Requirements for Investigations of Animal Food Chains

Fries\(^a\), Reinhard, Karl-Hans Zessin\(^b\), Moses Kyule\(^b\) and Maximilian Baumann\(^b\)

\(^a\) Freie Universität Berlin, FB Veterinärmedizin, Institut für Fleischhygiene und –technologie, Brümmerstr. 10, 14195 Berlin, Germany. Email: fries.reinhard@vetmed.fu-berlin.de
\(^b\) Freie Universität Berlin, FB Veterinärmedizin, FR International Animal Health, Luisenstr. 56, 10117 Berlin, Germany

Abstract

Concerns regarding the safety of foods of animal origin have increased in recent years as a consequence of ever-emerging hazards associated with food production and supply chains. These hazards cause various risk-threats to human health. Concerns are being spurred on by requirements related to the globalization of trade of agricultural goods and by perceptions of consumers regarding food safety. International mandated organizations like the Codex Alimentarius Commission and trade blocks like the European Union responding to the concerns in consequence have embarked on an integrated approach to assure a high level of food safety and quality from farm to table covering all sectors of the food chain, including feed production, primary production, food processing, storage, transport and retail sale.

Investigations of food chains and determination of risk factors for the transmission pathways of agents along the chain require a new research approach: the pre-harvest stage has to be added and integrated into investigations which traditionally have focused on the harvest and the post-harvest stages. Achievement of this requires high quality data on the various multiple stages of the food chain which in turn calls for the involvement of multi-disciplinary teams and multidimensional research approaches. Data would be used in the development and validation of state-transition risk assessment models that would provide insight in epidemiological consequences of food hazards and the efficiency of safety measures.

The paper presents experiences with the design and implementation of a food chain investigation approach for \textit{Salmonella} infection in the pork chain in Northern Thailand. Marked animals and samples from them were followed throughout the chain and \textit{Salmonella} prevalences were investigated in respective stages during the production line, transportation, lairage, slaughter and in pork products in retail markets. Investigation problems encountered and solutions used at different stages are presented.

1. Introduction and objectives

Recent changes in regulations governing safety and quality of global food trade of animal origins have created high demands for designing epidemiologically valid studies along the supply chains. To achieve this, internationally mandated organisations, for example WHO and the FAO/OIE, have recommended a multidimensional research approach and the involvement of a multi-disciplinary team in designing such surveys for the various segments of a supply chain. For example, the involvement of different disciplines such as agriculturalists, veterinarians,
hygienists, epidemiologists, economists, statisticians, laboratory technicians, and specialists in other food fields is strongly recommended. Results of food chain investigations aim at providing total transparency for a food commodity. For corrective measures, forward and backward tracing of results of hazards at the various stages in the supply chain is aimed at. Forward and backward tracing results are the basis for quantitative risk assessment and subsequently risk management. Here, various aspects of an investigation for the occurrence of Salmonella in a pork chain in a northern province in Thailand are presented. Particulars of this pork investigation, which centers on a laboratory-oriented planner’s point of view, can be used as examples for analyses of other chains.

2. Experiences and practices in food chain philosophy

2.1. Overall planning and coordinating

Due to globalization of food production, manufacturing and marketing food chain supplies have become very complex at national, regional and international levels. Thus, need exists for supply planning, implementing and controlling the flow and storage of raw materials, in-process inventory of finished goods as well as related information from the point of origin of animals to the point of consumption. Therefore, a team rather than a single person is needed for a chain analysis. The team must be coordinated by "one responsible person" who coordinates time schedules, sample collections, data organisation, and communicates progress/problems of the total study programme among the different groups/laboratories involved.

2.2. Biological sampling

In our example, representative samples of Salmonella from farm-level "almost" to retail markets yielded significant results (Table 1).

Table 1: Salmonellae in a Pork Chain in Northern Thailand (positive samples in %)

<table>
<thead>
<tr>
<th></th>
<th>Farm level (animals)</th>
<th>Abattoir-level (carcasses)</th>
<th>Retail-level (pork products)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feces</td>
<td>63 %</td>
<td>83 %</td>
<td>83 %</td>
</tr>
<tr>
<td>Lymphnodes</td>
<td>64 %</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Swabs prior to chilling</td>
<td>33 %</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Swabs after chilling</td>
<td>13 %</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pork during cutting</td>
<td></td>
<td>56%</td>
<td>Pork products: 71 %</td>
</tr>
</tbody>
</table>

Since most food safety risks stem from microbiological agents, pre-lab, in-lab and post-lab phases must be distinguished properly regarding such hazards.

Pre-laboratory phase (sampling in the field)

- **Targets:** Basically, most important zoonotic agents such as Salmonella or Campylobacter are widespread. If the focus is on the occurrence of the total spectrum of zoonotic agents, the whole line including different agents would be included. If the focus is on the hygiene of a particular chain sequence, only marker organisms such as Enterobacteriaceae or the aerobic plate count are suitable targets.

- **Sampling site:** Each site (“white” or “black” zone) in the farm, in the abattoir or in retail has a "different but individual" microbiological status: If one looks for the special
sequence of one machinery or for the particular impact of one particular circuit (e.g. water supply), only this particular part would be investigated.

- **Number and type of samples**: Prepare for sufficient laboratory capacity for working on the samples properly. For salmonellae, tests are mostly done on the basis of presence/absence results. But, a quantitative approach may be needed, e.g. cfu, etc., for risk assessment.

- **The age of animals defines the time of sampling during their life and the time of the day during the slaughter procedures**: From poultry it is known, that salmonellae occurrence depends on age; in the slaughter line the scalding, dehairing and evisceration stages are frequently salmonellae-positive.

- **Type of specimens**: Some specimens are more likely to carry the agent than others. In a survey in a pig processing line, all samples from the back of carcasses were salmonellae-negative; sampling feces, muscles or skin may thus yield different positivity rates.

- **Specimen size** (eg. 5 g, 25 g): The outcome of an analysis for *Salmonella* is strongly dependent on sample volume. The more material for analysis, the higher the probability to detect the agent.

- **Type of sampling** (destructive, conservative): Sometimes it is not possible to cut materials from a carcass due to financial/trade reasons. However, results from rinsing, scratching or cutting an area are known to yield quite different results.

**In-laboratory phase (“bacteriological technique” in a strict sense)**

- Preparation of specimens such as “pooling”: To save money, time and lab personnel, several samples may be pooled to be analysed in one single procedure. Cave: Enough materials of the same identity must be saved in order to repeat the analysis for each piece of specimen, should the result from the pooled sample turn out to be positive.

- The technique of the bacteriological analysis itself (non-selective, selective, molecularbiological): For the detection of a particular agent, selective and internationally accepted procedures such as ISO standards are to be followed. If the general ecological composition of a particular location or stage is to be observed, non-selective, blood containing media for culturing most bacteria should be used. In any case, Standard Operating Procedures (SOPs) of laboratories are to be followed.

**Post-laboratory phase**

- Differentiate between (a) direct lab results (internal log book) and (b) lab-based calculations which are the final results of laboratory work. Further calculations (c) based on such a dataset are not under the responsibility of the lab (see 2.3).

- Enter and analyse data in a beforehand customised data management system.

- If the objective is the implementation of a *Salmonella* surveillance systems, information from different lab techniques (ELISA, conventional isolation or PCR) deserves special attention in subsequent interpretation and comparison.

2.3. **Epidemiological sampling considerations: Interpretation of the dataset and consequences**

Data obtained from microbiological procedures are to be processed and interpreted in a different way to those collected through field observations. For one “chain approach” it may be sufficient to aim at descriptive information such as number or isolates, serotypes or management practices at a farm (e.g. feeding systems, all in-all out procedure). Another approach may aim at associations between chemical or bacteriological results and farm and management parameters.
2.4. The data set proper
Apart from biological sampling, observational or interview data may be collected. Questionnaires, if applied, deserve careful design and prior validation. It also may be useful or even necessary to obtain data from other “foreign” sources such as administrative records (meat inspection results, results from Salmonella surveillance systems). The use of such data raises questions on their validity and on their incorporation into data sets (cave: data compatibility).

2.5. The personal factor
At every stage of a study (sampling, lab, information from other, marginally involved persons), the reliability and quality of data obtained depends strongly on the human factor. Generally, if human beings are involved, bias never can be excluded, and not every information must be taken for granted.

- Farmers may know very well their own farming practices and flaws, but may be reluctant to let others know. In questionnaires the relevance and phrasing of questions (risk factors yes/no) has to be given particular attention.
- Because of the large number of persons involved in meat inspection services, the personal factor is crucial in regards to the validity of results. Although meat inspectors in general should have been instructed and trained beforehand, differences in interpretation of lesions cannot be excluded. So, performance of meat inspection remains subjective.

3. Major characteristics of the Thailand pork chain investigation
As learned from experience of an animal food chain investigation, general problem areas for safeguarding the quantity, quality and continuity of data at each stage of the chain include:

- Difficulties to access areas for sampling due to reluctance/non-cooperation of persons in charge
- Difficulties to identify the appropriate sampling sites along the chain (resulting in meaninglessly taken data)
- Laboratory failures (samples not taken, samples spoiled, media wrongly prepared)
- Use of different labs with different experience and sometimes even using different test media

Exemplary challenges experienced in the Thailand pork chain investigation and solutions found are summarised as follows:

Farm level:
Difficulties (Examples)
- Initial reluctance by company owner(s) to permit the study
- Accessibility to some of the pig farms
- Tagging/marking of sample pigs: No ear tags allowed by farm owners

Solutions:
- Meeting the company owners and extensive discussions on the study objectives
- Accessibility: The principal investigator did accompany the sampling personnel during their checks of the farms
- Spraying of identification numbers on the pig’s backs
Slaughterhouse level:
Difficulty:
- Identification of animals entering the abattoir
Solutions:
- Close cooperation between farm-level and slaughterhouse-level investigators in order to take over identified pigs unmistakenly
- At arrival: careful and repeated communication with slaughterhouse handling personnel prior to and during pig arrival

Slaughterhouse cutting room level:
Difficulty:
- Problems to keep track of all cut pieces and relate them to "same" carcass
Solution:
- Preservation of the marked carcass in order to sample cut pieces from the same carcass(es)

Retail-market level:
Difficulty:
- Follow-up of cut pieces from the marked carcass at the cutting room to the retail-markets because all cut pieces were transported in bulk to shops
Solution:
- Rather than sampling from the same cuts, samples were taken from the same batch of carcasses from the same day of slaughter

Laboratory-level:
Difficulties:
- Preservation of the samples and numbering
- Laboratory facilities, consumables and personnel (expertise)
- Data management and analysis: facilities and personnel (expertise)
- Facilities: Availability of computer software
- Personnel: Availability of laboratory personnel on a need-basis
Solutions:
- Adjustment of sampling design and laboratory schedules according to available technological structure and standard
- Use of customised standard test reagents from outside the country, e.g. Europe
- External technical support of laboratory expert and data analyst