Eastern Mediterranean. Whether the results presented here are relevant for these regions is unclear.

Although foodborne outbreaks receive media and political attention, the greater burden of foodborne diseases consists of sporadic cases. Thus far, few countries have implemented surveillance of sporadic cases of foodborne disease and so the majority of reported human cases are associated with foodborne outbreaks. In general, outbreak data have the advantage of being widely available worldwide, including in countries or regions where sporadic cases of disease are not likely to be reported. However, for both the outbreak data and the case-control studies, the data obtained were limited, and biased towards high-income countries. As outbreak investigation and surveillance capacity across the world increases, source attribution of STEC at global, regional and local levels will improve.

In the outbreak analysis, to investigate the relative contribution of different sources to severe cases of disease, we restricted the analysis to outbreaks leading to cases of HUS or to deaths. Due to limited data availability, these analyses were restricted to the AMR. No substantial differences were identified in the attribution proportions for milder cases, HUS cases or deaths.

In addition to a limited number of available case-control studies, it is important to note that the way in which case-control studies are conducted and have traditionally been reported also influences the type of data available. First, although case-control questionnaires about enteric illnesses like STEC often include an extensive list of potential risk factors (e.g. a detailed list of food items), the specific food items included are often those for which there is an established or suspected as-

sociation with illness, so as not to overburden study participants. Second, of the extensive list of food items for which data are collected, many may not have results reported in the final publication or they may be reported in aggregate. For example, in one study, the produce (fruits and vegetables) items in the questionnaire were lettuce, pre-cut lettuce, raw vegetables, sprouts and self-produced fruit juice, with numerical results reported in the publication for raw vegetables (which may have included lettuce and sprouts) and self-produced fruit juice. Finally, publication culture prioritizes reporting significant over non-significant results (both at the study level and at the food level within a given study). Here, significant findings were more often reported with extractable numbers, whereas in some cases non-significant findings were reported as text without numeric values (e.g. "eating ground beef was not associated with infection in this study" in O'Brien *et al.*, 2001), meaning they could not be included in the analyses. To address these limitations, publication bias was assessed, and a back-calculation to make use of all raw data was conducted. Nevertheless, the results of the case-control study analysis are likely skewed towards those food items for which there was established evidence or strong hypothesis of risk at the time of the study, as well as results for which there were statistically significant findings.

5.3.2 Temporal changes

Data collected covered a broad time period (outbreak data: 1998-2017; case-control studies: 1985-2012), but these analyses did not account for possible temporal changes in factors such as pathogen incidence, outbreak surveillance, regulations and interventions, and illness attribution. As these factors and food preferences change over time, these estimates may also change. The association of specific food categories with STEC illness reflects the historical practices of food production, distribution and consumption. Changes in production, distribution and consumption may result in changes in STEC exposure. Consequently, microbial risk management should be informed by an awareness of current local sources of STEC exposure.

Our study did not adjust for older data (i.e. discounting or reducing the weight of older outbreaks) as other studies have done (IFSAC, 2015) because data were sparse and discounting data would lead to a further reduction of available data.

5.3.3 Categorization of food items

These results highlight the food categories to which a large proportion of STEC illnesses at the global level can be attributed. However, it is important to note that these analyses used broad food categories, and that the results do not suggest that all food items within these large categories are frequent sources of STEC. As an example, "produce" includes a wide range of vegetable products, and STEC outbreaks have frequently been linked to a few food items within this category (e.g.

lettuce, spinach). Still, the limited number of categories identified as important suggests that interventions for STEC focusing on these areas may be most effective in reducing illnesses (IFSAC, 2015).

5.3.4 STEC serogroups

Significantly more information was available for STEC belonging to serogroup O157 than for other STEC serogroups. A limitation of the analyses is that findings from the many outbreaks and case-control studies covering only O157 were assumed to apply to STEC in general.

5.4 CONCLUSIONS

Data from case-control studies of sporadic infections and outbreak investigations offer different types of information about the sources of human illness. Here, beef, produce (fruits and vegetables), dairy products, other unspecified types of meat, and chicken all emerged as significant sources of STEC, depending on geographic region (AMR, EUR and WPR) and whether the illnesses being considered were from outbreaks or were sporadic cases. Care must be taken in extrapolating data from these regions to other regions for which there are no data. Similarly, absence of data for food categories, and absence of food items from any of the studies, does not necessarily mean that said food items are safe.

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Annexes

Annex 1

Total number of STEC outbreaks reported per country and WHO region*

Country	Region	Total
Argentina	AMR	18
Australia	WPR	23
Austria	EUR	8
Belgium	EUR	10
Canada	AMR	54
Croatia	EUR	2
Denmark	EUR	9
Finland	EUR	2
France	EUR	59
Germany	EUR	9
Hong Kong	WPR	3
Hungary	EUR	1
Ireland	EUR	10
Japan	WPR	6
Luxembourg	EUR	1
Malta	EUR	1
Netherlands	EUR	4
New Zealand	WPR	3
Norway	EUR	3
Poland	EUR	4
Portugal	EUR	2
Romania	EUR	1
Slovakia	EUR	1
Spain	EUR	6
Sweden	EUR	13
United Kingdom	EUR	30
United States	AMR	674
Total		957

*AMR: Region of the Americas; EUR: European Region; WPR: Western Pacific Region.

Annex 2

Comprehensive list of STEC serogroups*

O1	O32	O70	O107	O134	O168
O2	O36	O71	O108	O135	O169
O3	O37	O73	O109	O136	O169-O183
O4	O38	O74	O110	O137	O171
O5	O39	O75	O111	O138	O172
O6	O40	O76	O112	O139	O173
O7	O41	O77	O112ab	O141	O174
O8	O42	O78	O113	O142	O175
O9	O43	O79	O114	O143	O176
O9ab	O44	O80	O115	O145	O177
O10	O45	O81	O116	O146	O178
O11	O46	O82	O117	O147	O179
O12	O48	O83	O118	O148	O180
O13	O49	O84	O119	O149	O181
O14	O50	O86	O120	O150	O182
O15	O51	O87	O121	O151	O183
O16	O52	O88	O123	O152	O185
O17	O54	O89	O123-O186	O153	O186
O18	O55	O90	O124	O153-O178	O187
O19	O57	O91	O125	O154	O188
O20	O58	O92	O125ac	O156	O189
O21	O59	O93	O126	O157	OgC4-O118-O151
O22	O60	O96	O127	O158	OX3
O23	O61	O98	O128	O159	OX7
O24	O62	O100	O128ab	O160	OX177
O25	O63	O101	O128ac	O161	OX178
O26	O64	O102	O129	O162	O-Dys1
O27	O65	O103	O130	O163	O-Rough
O28	O66	O104	O131	O164	O-Untypeable
O29	O68	O105	O132	O165	
O30	O69	O106	O133	O166	

*STEC serogroups that have been associated with human infection, identified from the following references and in consultation with the Joint FAO/WHO Expert Group on Shiga toxin-producing *Escherichia coli* (serogroups "O", "OR", "ON", "OUT" were identified as associated with human infection, but were excluded from the list of search terms because they are also stand-alone words that returned substantial numbers of irrelevant results.)

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Annex 3

Results of the meta-analysis using the alternate method

(i.e. using the reported univariate OR for all instances where such values were reported, and then using the reported number of cases and controls who were either exposed or unexposed to a given food category for instances where ORs were not given).

Alternate method results for Table 6: Pooled univariate odds ratios (ORs) per food category (significant values shown in **bold**), ranked in descending order by the number of food items in the category.

Food Category (no. items within category)	Odds ratio (95% UI)	p-value	p-value Regression test	p-value Rank test	Trim-and-Fill Method – Odds ratio (95% UI)	p-value
Beef (80)	**1.650 (1.399, 1.947)**	**<0.001**	**<0.001**	**0.023**	**1.436 (1.212, 1.701)**	**<0.001**
Meat – unspecified (60)	**1.194 (1.020, 1.397)**	**0.027**	**0.001**	**0.029**	1.017 (0.867, 1.192)	0.904
Produce (fruits and vegetables) (38)	**0.645 (0.514, 0.810)**	**<0.001**	**0.024**	0.083	**0.618 (0.490, 0.780)**	**<0.001**
Dairy (23)	**0.719 (0.548, 0.942)**	**0.017**	0.202	0.497		
Chicken (9)	0.795 (0.356, 1.775)	0.576	0.681	0.612		
Seafood (8)	0.700 (0.467, 1.050)	0.084	0.254	0.905		
Pork (7)	1.036 (0.625, 1.716)	0.892	0.297	0.239		
Eggs (5)	**0.646 (0.491, 0.851)**	**0.002**	0.852	0.817		
Lamb (3)	1.899 (0.570, 6.330)	0.296	0.083	0.333		
Turkey (2)	1.038 (0.112, 9.590)	0.974	N/A	N/A		
Poultry/Game unspecified (2)	**0.386 (0.179, 0.834)**	**0.015**	N/A	N/A		

* This number is lower than in Table 5 because some food items as reported did not have sufficient useable data

(cont.)

Alternate method results for Table 7: Results for each World Health Organization (WHO) sub-region, showing pooled univariate odds ratios (ORs) per food category (significant values shown in **bold**).

Food Category	WHO sub-region AMR A1		WHO sub-region AMR B2		WHO sub-region EUR A3		WHO sub-region WPR A4	
	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)	No. items per category	OR (95% UI)
Beef	22	1.420 (0.993, 2.031) †	32	**1.573 (1.199, 2.063)** †	19	**1.374 (1.018, 1.855)**	7	1.303 (0.782, 2.170)
Meat unspecified	9	1.167 (0.861, 1.583)	8	**0.461 (0.369, 0.576)** †	38	**1.207 (1.045, 1.394)** †	5	1.268 (0.846, 1.900)
Produce (fruits and vegetables)	9	**0.515 (0.356, 0.745)**	0	N/A	17	0.938 (0.713, 1.233) †	12	0.617 (0.310, 1.227) †
Dairy	1	9.774 (0.981, 97.360)	0	N/A	20	**0.653 (0.498, 0.855)**	2	1.221 (0.696, 2.141)
Chicken	4	**0.354 (0.234, 0.536)**	0	N/A	2	1.146 (0.104, 12.606)	3	**2.677 (1.264, 5.671)**
Seafood	2	0.683 (0.417, 1.118)	0	N/A	5	0.688 (0.227, 2.085)	1	**0.480 (0.309, 0.747)**
Pork	2	1.469 (0.863, 2.500)	2	1.107 (0.320, 3.830)	0	N/A	3	0.685 (0.361, 1.301) †
Eggs	0	N/A	0	N/A	1	**0.600 (0.398, 0.905)**	4	**0.686 (0.474, 0.993)**
Lamb	0	N/A	0	N/A	3	1.899 (0.570, 6.330)	0	N/A
Turkey	1	0.400 (0.088, 1.815)	0	N/A	0	N/A	1	4.000 (0.361, 44.277)
Poultry/ Game unspecified	0	N/A	0	N/A	2	**0.386 (0.179, 0.834)**	0	N/A

[1] AMR A includes the following countries: Canada; Cuba; United States of America.
[2] AMR B includes the following countries: Antigua and Barbuda; Argentina; Bahamas; Barbados; Belize; Brazil; Chile; Colombia; Costa Rica; Dominica; Dominican Republic; El Salvador; Grenada; Guyana; Honduras; Jamaica; Mexico; Panama; Paraguay; Saint Kitts and Nevis; Saint Lucia; Saint Vincent and the Grenadines; Suriname; Trinidad and Tobago; Uruguay; Venezuela (Bolivarian Republic of).
[3] EUR A includes the following countries: Andorra; Austria; Belgium; Croatia; Cyprus; Czech Republic; Denmark; Finland; France; Germany; Greece; Iceland; Ireland; Israel; Italy; Luxembourg; Malta; Monaco; Netherlands; Norway; Portugal; San Marino; Slovenia; Spain; Sweden; Switzerland; United Kingdom.
[4] WPR A includes the following countries: Australia; Brunei Darussalam; Japan; New Zealand; Singapore.

† Using trim-and-fill method

Annex 4

Results of the analysis of univariate moderating effects of study characteristics, using the alternate method

(i.e. using the reported univariate OR for all instances where such values were reported, and then using the reported number of cases and controls who were either exposed or unexposed to a given food category for instances where ORs were not given).

Alternate method results for Table 8: Univariate moderating effects of study characteristics, by food category (n.b. turkey and poultry/game-unspecified excluded because n=2), with significant values shown in **bold**.

Study Characteristic	Characteristic level	OR	95% C.I.	p-value
Beef (n=80)				
WHO subregion	AMR A (reference)	—	—	—
	AMR B	1.135	0.752, 1.713	0.546
	EUR A	0.820	0.514, 1.308	0.405
	WPR A	0.758	0.391, 1.470	0.412
Publication year		1.021	0.997, 1.045	0.084
Study population age	All (reference)	—	—	—
	Adults	1.295	0.692, 2.424	0.419
	Children	1.364	0.969, 1.921	0.075
Food item status	Not raw (reference)	—	—	—
	Raw or undercooked	2.372	0.961, 5.855	0.061
	Unknown	1.011	0.412, 2.481	0.982
Robin's I	1 (reference)	—	—	—
	2	1.451	0.947, 2.222	0.087
	4	1.036	0.474, 2.268	0.929

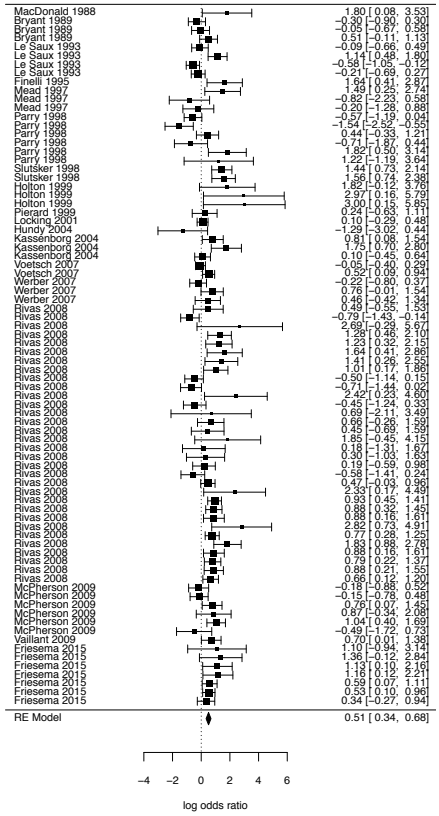
(cont.)

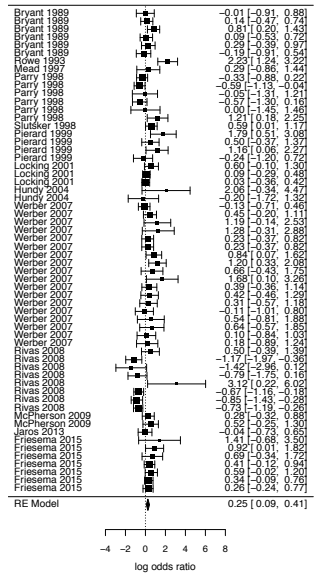
	6	4.193	0.769, 22.868	0.098
Believeable findings	Yes (reference)	—	—	—
	No	3.155	0.596, 16.700	0.177
Meat–unspecified (n=64)				
WHO subregion	AMR A (reference)	—	—	—
	AMR B	**0.333**	**0.233, 0.475**	**<0.001**
	EUR A	0.961	0.717, 1.289	0.792
	WPR A	0.915	0.556, 1.505	0.727
Publication year		1.001	0.980, 1.021	0.952
Study population age	All (reference)	—	—	—
	Adults	1.410	0.949, 2.094	0.089
	Children	0.795	0.575, 1.101	0.167
Food item status	Not raw (reference)	—	—	—
	Raw or undercooked	1.501	0.909, 2.479	0.112
	Unknown	1.157	0.761, 1.760	0.495
Robin's I	1 (reference)	—	—	—
	2	0.781	0.553, 1.103	0.160
	4	0.929	0.561, 1.537	0.773
	6	N/A	N/A	N/A
Believeable findings	Yes (reference)	—	—	—
	No	1.683	0.336, 8.432	0.526
Produce (fruits and vegetables) (n=38)				
WHO subregion	AMR A (reference)	—	—	—
	AMR B	N/A	N/A	N/A
	EUR A	1.636	0.972, 2.755	0.064
	WPR A	1.050	0.598, 1.844	0.864
Publication year		0.978	0.941, 1.017	0.270
Study population age	All (reference)	—	—	—

(cont.)

	Adults	1.040	0.491, 2.200	0.919
	Children	1.117	0.608, 2.054	0.721
Food item status	Not raw	N/A	N/A	N/A
	Raw or undercooked (reference)	—	—	—
	Unknown	1.065	0.669, 1.694	0.792
Robin's I	1 (reference)	—	—	—
	2	0.778	0.488, 1.238	0.289
	4	N/A	N/A	N/A
	6	N/A	N/A	N/A
Believeable findings	Yes (reference)	—	—	—
	No	N/A	N/A	N/A
Dairy (n=23)				
WHO subregion	AMR A (reference)	—	—	—
	AMR B	N/A	N/A	N/A
	EUR A	0.067	0.006, 0.752	0.028
	WPR A	0.126	0.010, 1.583	0.109
Publication year		0.958	0.911, 1.007	0.089
Study population age	All (reference)	—	—	—
	Adults	0.937	0.395, 2.224	0.883
	Children	0.610	0.354, 1.053	0.076
Food item status	Not raw (reference)	—	—	—
	Raw or undercooked	1.116	0.455, 2.736	0.810
	Unknown	0.531	0.234, 1.205	0.130
Robin's I	1 (reference)	—	—	—
	2	0.637	0.351, 1.157	0.138
	4	10.326	0.827, 128.858	0.070
	6	N/A	N/A	N/A
Believeable findings	Yes (reference)	—	—	—
	No	**14.054**	**1.210, 163.205**	**0.035**

Alternate methods results for Figure 6: Forest plots of the log odds ratio (OR) of the risk of human STEC infection from beef (left) and meat-unspecified (right), showing the overall pooled OR together with the 95% UI.





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Microbiological Risk Assessment Series

Shiga toxin-producing Escherichia coli (STEC) infections are a substantial public health issue worldwide. Circa 2010, foodborne STEC caused more than 1 million illnesses, 128 deaths and ~ 13 000 Disability-Adjusted Life Years (DALYs).

To appropriately target interventions to prevent STEC infections transmitted through food, it is important to determine the specific types of foods leading to these illnesses. This report conducted an analysis of data from STEC foodborne outbreak investigations reported globally, and a systematic review and meta-analysis of case-control studies of sporadic STEC infections published for all dates and locations.

This work was undertaken in response to a request from the Codex Alimentarius Commission to support the development of the international standards on foodborne STEC. The advice herein is useful for both risk assessors and risk managers, at national and international levels and those in the food industry working to control this hazard.

For further information on the Joint FAO/WHO activities on microbiological risk assessment and related areas, please contact

Food Safety and Quality Unit
Agriculture and Consumer Protection Department
Food and Agriculture Organization of the United Nations
Viale delle Terme di Caracalla
00153 Rome, Italy
Email: jemra@fao.org
Website: http://www.fao.org/food/food-safety-quality

Department of Food Safety and Zoonoses
World Health Organization
20 Avenue Appia
1211 Geneva 27, Switzerland
Email: jemra@who.int
Website: www.who.int/foodsafety

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