CHAPTER II

LITERATURE REVIEW

2.1 Microbiology of *Salmonella*

2.1.1 Genus *Salmonella*

Salmonellae is gram-negative motile bacteria. The genus *Salmonella* belongs to the family Enterobacteriaceae, it was named after Daniel E. Salmon, an American veterinarian who was the first to isolate *Salmonella* Choleraesuis from pigs with hog cholera in 1885 (Hagan et al., 1966).

Salmonella genera are usually motile by peritrichous flagella. But *S.* Gallinarum or *S.* Pullorum is always non-motile (Krieg, 1984). *Salmonella*e produce hydrogen sulfide except S. Choleraesuis and most strains of S. Paratyphi A (ISO-6579, 2002). The bacterium is facultative anaerobic and grows optimally at 37 °C. *Salmonella are* oxidase negative, catalase positive, indole and Voges-Proskauer negative, but methyl red and Simmons citrate positive (Holt, 2002).

Salmonellae live in the intestinal tract of a wide range of both, warm and cold-blooded animals, they usually can adapt to a wide host range. Some are specifically adapted to a particular host such as *Salmonella* Typhi to primates, *Salmonella* Dublin to cattle, *Salmonella* Choleraesuis to pigs (WHO, 2005).

Morphologically, a *Salmonella* bacterium is a straight rod of 0.7-1.5 µm in width and 2-5 µm in length (Holt, 2002). There are three common compartments of *Salmonellae*. The first compartment is cytosol, in which the processes of genetic replication and protein expression occur. The second compartment is a cell envelope, containing a cell wall and cytoplasmic membranes critical to the structure and function of the pathogen. The other compartments are surface structures that lie externally to the cell envelope (e.g., capsules; O or K antigen, flagella; H antigen) (McClane, 1999, Murray, 2002).

The ***O antigen*** is a carbohydrate (also called a polysaccharide) that is the out-most component of lipopolysaccharide (LPS). It is a polymer of O subunits; each O subunit is composed of four to six sugars defining the O antigen. O antigens are designated by numbers and are divided into O serogroups, also called O groups (e.g., *S.* Typhimurium in group O:4 within Group B, *S.* Enteritidis in group O:9 within group D1; S. Paratyphi A in group O:2 within group A) (Anonymous, 2004b).

The ***H antigen*** is the filamentous portion of the bacterial flagella. The antigenically variable portion of flagella is the middle region of the protein, which is surface-exposed. *Salmonella* can express two different H antigens, which are encoded by two different genes. Expression of the two genes is coordinated so that only one flagellum antigen is expressed at a time in a single bacterial cell (McClane, 1999). The two distinct flagella antigens are referred to as Phase 1 and Phase 2. Most serotypes express phase 1 and phase 2 antigens (diphasic), while others might only express one (monophasic). Some serotypes do not express H-antigens and are non-motile.

Surface (envelope or capsular) Antigens in *Salmonella* may cover O antigens, so the bacteria may not be agglutinated with O antisera. The Vi antigen was recognized as one specific surface antigen. The Vi antigen appears in some *Salmonella* serotypes, e.g: *S.* Typhi, *S.* Paratyphi C, and rarely in S. Dublin (Todar, 2005).

2.1.2 *Salmonella* serotypes

When serological analysis was adopted into the Kauffmann-White scheme in 1946, a *Salmonella* species was defined as "a group of related fermentation phage-type" with the result that each *Salmonella* serotype was considered as a species

(Todar, 2005).

Nowadays, the genus *Salmonella* consists of only two species, *Salmonella* enterica and *Salmonella* bongori. S. enterica is divided into six subspecies: *S. enterica* subsp. *enterica, S. enterica* subsp. *salamae, S. enterica* subsp. *arizonae*, S. *enterica* subsp. *diarizonae*, *S. enterica* subsp. *houtenae* and *S. enterica* subsp. *indica*. This nomenclature reflects the present understanding of *Salmonella* taxonomy (Commission, 2005, Tindall, 2005). Serotypes belonging to *S. enterica* subsp. *enterica* are typically designated by a name frequently related to the geographical place where the serotype was first isolated. The serotype name is written in non-italicized Roman letters and the first letter is capitalized. Serotypes belonging to other subspecies are designated by their antigenic formulae, following the subspecies name. The antigenic formulae of *Salmonella* serotypes are listed in a document called the White-Kauffmann-Le Minor scheme (Grimont, 2007).

Few years before, the nomenclatural systems are based on recommendations from the WHO Collaborating Centre (Brenner, 2000). In 2010, M. Guibourdenche et al. reported the supplement report No.47 about characterization of 70 new *Salmonella* serotypes recognized between 2003 and 2007 by the WHO Collaborating Center for Reference and Research on *Salmonella*. The number of *Salmonella* species and *Salmonella* nomenclature is summarized in Table 1 (Guibourdenche et al., 2010).

Table 1 Present number of serotypes in each species and sub-species of *Salmonella*

Species and subspecies of *Salmonella*	**Number of serotypes**		
	1998	***2001***	***2010***
***S.* enterica**			
subsp. enterica	1454	1478	1547
subsp. salamae	489	498	513
subsp. arizonae	94	94	100
subsp. diarizonae	324	327	341
subsp. houtenae	70	71	73
subsp. indica	12	12	13
***S.*bongori**	20	21	23
Total	**2463**	**2501**	**2610**

Source (Guibourdenche et al., 2010)

2.1.3 Epidemiology of *Salmonella*

Salmonella is a zoonotic bacterium and can infect a wide range of host species, including poultry, cattle, pigs and humans. A bacterium is known to be zoonotic if it is naturally transmitted between vertebrate animals and humans. In particular, *Salmonella* bacteria can be transmitted to humans through direct contact with infected persons or animals or through indirect contact with products of animal origin (e.g. meat, eggs) that are contaminated with the *Salmonella* bacteria. The serotype S. Typhimurium is presently the predominant serotype isolated from pork during monitoring in the European Union (EFSA, 2006) and in Belgium (FASFC, 2007).

Salmonellae live in the intestinal tract of humans and other animals, including birds. Contaminated foods are often of animal origin, such as beef, poultry, milk, or eggs. Reptiles, such as turtles, lizards, and snakes, are particularly likely to harbour *Salmonella*. Many chicks and young birds carry *Salmonella* in their feces (CDC, 2012c).

Salmonella are known to be hardy and ubiquitous bacteria that multiply at 7-45ºC. Persistence of *Salmonella* in the environment for long periods (months or even years) is possible in the presence of suitable organic subtracts (Schwartz, 1999). *Salmonella* can be isolated from intestinal tract of mammals, reptiles, birds and insects as well as water, food or environment (Grimont, 2000).

2.2 Salmonellosis

2.2.1 Salmonellosis in humans

Human salmonellosis is caused by ubiquitous *Salmonella* serotypes (e.g., *S*.Typhimurium, *S*. Enteritidis). *Salmonella* may be associated with all kinds of food and other carriers. Contamination of meat (cattle, pigs, goats, chicken, etc.) may originate from animal salmonellosis, however most often it results from contamination with the intestinal contents during evisceration of animals, washing, and transportation of carcasses. Although handling of contaminated meat may result in contamination of hands, tables, kitchenware, towels, other foods, etc, vegetables and fruits may carry *Salmonella* due to contamination with fertilizers of fecal origin, or when it was washed with polluted water (Todar, 2005). Moreover, egg-associated salmonellosis is also an important human health concern in several countries. *Salmonella* Enteritidis can be inside of perfectly normal-appearing eggs, and if these eggs are eaten raw or undercooked, the bacterium can cause illness (Todar, 2005).

Human salmonellosis often occurs through ingestion of contaminated food or water, less commonly via direct contact to infected animals. Occupational risk for farmers, workers and veterinarians in contact with infected animals should not be disregarded, even though it seems to play a minor role (Baker, 2007, Hendriksen et al., 2004, Humphrey, 2000).

Many case studies, investigations of outbreaks showed that there have been association with a variety of foods including eggs, poultry, red meat but also other food stuffs such as chocolate, milk and milk products, salads, fruits and vegetables, fish and fishery products, etc (Bell, 2002, Barber, 2002, Hohmann, 2001, Hughes, 2007).

Most people infected with *Salmonella* develop diarrhea, fever, and abdominal cramps 12 to 72 hours after infection. The illness usually lasts 4 to 7 days, and most persons recover without treatment. However, in some persons, the diarrhea may be so severe that the patient needs to be hospitalized. In these patients, the *Salmonella* infection may spread from the intestines to the blood stream, and then to other body sites and can cause death unless the person is treated promptly with antibiotics. The elderly, infants, and those with impaired immune systems are more likely to have a severe illness (CDC, 2012c). The common symptoms of *Salmonella* infection are shown in Table 2.

Table 2 Symptoms of *Salmonella* infection in human

Symptom	**Case (%)**
Diarrhea	87
Abdominal pain	84
Feeling feverish	75
Nausea	65
Muscle pain	64
Vomiting	24
Headache	21
Blood in stools	6

Source: (Humphrey, 2000)

Foodborne *Salmonella* infection might result in gastro-intestinal illness. *Salmonella* infections are usually self-limiting. Severe and fatal illness is rare and mainly associated with impaired immune systems. However, in a minority of cases, non-typhoid salmonellosis might evolve to chronic disease resulting in localized infections and reactive arthritis and furthermore, in neurological and neuromuscular illnesses. In humans, a seasonal trend has been reported, with salmonellosis presenting a peak in summer months, which might be explained by higher temperatures and different consumption patterns (e.g. consumption of salads, barbecues) (Hald and Andersen, 2001, EFSA, 2008a).

2.2.2 Salmonellosis in animals

Salmonellosis commonly occurs in domestic animals, the consequences of infection range from sub-clinical carrier status to acute fatal septicemia. Some *Salmonella* serotypes such as *Salmonella* Pullorum in poultry, *Salmonella* Dublin in cattle and *Salmonella* Choleraesuis in pigs are relatively host-specific. In contrast, *Salmonella* Typhimurium has a comparatively wide host range. The *Salmonella* serotypes of importance in domestic animals and the consequences of infection are shown in Table 3.

Swine are considered important carriers. *Salmonella* Typhimurium can be found in the pharynx, tongue, tonsils, ileum, caecum, mandibular, ileocaecal and superficial inguinal lymph nodes, liver, stomach contents and faeces (Hurd et al., 2001, Swanenburg et al., 2001).

Up to 30% of pigs may shed *Salmonella* on the farm (Stärk, 2002). This percentage increases during transport and in lairage as there is increased stress of the pigs caused by crowding, and transport, leading to increased exposure of naive pigs (Barber, 2002, Kranker, 2003).

Table 3 *Salmonella* serotypes in humans and animals; clinical and other consequences of infection

***Salmonella* serotype**	**Host**	**Consequences of infection**
Salmonella Typhimurium	Many animal species Humans	Enterocolitis and septicemia Food poisoning
Salmonella Dublin	Cattle	Many disease conditions Enterocolitis and septicemia
Salmonella Choleraesuis	Pigs	Enterocolitis and septicemia
Salmonella Pullorum	Chicken	Pullorum disease (bacillary white diarrhoea)
Salmonella Gallinarum	Adult birds	Fowl typhoid
Salmonella Arizonae	Turkeys	Arizona or paracolon infection
Salmonella Enteritidis	Poultry Humans Other mammal species	Often sub-clinical in poultry Food poisoning Clinical diseases
Salmonella Brandenburg	Sheep	Abortion

Source: (Quinnand, 2003)

2.3 *Salmonella* prevalence and risk factors at farm, slaughterhouse and market

2.3.1 *Salmonella* prevalence in pigs and pork

2.3.1.1 Salmonella prevalence in pigs and pork overseas

Pork is considered to be one of the main sources of *Salmonella* infection in humans (van der Gaag et al., 2004, Berends et al., 1998). So, more studies on *Salmonella* in pig have been done. In the 1990's, swine-specific *Salmonella* studies increased more than five-fold compared to ten years ago (Bahnson, 2001).

Hald et al. used Danish estimation principles to quantify sources of human salmonellosis and assessed the role of pork in the transmission of *Salmonella* to humans in Denmark, the Netherlands, Germany, Sweden and England and Wales. Although *Salmonella* types can occur in almost all food-producing animals, there are often rather strong associations between certain types and a particular animal reservoir (Hald et al., 2003).

In Germany, the overall prevalence of *Salmonella*e was estimated 6.2% in the slaughtered pigs from Germany (Käsbohrer, 2000).

In Belgium, prevalence of *Salmonella* in pigs was 37% of the carcass samples as a mean value; there were high variations from different slaughterhouses between 0 and 70%. Distribution of S. Typhimurium predominated on pig carcasses with 71% (Botteldoorn et al., 2003).

In two large U.S. commercial pork processing plants, the overall prevalences of *Salmonella* on carcasses at three sampling points (prescald, preevisceration, and after chilling) were 91.2%, 19.1%, and 3.7%, respectively. The prevalence of carcasses with enumerable *Salmonella* at prescald, preevisceration, and after chilling were 37.7%, 4.8%, and 0.6%, respectively (Schmidt et al., 2012).

Kuhn et al. (2012) showed that the outbreak strain from the slaughterhouse environment and in pork and products as well as patient interviews strongly suggested different pork products as the source of infection (Kuhn et al., 2012).

In the Netherlands, contamination at the slaughter-line is initiated by pigs carrying *Salmonella* on skin or in faeces, another contamination route could be resident flora present on the slaughter equipment. On 96.6% of the skin samples *Salmonella* was identified, whereas a lower number of animals tested positive in their rectum (62.5%) (van Hoek et al., 2012).

In 2008, in Ireland, *Salmonella* was detected on 24/720 (3.3%) pork cuts and in 7/56 (12.5%) of environmental swabs. The predominant serotype was *Salmonella* serotype Typhimurium followed by *Salmonella* Derby (Prendergast et al., 2008). Study on cross-contamination within the slaughter plant environment in slaughter process in Ireland accounted for up to 69 % of *Salmonella* carcass contamination (Duggan et al., 2010).

In small Wisconsin abattoirs in U.S, a study on processing conditions and indicator bacteria levels showed the *Salmonella* prevalences on skinned and unskinned prewash carcasses were 11.7 and 8.3%, respectively (Algino et al., 2009).

In Thailand, there were 26 (65%) pork samples and 30 (75%) chicken meat samples contaminated with *Salmonella*. The most prevalent serotype in pork was S. Rissen (61.5%), followed by S. Stanley and S. Lexington (11.5%) (Angkititrakul et al., 2005). Prevalence of *Salmonella* on pigs in farms, slaughterhouses and pork in market were 6%, 28% and 29%, respectively (Padungtod and Kaneene, 2006). In Chiang Mai (Thailand), the prevalence of *Salmonella* in pre-slaughterhouse pig was 69,5% (Patchanee, 2002) and 55.5% of freshly cut pork, 70.5% of transported pork, and 34.5% of retail products. The five most prevalent *Salmonella* serotypes identified were Rissen (45.3%), Typhimurium (16.3%), Krefeld (10.6%), Stanley (6.3%), and Lagos (6.0%) (Sanguankiat et al., 2010).

2.3.1.2 Salmonella prevalence in pigs and pork in Vietnam

Salmonella prevalence in domestic animals in 6 provinces of the Mekong Delta, in 2000, was 5.2% in pigs and 69.9% of the pork samples. Eighty *Salmonella* strains were isolated and 25 serotypes were identified. The predominant serotypes were S. Javiana, S. Derby, and S. Weltevreden. S. Javiana and S. Weltevreden were detected together in pigs, chickens, and ducks (Tran et al., 2004, Phan et al., 2005a). Overall *Salmonella* in pigs was 49.4% in the Mekong Delta, Vietnam (Vo et al., 2006b).

A study examined the level of *Salmonella* contamination in raw food samples and determined their antibiotic resistance in Hanoi. There was 61% of meat (chicken, beef, pork) and 18% of shellfish samples were contaminated with *Salmonella* (Ha and Pham, 2006).

A study on prevalence and epidemiology of *Salmonella* in small pig abattoirs in Hanoi showed that the *Salmonella* prevalence from cecal, carcass swabs, and tank water samples was 52.1%, 95.7% and 62,5% respectively (Le Bas et al., 2006).

In Hue, an epidemiological analysis an detection for *Salmonella* spp. in specimens collected from pork production chains reported 7 serotypes of *Salmonella* that were detected in retail pork, slaughterhouse carcasses and environmental specimens. The following detection rates were 32.8% of retail pork, 15.5% of slaughterhouse carcasses, 47.4% of floors, 38.1% of weighing bowls, 28.6% of cooking boards and 16.7% of tank water samples (Takeshi et al., 2009).

In North Vietnam, the prevalence of *Salmonella* was approximately 39.6% (n=126) of pork and 42.9% (n=115) of chicken samples, and 14 *Salmonella* serotypes were identified. *S.* Anatum (15.8%) was the most common serotype, followed by *S.* Infantis (13.3%), *S.* Emek (10.4%), *S.* Derby and *S.* Rissen (9.5%), *S.* Typhimurium (9.1%), *S.* Reading (7.5%) and *S.* London (6.2%). The isolation frequency of *S.* Enteritidis, *S.* Albany, *S.* Hadar, *S.* Weltevreden, *S.* Newport and *S.* Blockey ranged from 1.2%–5.8%. Among those *Salmonella* serotypes, resistance to at least one antibiotic agent was detected in 78.4% of isolates (n=189) (Thai et al., 2012).

2.3.2 Risk factors related to *Salmonella* at pig farm, slaughterhouse and market

2.3.2.1 Pig farm

The source of *Salmonella* at herd-level is often difficult to determine because the host and vector range is broad. Contaminated environment, feed and water, biological and mechanical vectors are all known to be possible sources of infection

(Gebreyes, 2004, Fosse et al., 2009). Therefore, biosecurity is considered to be an important factor to prevent *Salmonella* introduction and spreading at herd-level. Feed is one of potential sources of *Salmonella* to pig herds (EFSA, 2008e). Poor hygiene practices are recognized as a risk factor for *Salmonella* infection at farm (Berends et al., 1996) also all-in-all-out management practices (Farzan, 2006). In addition, it was reported that smaller herds have a significantly higher *Salmonella* prevalence than larger herds (van der Wolf, 2001). In contrast, Mousing et al. showed, that large herds were associated with higher *Salmonella* sero-prevalence (Mousing et al., 1997).

2.3.2.2 Pig slaughterhouse

Transportation and lairage: Salmonella shedding is significantly increased during transport and lairage, which might be a result of high animal density, stress and feed deprivation (Berends et al., 1996). In association with stress, catecholamine might be released, resulting in increased gastric pH due to decreased gastric acid production, favoring survival of *Salmonella*. Increased intestinal motility leads to increased defecation frequency also favoring *Salmonella* spread into the environment (Schwartz, 1999). Additionally, contaminated trucks might act as a source of *Salmonella* (Fedorka-Cray, 1997). Different studies indicate that lairage allows for *Salmonella* cross-contamination and infection (Wong et al., 2002, Beloeil et al., 2004). Hence, the longer the time spent in lairage, the higher the probability of *Salmonella* contamination and infection.

Slaughter processing line: Cross-contamination during slaughter also represents a major public health problem. *Salmonella* contamination of carcasses after slaughter was partially caused by *Salmonella*-infected herds that were slaughtered before, and partially by residential flora of the slaughterhouse (Swanenburg et al., 2001). *Salmonella* can be transferred from the intestinal content to edible tissues of the individual carcass, but also between carcasses and through contact with the equipment, other surfaces and workers or by dripping or washing of contaminated water onto carcasses (Wong et al., 2002, Botteldoorn et al., 2004, Botteldoorn et al., 2003). Along the slaughter line, certain stages represent potential cross-contamination points, which are considered as critical control points. Evisceration is also a major source of contamination, different studies showed an increased contamination after this point (Berends et al., 1997, Hald et al., 2003). Moreover, hygiene levels vary between abattoirs and significantly affecting *Salmonella* carcass contamination (EFSA, 2008c).

2.3.2.3 Pork market

Positive pork at retail or market can be attributed to pig carcasses from slaughterhouse, vehicle and equipment in transportation and handling. *Salmonella* on carcasses at slaughterhouse might lead *Salmonella* positive pork. Many potential risks can induce contamination including hygienic practice or human handling, equipment, storage conditions at retail or market. The hygiene performance, particularly at retail, has a significant impact on the occurrence of *Salmonella* (Hansen et al., 2010). *Salmonella* Typhimurium was the most common serotype and the majority of isolates from pork shop. PFGE analysis showed evidence of persistence of some strains, with an S. Typhimurium U310 recovered from a pork abattoir being identical (100%) to a strain found a year later in a sample from a retail outlet (Prendergast et al., 2009).

2.3.3. Practice and perception of relevant groups on food safety

2.3.3.1 Practice and perception of relevant groups on food safety

Regulation on slaughter conditions and requirements in Vietnam (brieft): Animal slaughter has to operate at slaughterhouse or slaughter-point which has authority certification according to current regulation. Prohibit slaughter disease animal or suspected disease animals named in the list of MARD. There has a separate slaughter compartment, far from residential and public building areas at least 100 meters. Slaughter point, which slaughters less than 10 pigs per day may not oblige, but have to ensure not polluted environment. Slaughterhouse has to have enough equipment for slaughtering according to slaughterhouse scale. Equipments for slaughtering have to be clean well before and after slaughter; container keep meat or by-product have to have cover and dryable. Water source has to be clean. Slaughterhouse has drainage, container to treat waste, waste water for ensuring environment sanitation. People working in slaughterhouse have to be free of contagious diseases and check health once within 12 month period. Centralized slaughterhouses have to have veterinary staff to inspect animals' disease situation before animals go into slaughterhouse in city, town or municipal. Pigs, cattle or buffaloes admitted to slaughterhouse have to have inspection certificate or vaccination document of veterinary authorities (MARD-MOIT, 1997).

Regulation on meat selling conditions and requirements in Vietnam (brieft): Pork, cattle or buffalo meat, by-product or meat products sold on the market have to originate from legal slaughterhouses and be inspected and stamped or attached with hygienic stamp (for processed products) by veterinary authorities, or issued veterinary hygiene certificate. Organizes, individuals are issued certificate for meat and/or processed meat trade if they fulfill requirements such as have enough and appropriate equipments, clean location, etc. Sellers have to be free of contagious diseases and check health once within 12 month period. Shop owner at selling place has responsibility for origin and quality of meat and processed meat. Prohibit selling meat which doesn't pass over veterinary inspection. Meat processing plants for selling or serving meals at site have to use meat from legal slaughterhouse. Processed meats for sale has to have covers equipments to avoid dust, insects and ensure hygiene and quality when selling (MARD-MOIT, 1997).

Regarding perception of food quality in terms of purchase criteria for food in Germany, Rohr et al. (2005) showed that price appeared to be the most important purchase criterion: 66% of the responders mentioned the price. 37% of the participants called for freshness/not spoiled. Thirty three percent named quality, 15% appearance, 15% ingredients (fat, sugar, nutritive value) and 14% specified the best before date. This study also showed that eighty percent of consumers expressed a willingness to pay a premium price for notably approved products. Consumers seemed to be willing to pay 30% extra for eggs as well as minced beef and 22% extra for apples which were exposed in terms of food safety. Willingness to pay was more pronounced for animal than for plant food (Rohr et al., 2005).

A study in Ha Tay, Vietnam (2010) revealed that nearly half the consumer (43%) had concerns about pork: most common was fear of disease from pork, followed by fear of chemical contaminants, unfresh pork and bad smell. Only one person mentioned nutritional concerns. Consumers bought from known and trusted source in order to reduce risk (Grace, 2010). In another study in Vietnam, Lapar et al. (2009) reported that high and increasing demand for pork and strong preference for fresh, unchilled meat supplied by traditional market outlets in Vietnam (Lapar et al., 2009). A study on consumer perspective in Vietnam (2012) showed that housewife often bough food for their family and was in charge of cooking pork. Meat was bough mainly from the informal market and quickly prepared, cooked and consumed. People had high trust in pork safety and quality and rarely attributed health issues to pork consumption. The main concern was growth promoters, pork refresher (chemicals used to make not fresh pork appear fresh) as well as diseased pork. There was little knowledge of zoonoses (Hung et al., 2012).

An on-going study in Hung Yen and Nghe An, Vietnam (2012) described producers' perspectives that pork sold at the local market was safe because local butcheries often buy healthy pigs from local farms, however they had no ideas concerning possibility of low quality pork sold at the local market. Pig collectors groups expressed their less concern on pig diseases and diseases spreading. Slaughterhouse owners and workers/butchers showed their opinions that they keep hygienic practice in slaughtering by washing pigs before slaughtering with detergent or sometimes boiling. Slaughtered waste water was discharged directly into pools/rivers/drainage system. Whereas solid wastes such as pig hair and skin, etc were gathered into plastic bags then brought to public waste bins. Slaughter workers were provided with knives and boots only, no other protective cloth or devices. From consumers' perspectives, they mentioned that pork was purchased based on their experiences such as color, texture, odor, viscosity and relationship with sellers (ILRI, 2012).

2.3.3.2 Focus groups discussion and In-depth interview

Focus groups discussion are a qualitative data collection method effective in helping researchers learn the social norms of a community or subgroup, as well as the range of perspectives that exist within that community or subgroup. Focus groups are often used to determine what service or product a particular population wants or would like to have, such as in marketing studies. One or two researchers and several participants meet as a group to discuss a given research topic. These sessions are usually tape recorded, and sometimes videotaped. One researcher (the moderator) leads the discussion by asking participants to respond to open-ended questions – that is, questions that require an in-depth response rather than a single phrase or simple “yes” or “no” answer. A second researcher (the note-taker) takes detailed notes on the discussion. A principal advantage of focus groups is that they yield a large amount of information over a relatively short period of time. They are also effective for accessing a broad range of views on a specific topic, as opposed to achieving group consensus. Focus groups are not the best method for acquiring information on highly personal or socially sensitive topics; one-on-one interviews are better-suited for such topics (Mack et al., 2005).

Though moderator can control the discussion, the extent to which he/she can control the discussion depends on his/her experience. Inexperienced moderator may face problems in controlling some participants who try to dominate the group. Respondents may be reluctant to share some sensitive ideas and concerns publicly. Groups can be difficult to assemble. It is difficult to persuade people to give up their time and to find a time suitable for all participants Due to small sample size and heterogeneity of individuals, focus groups findings may not be adequate to make projections. A focus group can be a very artificial set up which influence the respondents to express and act unnaturally. Data can be not easy to summarize and analyze (ICS, 2012).

The ***in-depth interview*** is a technique designed to elicit a vivid picture of the participant’s perspective on the research topic. During in-depth interviews, the person being interviewed is considered the expert and the interviewer is considered the student. The researcher's interviewing techniques are motivated by the desire to learn everything the participant can share about the research topic. In-depth interviews are useful for learning about the perspectives of individuals, as opposed to, for example, group norms of a community, for which focus groups are more appropriate. They are an effective qualitative method for getting people to talk about their personal feelings, opinions, and experiences. They are also an opportunity for us to gain insight into how people interpret and order the world. We can accomplish this by being attentive to the causal explanations participants provide for what they have experienced and believe and by actively probing them about the connections and relationships they see between particular events, phenomena, and beliefs. Interviews are also especially appropriate for addressing sensitive topics that people might be reluctant to discuss in a group setting (Mack et al., 2005).

The primary advantage of in-depth interviews is that they provide much more detailed information than what is available through other data collection methods, and a more relaxed atmosphere. However, interviews can be a time-intensive evaluation activity because it takes time to conduct interviews, transcribe them, and analyze the results. Program or staff might want to "prove" that a program is working, so their interview responses might be biased. Responses from community members and program participants could also be biased due to their stake in the program or for a number of other reasons. Interviewer must be appropriately trained in interviewing techniques and use effective interview techniques. When in-depth interviews are conducted, generalizations about the results are usually not able to be made because small samples are chosen and random sampling methods are not used. In-depth interviews however, provide valuable information for programs, particularly when supplementing other methods of data collection (Carolyn and Palena, 2006).

2.4 Pork production and consumption in Vietnam

In the livestock sector, pig farming takes a dominant role. The estimated total of pig population in 2010 is 27,4 million (decrease 9% as compared to 2009). The living weight of pigs per capita was average 35 kg (GSO, 2010). About 40% of the country's pig population and half of the pork products are produced in the Mekong River Delta and Red River Delta (Dinh, 2001). Pork production is mainly based on the household level; about 60% of the rural population keeps pigs (Costales, 2007). There were 548 commercial pig farms which keep at least 100 pigs (Dinh, 2001), contributing to 20-25% of the total pork production (GAIN, 2006). The pork production and consumption is shown in Figure 1.

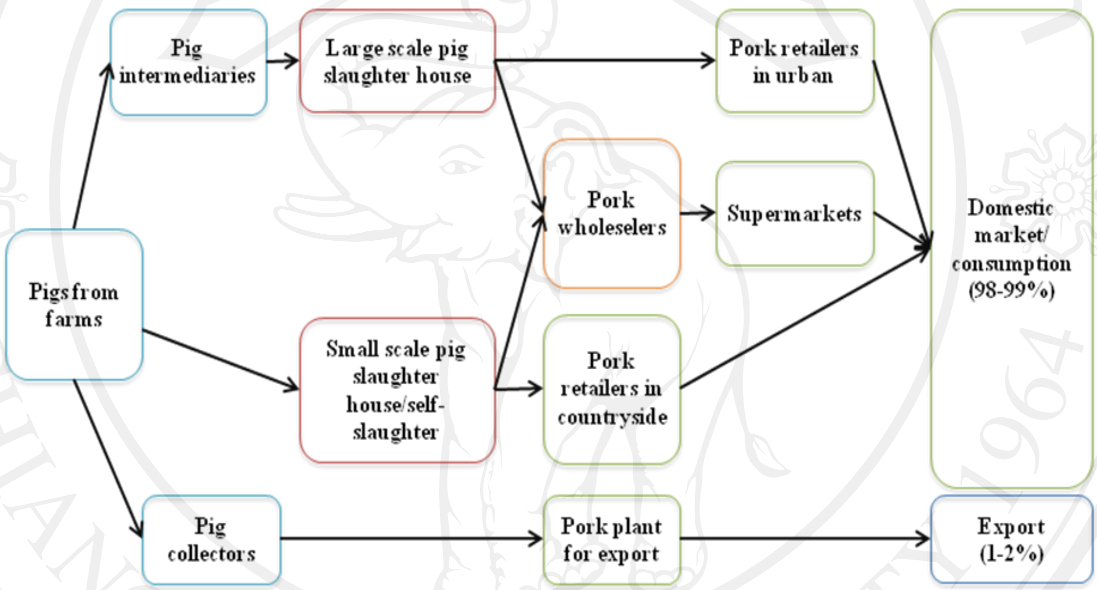

Figure 1 Pork chain from production to market in Vietnam

(Extracted from Vo T.T 2011)

Most of the pork produced in Vietnam is mainly for domestic demands and only 1-2 % for export (Nguyen, 2006). Vietnamese's taste prefers fresh meat to frozen. It is estimated that 95 % of the households are consuming pork in their daily diets (Dinh, 2001). Therefore pigs are slaughtered on a daily basis and most of the pork is consumed as fresh meat within a day (GAIN, 2006).

2.5 Impacts of salmonellosis and control measures

2.5.1 Public health impacts

In humans, salmonellosis has been reported as one of the majority foodborne disease worldwide for decades. The number of nontyphoidal *Salmonella* gastroenteritis cases was estimated at 93.8 million globally each year, with 155,000 deaths. Of these figures, about 80.3 million cases were foodborne (Majowicz et al., 2010). In 2012, *Salmonella* was ranked of the highest number of lab confirmed infections with 7,800 cases (CDC, 2012a). In the European Union, a total of 95,548 confirmed salmonellosis cases in humans were reported in 2011 (EFSA, 2011).

Moreover, antimicrobial resistance among *Salmonella* serotypes is also known as another major public health issue. *Salmonella* infections in humans may be usually self-limiting. Increased number of cases of multi-resistant S. Typhimurium and S. Typhimurium DT104 caused great concern. Antimicrobial resistance might be associated with increased virulence of the organism and/or poor response to treatment (Helms et al., 2004, Boyen et al., 2008). In addition, resistant and multi-resistant *Salmonella* serotypes are commonly isolated from pigs (Gebreyes, 2004).

Salmonella infection is a significant burden in both developing and developed countries and it has increased dramatically in the number of human cases. This might be a consequence of enhanced monitoring and reporting systems, increased food safety and consumer awareness, increased of consumption of animal food related, changes in consumption habits as well as widespread distribution of zoonotic *Salmonella* serotypes in the human population (Schwartz, 1999). Moreover, it can also be explained by different herd and slaughter management associated with increase herd and abattoir size. These might lead to higher infection levels in herds, cross-contamination during transportation, at lairage and slaughtering process, as well as presence of the agent at retail, including consumer households (Boyen et al., 2008).

In Belgium, Czech Republic and France, there had 3,177, 8,499 and 8,685 cases of human salmonellosis were reported in 2011, respectively. The number of human salmonellosis cases reported in Germany in 2011 was 23,982, decreased in comparison with number of cases in 2010 (24,833) (EFSA, 2011). It was estimated that 15–20% of all human cases of salmonellosis were associated with the consumption of pork in Germany (Botteldoorn et al., 2003). However, reported cases are likely to represent only a minor proportion of the total number of cases found in human population (EFSA, 2008b). According to a study on infectious intestinal disease in England, for every 3.2 cases of salmonellosis, only one case was reported to national authorities (Wheeler, 1999).

In the United States, every year, approximately 40,000 cases of salmonellosis are reported. Many milder cases are not diagnosed or reported, the actual number of infections may be thirty or more times greater. Salmonellosis is more common in the summer than winter. Young children, the elderly, and the immunocompromised are the most likely person groups to get and have severe infections. It is estimated that approximately 400 people in the US die each year with acute salmonellosis (CDC, 2012b).

In the South East Asia region, official *Salmonella* surveillance data do not exist, but it is estimated that up to 22.8 million cases occur annually with 37,600 deaths (Majowicz et al., 2010). In Thailand, *Salmonella* is the main bacterium which causes gastrointestinal and systemic infections. In Vietnam, according to the report of the Department of Food Administration, Ministry of Health, Vietnam, from 2007 to 2011, 927 outbreaks of food poisoning occurred, with 30,734 cases and 230 deaths. Of these, in 2011, 148 outbreaks of food poisoning with 4,700 people infected and 27 deaths were reported. A proportion of over 28% of these cases were caused by several pathogens (VFA, 2011). However, human salmonellosis cases were not recorded. For non-typhoid *Salmonella* infections in humans, S. Typhimurium is the most common cause in South Vietnam (Vo et al., 2006b).

2.5.2 Economic impacts

Besides health impacts, salmonellosis also impacts on economics. *Salmonella* infections are associated to decreased livestock and food production, especially impact on human health is costly. Costs are spent for surveillance, investigation, reporting systems and intervention in general as well as for treatment, compensation for loss of production and indirect costs. However, very few countries report data on the economic cost of *Salmonella*; data related to the cost of foodborne disease are generally not available from developing countries (WHO, 2005).

The USDA's Economic Research Service (ERS) estimated that *Salmonella* infections from all sources cost about $2.65 billion per year in the US. That is based on an estimate by the Centers for Disease Control and Prevention (CDC) of almost 1.4 million *Salmonella* cases annually from all sources, with 415 deaths. The estimated average cost per case is $1,896 (ERS, 2012).

In the EU, a total estimated cost of human salmonellosis was about € 608 million in 2008. In Germany, United Kingdom and France, a total estimated cost of human salmonellosis was about € 255, 82 and 38 million, respectively and the average cost per *Salmonellosis* case in these Members States was estimated at € 832, 1,066 and 207 (FCC, 2010). In the Netherlands, annual social costs caused by human salmonellosis were estimated between 32 and 90 million Euro (van Pelt, 2001).

2.5.3 Control measures of Salmonellosis

Along the pork production chain, *Salmonella* can enter at any point, from pre-harvest, harvest and post-harvest. So control measures of salmonellosis should be established at each stage. Control of *Salmonella* infection in farms included strategies to mitigate the risk of introduction, transmission and spreading within and between farms. *Salmonella* introduction and spreading at herd-level is critically determined by multiple factors (bio-security measures, feed or water...) (Fedorka-Cray, 2000). Therefore, bio-security measures, incoming materials should be under control.

A study using Danish data showed that control measures included additional cleaning and disinfection, addition of organic acids to feed and water, change to all-in-all-out system, among others (Anonymous, 2000). High biosecurity standards are additionally expected to further reduce the prevalence of other important pathogens besides *Salmonella* (Anonymous, 2006).

Well-documented studies on the efficacy of vaccines are lacking (Denagamage, 2007). Live attenuated orally administered vaccines are considered to stimulate an effective cell-mediated immune response against host-specific serotypes.

During transport and lairage, pigs should be managed carefully and mixing of unfamiliar pigs should be avoided. Thorough cleaning and disinfection of trucks and lairage should be ensured between batches of pigs. Lairage time should be kept to a minimum (Wong et al., 2002).

At slaughterhouse level, strict compliance with hygiene measures should be followed. Critical control points should be identified and systematically monitored assuring compliance with Hazard Analysis Critical Control Point (HACCP) principles (Wong et al., 2002).

At manufacturing and retail, temperature should be kept below 7ºC, to inhibit growth of *Salmonella*. HACCP principles should be strictly followed, namely good manufacturing practices, including hygiene and processing procedures. To enhance microbiological stability, pork products can be further processed (e.g. acidification, fermentation, curing, and smoking) (Wong et al., 2002).