FOOD, AGRICULTURE AND FISHERIES, AND BIOTECHNOLOGY

FRISBEE

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1. Introduction

The objective of WP3 is the establishment of QEEAT tools for evaluation of quality, energy consumption and environmental impact of refrigeration technologies. The objective of this work package is also to provide evaluation tools (sensors, equipment, software, protocols and methodologies) to assess and improve existing refrigeration technologies and emerging new technologies and concepts which will be addressed in further work packages, with respect to energy efficiency, quality of refrigerated foods and sustainability based on the framework developed in WP2 and for the selected refrigerated foods. **Subtask 3.2.4. is concerning by safety and quality modeling.** The quality and safety attributes of selected products considered were identified in this subtask (DOW FRISBEE). The accompanying description in the DOW is as follows: "NTUA, KUL, SINTEF and ACTIA (via its third parties AERIAL and ADRIA) will compile existing microbiological and quality kinetic models and data related with their food product(s) under chilled, superchilled and/or frozen conditions, as listed in Table 1. The compilation will include an extended literature search and, with respect to microbiological safety issues for the food products under consideration, the careful exploitation of the public domain microbial data and model ".

The D. 3.2.4.2 is a review on chilled pork meat and Ready to Eat pork meals and frozen pork meat. For chilled RTE pork meals, the safety indicators are "*Listeria monocytogenes* and spoilage lactic acid bacteria" and quality indicators are "texture and odor". For frozen pork meat, the quality indicators are "texture and odor". "This compilation will be completed with carefully designed (static and dynamic) experiments on the different foods using experimental facilities at the different partners involved, if necessary" (DOW FRISBEE).

2. Safety indicators and models for chilled pork meat and Ready To Eat (RTE) pork meals

2.1. *Listeria monocytogenes,* challenge in food production

Listeria monocytogenes may cause disease in humans and it is typically transmitted as a food-borne pathogen. *L. monocytogenes* is frequently present in the environment, faeces of animals. The organism can be found in raw foods such as fresh meat, raw milk and fish. The ubiquitous occurrence and the ability to grow or survive in a chilled environment compared to most other microorganisms, **makes *L. monocytogenes* a significant challenge in food production**. This is especially the case for ready-to-eat (RTE) foods in which *L. monocytogenes* can grow. It is crucial that producers of RTE foods (food intended by the producer for direct human consumption without the need for cooking or other processing effective to eliminate or reduce microorganisms of concern to an acceptable level) take actions to control contamination of *L. monocytogenes*, as well as its growth in the product until the end of shelf-life.

Food business operators (FBO) producing RTE foods must be able:

- to demonstrate to the satisfaction of the competent authority that the products will comply with the Community Regulation until the end of the shelf-life,
- to understand the range of different approaches available to help establish a safe product shelf-life in relation to *L. monocytogenes* and to decide the appropriate approach for their products and
- to classify their products into RTE foods in which growth of *L. monocytogenes* can occur or in RTE foods in which growth of *L. monocytogenes* will not occur during their shelf-life.

The EU has established microbiological food safety criteria for *L. monocytogenes* in RTE foods in **Regulation (EC) No 2073/20053** of 15 November 2005 on microbiological criteria for foodstuffs. Article 3 of Regulation (EC) No 2073/2005 indicates that Food Business Operators

shall ensure that foodstuffs comply with the relevant microbiological criteria and limits set out in the Regulation. In particular, this applies to RTE foods that are able to support the growth of *L. monocytogenes* and that may pose a *L. monocytogenes* risk for public health.

The specific food safety criteria for *L. monocytogenes* in RTE foods are laid down in **Annex I** of the Regulation. Food safety criteria define the acceptability of a product or a batch of foodstuff applicable to products placed on the market. Furthermore, corrective actions at the production plant according to the hazard analysis of critical control point (HACCP) plan shall be taken.

Annex II of the Regulation describes the shelf-life studies that the FBO shall conduct, as necessary, in order to investigate compliance with the criteria throughout the shelf-life. These shelf-life studies shall always include:

- specifications of physico-chemical characteristics of the product (such as pH, aw, salt content, concentration of preservatives and the type of packaging system) taking into account the processing steps and conditions storage and the possibilities for contamination and the foreseen shelf-life, and
- consultation of the available scientific literature and research data regarding the survival and growth characteristics.

The FBO is responsible for setting the quality and safety defined conditions, which should take into account reasonably foreseen conditions of distribution, storage and use**. An important part of these foreseen conditions is the kinetics of temperature** during the entire life.The determination of the quality and safety of foodstuffs shall always include the consideration of the different factors such as **cold chain** food sector, type of product and type of process. The inherent variability of manufactured batches and the variability linked to microbial species shall also be taken into account, **as well as all the reasonably foreseeable conditions during processing, distribution, storage and use, included those applied by the consumer.**

2.2. Factors influencing the behaviour of *Listeria monocytogenes and* existing data

Listeria monocytogenes is a ubiquitous pathogen that can be found in a large number of food products, including **pork meat and processed pork products**. Pork meat is potentially contaminated (Lianou A, Sofos JN 2007; Thévenot et al 2006 ; Bërzins et al 2007 ; Lin et al 2006a et b ; Vorst et al 2006 ; Tompkin 2002). Certain products like jellied pork tongue have been responsible for epidemic listeriosis, as well as potted minced pork (De Valk and al, 2000 ; Thevenot and al, 2005). The occurrence of *Listeria monocytogenes* **in the pork-processing industry from the slaughterhouse to the cutting room makes it difficult to avoid minor contamination during the meat process** (Stekelenburg, 2003).

Listeria monocytogenes is a psychotrophic bacterium which can develop during refrigerated storage. To assure the safety of the products, industries can limit the contamination using :

- Good manufacturing processing
- Hazard Analysis Critical Control Points
- Relevant processing steps
- **Good control of the chilled chain.**

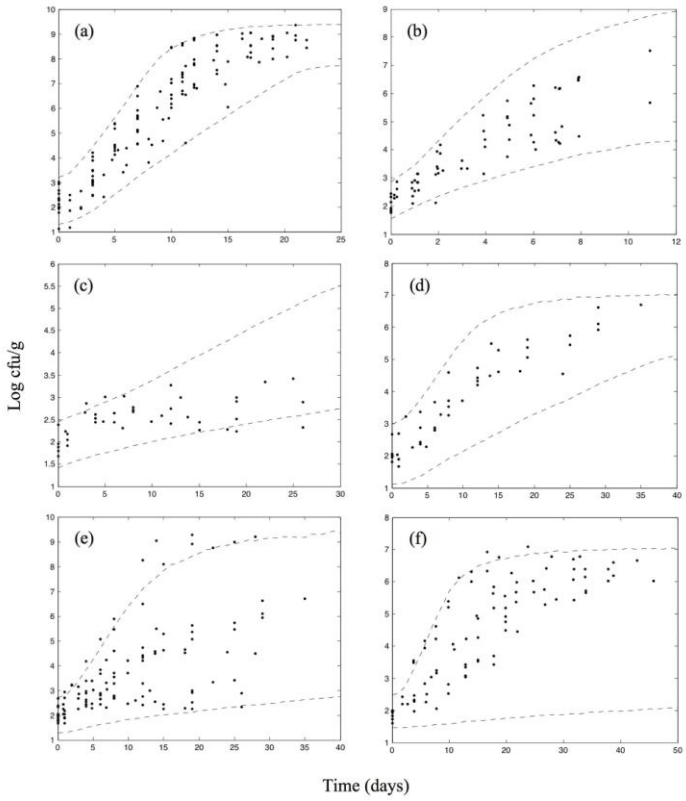
The control of factors such as pH, water activity, **temperature**, additives, organic acids allow control of the growth of the pathogen bacteria and improve the shelf life of the pork products (Zuliani and al, 2007 ; Augustin and al, 2005). The influence of these factors are known (Notermans and In't Veld, 1994 ; Koutsoumanis and al, 2004 ; Tienungoon and al, 2000).

The behaviour of *Listeria monocytogenes* in a specific food matrix is dependent on the product's ecological characteristics : structure, pH, type of the acid, water activity, presence of additives, presence of natural dominant flora **like lactic flora** (Duffes and al, 1999a and b ; Bredholt and

al, 1999) and environmental conditions of storage [temperature / time ; modified atmosphere] (Devlieghere and al, 2001).

Another aspect is the assessment of the evolution of microorganisms naturally contaminating food which must take into account the **variability** of biological factors, food characteristics and storage conditions. The estimation of the variability of maximum growth rate and population lag time in natural conditions of contamination is generally impossible for pathogenic microorganisms and food business operators must usually perform challenge testing. A research project (Augustin and al 2010) was conducted to quantify the variability of growth parameters of *L. monocytogenes* obtained by challenge testing in five different foods. The objective was to evaluate the within and between-batch, between-manufacturer, and between-initial microbial physiological state variability of the growth parameters to optimize the challenge testing methodology applied when evaluating the variability of the behaviour of microorganisms in foods. Challenge tests for five foods were conducted, including vacuum-packed pork pie, and sliced cooked packed ham under modified atmosphere (Figure 1):

Figure 1. Observed (●) and 95% confidence bands (dashed lines) of simulated growth of *L. monocytogenes* at 8°C in (a) pork pie, (b) surimi salad, (c) cooked ham from manufacturer CHC, (d) cooked ham from manufacturer CHA, (e) cooked ham from the three manufacturers, (f) smoked herring from the two manufacturers.



It appeared that repeating a limited number of three challenge tests in different batches/manufacturers for different initial physiological states is often necessary and adequate to accurately assess the variability of the behavior of *L. monocytogenes* in a given food.

Survival of *Listeria monocytogenes* on cooked bacon cubes (aw = 0.910±0.08), strips (aw = 0.726±0.054), and bits (aw = 0.62±0.038) was determined during a 25 week storage period at 20, 4.4 and 22°C (Taormina and al 2010). Samples inoculated at ca. 5.5- log10 CFU/g (HI). At 20°C a decline of about 1-log10 CFU/g occurred on all HI cooked bacon types by 14 weeks, although most LI samples remained positive for 25 weeks. At 4.4 and 22°C, some strips and bits LI samples were negative for the pathogen within 3 weeks and > 1.5 log10(UFC/g) reductions occurred on HI strips and bits by 8 weeks. Reductions on cubes at refrigeration and ambient

temperature were ca. 0.5 log10 (UFC/g), and cubes remained positive on LI samples for 25 weeks. Rate parameter estimates indicated that the population declined fasted on strips and bits at 22°C compared to all other product and temperature combinations. In this study, cooked bacon does not support the growth of *L. monocytogenes*; the pathogen gradually dies off during storage.

Commercial cured ham (Lianou and al 2007) formulated with or without potassium lactate and sodium diacetate was inoculated with *L. monocytogenes* and stored to simulate conditions of processing, retail, and home storage. The ham was sliced, inoculated with a 10-strain composite of *L. monocytogenes* (1 to 2 log CFU/cm²), vacuum packaged, and stored at 4°C to simulate contamination following lethality treatment at processing (first shelf life). After 10, 20, 35 and 60 days of storage, samples were tested. At the same times, portions of original product were sliced, inoculated, and packaged in delicatessen bags to simulate contamination during slicing and retail (second shelf life). Aerobic storage of both sets of packages at 7°C for 12 days was used to reflect domestic storage conditions (home storage). ***L. monocytogenes* populations were lower during storage in ham with lactate-diacetate than in product without antimicrobials under both contamination scenarios. Inoculation of ham without lactate-diacetate allowed prolific growth of *L.monocytognes* in vacuum packages during the first shelf life and was the worst case contamination scenario with respect pathogen numbers encountered during home storage.**

2.3. Models applicable in the cold chain of chilled pork meat and RTE pork meals

The successfully validated models are useful for assessment and management *of L. monocytogenes* in processed and ready-to-eat (RTE) foods. Evaluation of predictive microbiology growth models includes a comparison of predicted growth responses with those observed in food. Typically, food data are obtained from challenge tests with inoculated products but data from naturally contaminated food are important and should be used when they can be obtained. These evaluations and the resulting indices of performance (e.g. bias and accuracy factors) are important to determine if a predictive model can be used with confidence.

Several predictive models to quantify the effect of several factors on the growth rate of *L. monocytogenes* have been proposed (not exhaustive: Rosso and al, 1995 ; Pinon and al, 2004 ; Zuliani and al, 2006 ; Zuliani and al, 2007 ; Coroller and al, 2005; Dalgaard and al 1998; Mejlholm and al 2010; Koutsoumanis and al 2004).

The behaviour of *L. monocytogenes* in meat products was studied by many authors (Zamora and al 1985 ; Deumier and al 2003 ; Mbandi 2002 ; Stekelenburg and al 2003, Coleman and al 2003 ; Lu and al 2005 ; Pal and al 2008a et b ; Uyttendaele M. 2004 ; Yang et al 2006 ; Dykes et al 2003 ; Jones et al 2008). As *L. monocytogenes* is a psychrotropic bacterium, it can develop during refrigerated storage. In order to assure the safety of their products, industries can limit the contamination of *L. monocytogenes* using good manufacturing processing and using additives to control the growth. For example, salts of acids (potassium sorbate, sodium acetate, sodium lactate, mixture of acid salts) act by lowering water activity, the cell pH and by inhibiting enzymes (Zuliani and al 2007 ; Choi and Chin, 2003 a and b).

Several predictive models integrate the effect of these inhibitors and the effect of the interactions between environmental factors (Coroller and al, 2005 ; Augustin and al, 2005 ; Zuliani and al, 2007 ; Mejlholm. and al 2010).

Mejlholm et al 2010 make an exhaustive review of the subject. The authors evaluate recently the performance of six predictive models for *Listeria monocytogene*s using 1014 growth responses of the pathogen in meat, seafood, poultry and dairy products. For example, growth of *L. monocytogenes* and background flora (like lactic acid flora) and storage data are available concerning ham (Stekelenburg and al 2001; Hwang and al 2007), sausages (Pal and al 2008),

pork Bologna (Barmpalia and al 2005), Bratwurts, Frankfurters (Pal and al 2008), Servela, Wieners (Glass and al 2002).
Predictive models studied are well described. The performance of the growth models was closely related to their complexity i.e. the number of environmental parameters they take into account. The most complex model included the effect of nine environmental parameters and it performed better than the other less complex models both for prediction of maximum specific growth rates (μmax values) and for the growth boundary *of L. monocytogenes*. The performance of three other models, including the effect of five to seven environmental parameters, was considered acceptable. These models all included the effect of acetic acid/diacetate and lactic acid, one of the models also included the effect of CO2 and nitrite but none of these models included the effect of smoke components. Less complex models that did not include the effect of acetic acid/diacetate and lactic acid were unable to accurately predict growth responses of L. monocytogenes in the wide range of food evaluated in the present study.
When complexity of *L. monocytogenes* growth models matches the complexity of foods of interest, i.e. the number of hurdles to microbial growth, then predicted growth responses of the pathogen can be accurate.

Other model software is available in the context of meat and processed meat:
COMBASE www.combase.cc
Sym'Previus www.symprevius.org
PMP Pathogen Modeling Program www.arserrc.gov/mfs/pathopgen.htm
DRMI 2007 *Listeria* model from Danish Meat Research Institute http://1.test.dezone.dk
PURAC http://purac.com/EN/Food/Contact.aspx.

Probabilistic approach
Predictive microbiology aims at providing reliable models for simulation of microbial behaviour in food products. Traditional approaches are often based on 'worst-case' scenarios estimating the bacterial growth likely to occur in a food product: models are typically developed using a fast-growing strain (or cocktail) in an optimal growth medium (e.g., liquid broth or ground meat without adverse microflora). Such approaches generally give safe results. Furthermore, worst-case scenarios may also omit important cases: an even faster growing strain may be found, growth conditions in food may prove more favourable to adapted strains. Describing a general trend while accounting for variability sources within the model would, thus, be more efficient for food industry applications, by providing safe enough predictions to avoid unacceptable health risks for consumers while maintaining cost-effective processes and decisions (Juneja et al., 2003; McMeekin et al., 2002). Output from a stochastic model should indicate a distribution of probabilities for contamination, instead of a single deterministic value, in relation to a threshold level and an acceptable probability of exceeding a critical level (Koutsoumanis and Angelidis, 2007; Whiting et al., 2006).

Couvert and al 2010 built a bacterial growth model using a stochastic approach: sources of variability were included in the model to predict the evolution of the distribution of the *Listeria monocytogenes* contamination in foods throughout their shelf life. This model is part of the Sym'Previus software (http://www.symprevius.org) designed for prediction of microbial behaviour. The biological variability of bacterial cardinal values is already set in the software but the variability of growth parameters, initial contamination, food characteristics and storage conditions must be specified by the users. The individual cell variability of lag times is deduced from the population lag time or physiological state assuming an extreme value type II distribution of individual cell lag times in the bacterial populations (Guillier and Augustin, 2006). A stochastic approach may consider influences of variability and uncertainty on the growth parameters. So far, modelling of variability in stochastic models has mostly dealt with biological parameters (Marks and Coleman, 2005) and single cell lag times (Guillier et al., 2005; Métris et al., 2005). Other authors have built more complete models including variability and uncertainty of parameters using a Bayesian approach (Delignette-Muller et al., 2006; Pouillot et al., 2003). These models are particularly useful in the framework of quantitative risk assessment: distribution of contamination probabilities may be obtained as a function of time or stage in the product life.

Model performance was evaluated by comparing the results of the software simulations with experimental data obtained in artificially or naturally contaminated foods.

This model determines probability to exceed targeted contamination criteria at the end of shelf-life. Fig. 2 shows an example of this distribution.

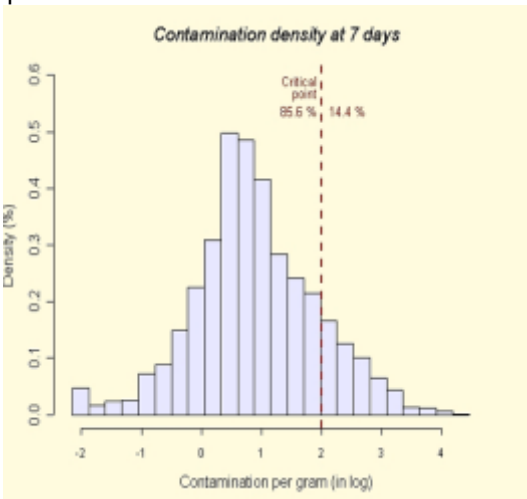


Fig.2 : Example of contamination density result given in Symprevius.

Fig.3 illustrates the flow diagram of the stochastic model used in Sym'Previus to evaluate the *L. monocytogenes* contamination of food after a storage step. Circles are used for random variables and squares indicate fixed parameters. Dotted arrows are used when two variables are linked by an equation and solid arrows are used when there is a random process between two variables.

w is the portion size; c_0 is the initial *L. monocytogenes* concentration; n_0 is the initial number of cells in the portion; $n_{\text{end storage}}$ ($c_{\text{end storage}}$) is the number (concentration) of cells in the portion at the end of the storage, $\mu_{\text{max,ct}}$, lag_{ct} and $x_{\text{max,ct}}$ are the growth parameters observed during challenge tests performed in conditions T_{ct}, pH_{ct}, aw_{ct}; T_{card}, pH_{card}, aw_{card} are cardinal values for *L. monocytogenes*; μ_{opt} and lag_{min} are the maximum specific growth rate and lag time when T, pH and aw are set to their optimal values; K and k_i are the population and individual physiological parameters, respectively; μ_{max} and lag_i are the maximum specific growth rate and the individual lag time for cell i in growth conditions T, pH, aw.

Model performance was evaluated by comparing the results of the software simulations with experimental data obtained in artificially or naturally contaminated foods.

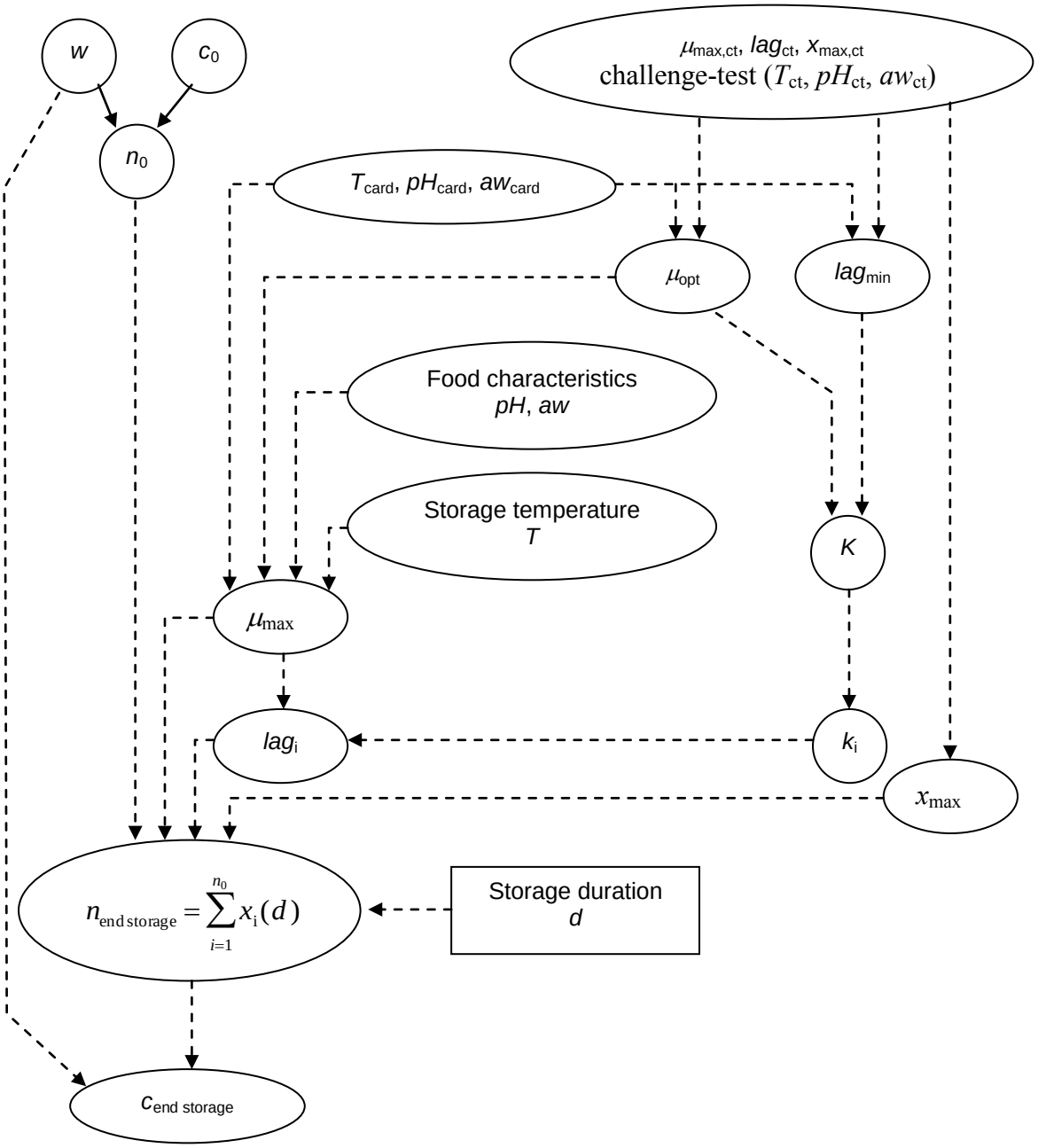


Fig. 3 Flow diagram of the stochastic model used in Sym'Previus

2.4. Competition between *L. monocytogenes* and indigenous flora

Other factors (structure of food stuff, formulation conditions, indigenous natural flora **like lactic flora**) are included in other modelling tools. Authors have studied interactions between indigenous flora and *Listeria monocytogenes* (Duffes et al 1999ab, Guillier et al 2008, Leroi et al 1998-2000-2001).The growth of *L. monocytogenes* in RTE food product is affected by the competition with the simultaneous growth of the food flora. This effect known as the "Jameson effect" implies that the growth of each of the two competitive flora is stopped as soon as one has reached its maximum level in the product (Dalgaard and Jorgensen, 1998; Ross *et al.,* 2000 ; Cornu et al 2003, Delignette et al..2006, Lakshmanan et al, 2004). The growth of the food flora of cold-smoked salmon is modeled similarly and independently of the *L. monocytogenes* growth. The aim is to be able to predict the simultaneous growth of *L. monocytogenes* and food flora in cold-smoked salmon in a competitive model such as the one proposed by Gime´nez and Dalgaard (2004); Delignette and al (2006) ; Cornu and al 2006 ; Pouillot and al 2009 ; Authors studied for meat products theses interactions (Taorma 2010; DEVLIEGHER et al 2001; Coleman et al 2003; Summer et al 2002). Malakar and al (2002 ; 2003) observed that microbial interactions are strongly related to a high population density in liquid laboratory media and related to the initial size of the inoculums. Van Impe (2005) described a new type of predictive models with the objective to integrate microbial interactions (production of metabolites and/or nutritional competition between *Lactococcus lactis* and *Listeria inocua*) inside structured food products. Frequently, nutritional competition is identified (Cornu et al 2003-2006 ; Coleman et al 2003 ; Giménez et Dalgaard 2004 ; Mejlholm et al 2007 ; Gnanou Besse *et al.* 2006 ; Crépet and al 2009). Authors suggested a geometric effect related to the bacteria, immobilized into a colony inside solid media, due to diffusion limited for metabolites, nutriments and cells. There is also a relation between inoculum size of *L. monocytogenes* and maximum population. This phenomenon was studied in different food stuffs (Guyer et Jemmi, 1991 ; Peterson *et al.,* 1993 ; Gnanou Besse *et al*., 2006). The effect is dependent on many factors interactions, like indigenous flora.

Today, in the food retailing sector, economic pressures has increased the development of fresh, ready-to-eat or ready-to-cook products. Packaging technological developments make it possible to consider the use of Modified Atmosphere Packaging (MAP). Protecting food is the main function of modified atmospheres. From a technical point of view and in terms of food safety, the gaseous environment (CO_2, O_2 and $N_{2)}$ in a package can slow down microbial development. MAP can thus change the microbiological ecology of the food. The changed environment may support the growth of certain pathogens (Farber, 1991) while inhibiting microorganisms responsible for spoilage. A food product may therefore appear edible even though it contains high numbers of pathogens that may have multiplied due to a lack of indigenous competition. N_2 has no specific effect on preservation, but due to its insolubility, it is used as a filler gas to avoid the collapse that occurs with soluble CO_2. High levels of O_2 can inhibit the growth of both anaerobic and aerobic microorganisms, since the optimal O_2 level for growth (21% for aerobes, 0.2% for anaerobes) is surpassed. CO_2 has an antimicrobial effect and it is the most important component in the choice of a gas mixture. When CO_2 is introduced into the package, it is partly dissolved in the aqueous phase and in the lipids present in the food product. The inhibition of microorganisms in modified atmosphere is determined by the concentration of dissolved CO_2 in the aqueous phase (Devlierghere *et al*., 1998 and 2000). Gram-negative microorganisms such as *Pseudomonas, Shewanella* and *Aeromonas* are very sensitive to CO_2 (Molin, 1983 ; Boskou and Debevere, 1997). Furthermore, pathogens such *Listeria monocytogenes* are minimally affected by CO_2 levels below 50%.

The effect of various parameters such as temperature and pH on the microbial growth was taken into account in this modelling approach and some authors have even tried to introduce the effect of CO_2 and O_2 concentration in the equations. Mathematical models have been developed to describe the effect of CO_2 for spoilage microflora (Koutsoumanis *et al*., 2000), *Aeromonas* (Pin *et al.*, 2004). In many cases, gas concentrations are fixed parameters. For example, Lee et al. (2008) studied the effect of MAP with different CO_2 concentrations on the microbial quality of Korean ready-to-eat food products stored at different temperatures. Then they modelled the bacterial growth but in fixed CO_2 concentration. Several mathematical models have been proposed for simulating the changes in gas concentration during shelf life. Many of these models

are designed for fruits and vegetables; the gas composition in the atmosphere of the package is modelled as the result of the interplay between product respiration and the transfer of gases through the packaging film (Charles *et al.*, 2003, 2005.) More recently, some models have been proposed for gas diffusion processes and soluble gas stabilisation systems for non-respiring foods (Simpson *et al*., 2009).

These data/models constitute a background for ACTIA's work at 2011, concerning lactic acid flora.

2.5. Data for spoilage lactic acid bacteria

Frisbee is interested particularly in natural lactic acid flora in this project, responsible for potential quality loss. Lactic acid flora (*Lactobacillus sp., Lactococcus sp., Leuconostoc sp*.) are ubiquitous flora that can be found in a large number of **meat products and RTE products** (Champomier-Verges and al, 2002 ; Hammes and al, 2004). The widespread occurrence of lactic acid flora in the pork-processing industry, its development during refrigerated storage into pork based RTE products are some major reasons to study it (Hüfner and al, 2007 ; Klaenhammer and al, 2002).

Pork meat:
Spoilage has always been, and still is a major concern in the food industry. **In meat, spoilage becomes apparent in the form of colour changes, off-odours and -flavours, as well as textural changes.** In uncooked, vacuum- (VP) or modified atmosphere-packed (MAP) meat, microbial growth is the main cause of meat deterioration (Borch et al. 1996; Gram et al. 2002). Extension of the shelf-life of fresh meat is often achieved by packaging products under vacuum or modified-atmosphere (MAP) conditions, in addition to chill storage (Tsigarida et al. 2000). The microbiota in vacuum-packed and MAP pork has been shown to be dominated by lactic acid bacterial and some other bacterial groups like *Brochotrix thermosphacta* and *Enterobacteriaceae* (Shaw and Harding 1984; Mcmullen and Stiles 1989 and 1993; Holley et al. 2004).

Marinated pork meat:
An increasing amount of meat is today sold as marinated meat products. In northern Europe, industrial marination is a process where meat products are injected with brine containing salt and phosphates before being packed in a flavouring sauce containing salt, sugar, organic acids and herbs or spices in the production plant. Even though antioxidants have sometimes been added, marination has so far mainly been associated with taste. The injection of meat with brine before marination has been shown to contribute to the total bacterial load of the products (Bohaychuk and Greer 2003; Greer et al. 2004). Schirmer and al study (2009) determine the microbial flora in fresh, vacuum-packed, marinated pork steak during storage and to identify important spoilage bacteria. The flora in marinated products was similar only for products from the same plant. Strains of *Lactobacillus algidus, Lactobacillus sakei, Lactobacillus curvatus, Carnobacterium divergens, Carnobacterium maltaromaticum, Leuconostoc mesenteroides, Leuconostoc carnosum and Leuconostoc sp.* were isolated and **tested for their spoilage** potential.

Processed pork meat products:
The shelf-life of vacuum- and modified atmosphere packaged (MAP) processed meat products is in general limited by the level and composition of the predominating lactic acid bacterial (LAB) flora (Samelis 2006; Nychas et al. 2008). There is an ill-defined time delay after attainment of maximum LAB numbers before onset of spoilage (McMullen and Stiles 1989; Korkeala and Bjo¨rkroth 1997; Mataragas et al. 2006). As an example, Danish ready-to-eat heat-processed MAP pork product was examined in a Laursen and al (2009) study. The aim was to evaluate the potential for developing a quality index for a Danish modified atmosphere packaged (MAP) heat-processed and naturally contaminated pork meat product stored at 5°C. The composition of the

predominating microflora and changes in contents of tyramine, arginine, organic acids and sensory characteristics were analysed.
The microflora was predominated by *Lactobacillus sakei, Leuconostoc carnosum and Carnobacterium divergens.*

Ready to eat pork meat:
Today, the evolution is to control in ready-to-eat pork products low concentration of additives, for health and nutritional aspects. The aim is also to control the **shelf-life in this new context**. Lactic acid bacteria mainly dominate and are potentially **sources of variation affecting sensory characteristics (odor, texture, …)**(Pal and al 2008). The shelf-life is limited by the level and composition of dominant lactic flora (Samelis, 2000 ; Nychas et al, 2008). The dominant lactic flora responsible of the spoilage are *Leuconostoc mesenteroïdes* (Diez, 2009 ; Vihavainen, 2008) ; *Leuconostoc carnosum* or *Leuconostoc gelidum* (Vasilopoulos, 2008) ; *Enterobacteriacae, E. facecalis, C. divergens* (Vasilopoulos, 2008). Indigenous flora are also closely specific for a type of pork RTE product (Schirmer, 2009).

3. Quality indicators and models for chilled pork meat / Ready To Eat pork meals and frozen pork meat

3.1. Freezing process: an overview

Introduction: Freezing is a widely accepted preservation method used to store meat for relatively long periods of time. Utilizing frozen products rather than chilled offers the advantages of increased storage time, greater flexibility in inventory and greater product control. During the freezing process, the product temperature is lowered and most water in the food is transformed into ice crystals. The exact mechanisms by which freezing, frozen storage and thawing kills or damages microbial cells are not fully understood, although a number of studies have been conducted on the nature and sites. The color of frozen meat varies with the rate of freezing. There is a direct relationship between freezing rate and muscle lightness. The faster the rate, the lighter the product is. These differences in frozen meat lightness result from the dependence of the ice crystal growth on the freezing rate. Small crystals formed by fast freezing scatter more light than large crystals formed by slow freezing and hence fast frozen meat is opaque and pale and slow frozen meat is translucent and dark. However, freezing and frozen storage of meat can affect the structural and chemical properties of muscle foods, and **influence meat quality attributes such as thawing loss, colour and tenderness**. It is commonly known that, during the freezing process, intracellular juice is expelled by osmosis to the extracellular space forming ice crystals that later cause juice loss from meat during thawing.
Yu et al (2010) adopted a uniform design and three factors, i.e., freezing rates with two levels, thawing rates with six levels and source of sample (six pork M. Longissimus dorsi from three pigs), were investigated. After thawing, thaw loss (TL), cook loss (CL), water-holding capacity (pressing loss) and protein solubility were measured. Structure was also viewed using transmission electron micrographs and scanning electron micrographs. The results showed that freezing rates, thawing rates and individual significantly affected TL and CL. Eliminating the effects of fixed factors of different individuals, the effects of thawing rates and freezing rates on TL and CL were nonlinear. Significant interactions between freezing rates and thawing rates on both TL and CL were observed. Water-holding capacity was correlated to different animals and was independent of freezing rates and thawing rates. Thawing rates and animal had some effects on total protein solubility (TPS) and myofibrillar protein solubility (MPS); however, freezing rates had almost no effects on them. Considering the CL, TL, TPS and MPS comprehensively, fast freezing rate and slower thawing rates (0.75 cm/h) were the optimal combination.

Mortensen and al (2006) studied the effects of fresh meat quality (PSE versus DFD), freezing temperature (−20 °C versus −80 C°) and duration of freezer storage on changes in water mobility and distribution at intervals of 1–2 months during 10-month freezer storage of pork using. Fresh meat quality was found to have a significant effect on the amount of loosely bound water after freezing, which was reflected in a significantly lower cooking yield in PSE meat compared with DFD meat. While no significant changes in the cooking yield were observed with increasing length of freezer storage, a significant increase in the amount of loosely bound water in PSE meat with increasing length of freezer storage was found.
Hansen and al (2004) studied **the colour stability and development of lipid oxidation** during chilled storage for 6 days of chops from *M. Longissimus dorsi* produced from pigs with high (6.3) and low (5.5) ultimate pH (pH_u). The chops from the same individual pigs were either chill stored at 2 days post-mortem or after frozen storage for 30 months (pre-frozen). Initial redness was lower for pre-frozen chops than for fresh chops. Chops with the high pH_u had a stable value during chill storage, while chops with the low pH_u showed a rapidly decreasing for fresh and pre-frozen chops. In contrast, initial lipid oxidation, measured as TBARS (Thiobarbituric acid reactive substances), was similar for pre-frozen and fresh chops prior to chill storage for both the high and the low pH_u meat but developed most significantly in pre-frozen, low pH_u meat. Individual differences in colour stability and development of lipid oxidation between pigs were notable for pre-frozen low pH_u meat and need to be considered in quality control since meat from single pigs otherwise might give problems.

Complete flavor and texture profiles on pork loins from 118 carcasses revealed the following findings (Jeremiah LE and al (1990): (1) freezing, storing and thawing pork loins produced a slight tenderizing effect by making them less cohesive and easier to chew; permitted greater moisture release during chewing, thereby, slightly increasing the juiciness; resulted in well balanced and blended texture; but resulted in a less appropriate and well balanced flavor; (2) pork with normal muscle quality had a firmer, more elastic, and cohesive texture which was stringier, more fibrous, and harder to compress; (3) the PSE condition appeared to result in a drier texture with less moisture and fat being released during mastication and a greater amount of moisture being absorbed from the mouth; (4) the DFD condition appeared to result in a juicier texture with greater amounts of fat and moisture being released into the mouth during mastication and a softer texture which was less cohesive, fibrous, and stringy and easier to chew, but as the DFD condition became extreme the texture became excessively soft, crumbly and mushy resulting in a relatively large proportion of small mealy and/or mushy particles which detracted from the texture amplitude; (5) the PSE condition resulted in a predominance of sour notes which detracted from the flavor amplitude; and (6) the DFD condition resulted in a predominance of porky, sweet, and fatty character notes which enhanced the flavor amplitude but as the DFD condition became extreme, more character notes contributing to off-flavors were detected, a portion of which were apparently due to sex taint.
Present findings clearly document the influence of differences in inherent muscle quality on palatability properties of pork loins and aid in resolving the controversy that presently exists within the literature regarding the effects of freezing, frozen storage, and thawing on pork palatability.

3.2. Quality indicators and measurements

Undesirable storage conditions can lead to microbial, biochemical, chemical and physical quality changes in stored food. Several quality parameters governed by these changes are used to characterize quality of foods. This review briefly discusses the quality attributes commonly used to describe muscle foods (meat) quality loss during fresh and/or frozen storage.

3.2.1. Lipid oxidation – TBA and TBARS tests

The oxidation of lipids is the most important chemical change that occurs in stored muscle foods (Man and al 2000). The unsaturated fatty acids in triglycerides and phospholipids of tissue primarily autooxidise and various oxidative and non oxidative reactions secondarily take place (Powrie and al 1984). The reactions lead to the formation of volatile compounds such as

carbonyls, alcohols and acids which are responsible of the development of rancidity and off-flavours that persist even after the cooking of the products [Igene and al 1980, Man et al 2000].

Various methods have been developed to assess the extent of lipid oxidation in muscle foods [Gray and al 1992, Ladikos and al 1990]. The methods have been globally divided into those that measure primary changes and those that measure secondary ones. The oldest and the most widely used method appears to be the 2-thiobarbituric acid (TBA) test [Andersen 1990; Chen 1989; Syed 1993]. Advantageous and limitations of the TBA method for assessment of lipid oxidation in meat products have been extensively discussed by Gray and Monahan [Gray 1992]. The TBA test is an assessment method based on the measurement of the concentration of malonaldehyde on food, food extract, or steam distillates of food. Malonaldehyde is a relatively minor secondary lipid oxidation which reacts with TBA to produce a coloured complex with an absorption maximum at 530 – 532 nm. Measurements are done by spectrophotometric determination and the extent of lipid oxidation is reported as a 'TBA number' or 'TBA value' expressed as milligrams of malonaldehyde equivalents per kilogram of sample. However, the term 'thiobarbituric acid-reactive substances' (TBARS) is now commonly used in place of the TBA number or value [Cava 2009; Jakobsen 2000; Muela 2010; Vieira 2009]. This term assigns the extent of lipid oxidation in general rather than to quantify only malonaldehyde. Indeed, other products of lipid oxidation, such as alka-2,4-dienals, also react with TBA to form complex with the same absorption maximum as the malonaldehyde-TBA complex [Gray 1992]. The TBA method generally well correlates with sensory data even some limitations of the procedure have been pointed out [Gray 1992, Shahidi 1998]

Several factors influence the rate of oxidation of lipids. This rate increases with the degree of unsaturation of the fatty acids. For example, fish muscle contains high concentrations of polyunsaturated fatty acids whereas the major part of fatty acids in beef muscle is monounsaturated, the development of rancidity in frozen fish occurs at a more rapid rate than that in frozen meat muscle [Powrie 1984] . The storage temperature and time are also important variables for the rate of lipid oxidation. Frozen meat stored at temperature lower than -15°C do not show significant changes on the TBA value. But storage at -10°C and higher temperatures significantly increases the TBA number with time and temperature. Fluctuating storage temperature was found to increase the rate of lipid autooxidation much faster than constant temperature [Chen 1989] .

3.2.2. Colour changes

Colour is the primary factor influencing the fresh muscle purchasing decisions of consumers, but it is also important for frozen meat quality. Myoglobin is the main muscle protein responsible for colour with variable amounts according to the species. Colour changes generally occur during muscle food frozen storage due to thermal and/or photochemical autooxydation of the red oxymyoglobin to the brown metmyoglobin [Andersen 1989]. The oxidation reaction is affected by the storage temperature and time. Moreover, interactions between pigment oxidation and lipid oxidation also results on meat discolouration. The assessment of colour changes is often realized by the use of colorimeter with the measurement of the Hunter redness value (a^*), the lightness value (L^*) and the yellowness value (b^*). Chen at al. [1989] reported that the redness value of stored hamburger meat consistently decreased over storage time at storage temperature of -22°c, -18°C and -15°C, without significant difference between the three temperatures. The redness value decreasing rate is faster at storage temperature of -10°C and -5°C. Soldatou et al [19] studied colour stability of lamb meat product under refrigeration (4°C). They found a decrease of the redness value with storage time. The decrease in a^* value has frequently been associated with the formation and the accumulation of metmyoglobin on muscle surface during storage.

3.2.3. Protein denaturation – Texture changes

The denaturation/aggregation of muscle proteins, especially myofibrillar proteins, during frozen storage is an important factor of quality loss of meat. This process is accompanied by changes in texture of muscle foods such as hardening and with functional alterations such as liquid loss and changes in gelation properties. The denaturation mechanisms are mainly attributed to the removal of water and to the interactions of proteins with oxidized fatty acids but the microstructural modifications are significantly affected by the temperature and the duration of the storage. The proteins extractability and solubility are often assessed in order to determine proteins structural changes reliable to textural deterioration during cold and frozen storage.. Texture changes are determined by mechanical analysis by the way of shear force and elastic

modulus measurements or compression tests [Herrero 20005]. Authors reported an increase of the hardness and the elasticity, which reflect muscle toughening, with a higher storage temperature and prolonged storage time. They correlate the texture changes with the protein denaturation by a decrease in protein solubility, an increase in hydrophobicity and the formation of non-covalently and covalently linked aggregates.These findings are in agreement with those authors who reported a strong dependence of the rate of decrease of protein solubility with the duration of freezing storage.

3.2.4. Water holding capacity

Other related changes in proteins of meat are those that can be observed in the changes in water content in the water and which also lead to the loss of quality. In this respect, the water holding capacity (WHC) is regarded as an essential quality parameter which has been related to important attributes such as juiciness or tenderness. WHC influences the appearance of fresh and frozen meat and fish, and affects their sensory properties. WHC is generally quantified by three methods : (1) measurement of the thawing losses, (2) measurement of the cooking losses after cooking the sample until it reaches an internal temperature of 70°C and (3) measurement of the press losses using a filter paper press [Muela 2010, Vieira 2009]. In all methods, water losses were determined by difference in weight and were expressed as a percentage of initial weight. Obviously, in non frozen products, only cooking and press losses were evaluated. Other WHC measurement methods such as centrifugation and spectroscopy are used [Herrero 2008, Pedersen 2003]. It is accepted that there is a loss of WHC during frozen storage and that this loss of WHC is as more important as high storage temperatures [Chen 1989, Muela 2010, Syed 1993, Vieira 2009]. WHC loss is related to conformational transitions of muscle proteins, to changes in the structure of muscle water and/or to alterations in protein-water interactions [Herrero 2008]. Smith [Smith 1987] reported that myofibril gel microstructure changed from a continuous filamentous matrix to a globular matrix with decreased WHC during frozen storage of the turkey meat. It also can be influenced by the freezing rate, size and location of ice crystals [Syed 1993].

3.2.5. Odor: application of electronic noses

Odor deteriorative changes are able to occur at a rate governed by storage temperature, time and product composition. Sensory analysis is a long and costly process since it requires highly trained panels to minimize subjectivity. The odour perceived by the human nose is of great importance in consumers' acceptance of muscle foods. Therefore, it is not surprising that, in 1982, Persaud and Dodd [Persaud 1982] proposed the concept of an artificial or electronic nose and developed an instrument operating on a similar principle to the human nose [Schaller 1998]. Repeated efforts and extended researches have been made over the years and contribute to the enhancement of the technology. This makes electronic noses very useful for food applications in the field of process monitoring, shelf-life investigation, freshness evaluation and quality control. The use of electronic noses in quality control of muscle foods (meat) during fresh/frozen storage is developed.

a) Main volatile compounds (VC) associated with muscle foods bacterial spoilage

The spoilage of stored meat and fish products is largely due to the action of bacteria. The composition of the muscle foods spoilage flora is greatly influenced by the storage conditions, such as temperature and environment (type of packaging for example). Spoilage commonly manifests itself as off-odours and off-flavours caused by the presence of volatile compounds (VC) produced as a result of bacterial metabolism. Several studies have been dedicated to the identification of these VC and their specific role in producing a given off-odour (Freeman 1976; Stanley 1981; Pittard 1982; Dainty 1984/1985/1989; Mayr 2003; Stutz 1991). The correlation between the presences of specific volatile components with that of individual bacteria is now well established [Stanley 1981, Pittard 1982,Stutz 1991].
Authors [Dainty 1985] reported a defined time sequence of production of VC in naturally contaminated non-comminuted beef samples. Amongst those appearing first, and therefore with the greatest potential for early detection of spoilage, were the known end-products of *Brochotrix thermosphacta* metabolism, i.e. acetoin, diacetyl, 3-methyl1-butanol and 2-methyl-1-propanol [Dainty 1985], though their detection was rapidly followed by that of esters and of sulphur-containing compounds such as dimethyl sulfide and dimethyl disulfide.

b) Basic principle of the electronic nose

The concept of an e-nose is based on an odour stimulus which generates a characteristic fingerprint from the sensor array. Patterns or fingerprints from known odours are used to construct a database and train a pattern recognition system, such as artificial neural networks, so that unknown odours can subsequently be classified and identified. In a broader sense, electronic nose instruments are composed of three elements namely: (1) a sample handling system, (2) a detector system (the sensor), and (3) a data processing system [Peris 2009]. For each element, several techniques and technologies are used. The choice must be made with care, taking into account the type of sample, the volatile compounds and their relative molar masses, and lastly the ability to characterize complex mixtures.

c) Meat applications

Several studies, in the past decade, have been devoted to the use of electronic nose for quality assessment of meat and meat products during their storage: El Barbri et al. (2008]; Musatow and al 2010; Hansen and al 2005, Boothe and al 2002, Rajamäki and al 2006.

Applications of electronic nose as an instrument for the assessment of quality of meat have been reviewed. All reported studies demonstrate that the technique is able to evaluate the freshness or degree of spoilage of muscle foods. However, research developments are required in order to remedy to the multiplicity of sensor types and pattern recognition systems. The interpretation of the results is not always obvious and also need further researches. Regardless of these concerns, this method will not be used by ACTIA in the FRISBEE project. The need of the FRISBEE project is to utilize improvement methods directly sustainably.

3.3. Modelling of food quality changes

Modelling of quality deterioration during food processing and storage has been studied during the past several years [Labuza 1984, Saguy 1980]. The determination of kinetic parameters and mathematical models allow characterisation of the rate of deterioration reactions in foods and is one of the prerequisites for the improvement and optimization of the supply chain including storage and transportation [Rong 2009].
Modelling of quality loss of refrigerated and frozen muscle foods during storage has previously been investigated. As mentioned above, temperature is the main environmental condition leading to muscle quality degradation. The varying duration required for the storage and the transportation of food products is also an important factor. Therefore, modelling meat product quality requires a focus on both time and temperature.
The general equation describing quality loss is often given as:

$$\frac{dC}{dt} = -kC^n \quad (1)$$

where *C* is the quality index, *n* is the order of the reaction and *k* is rate constant.

The value of the order reaction *n* can be a fraction or a whole number. The order number is strictly an empirical concept, but when the stoichiometric equation represents the mechanism, the reaction order and the molecularity have the same value. Most reaction orders that describe food deterioration are either zero or first order [Labuza 1984]. Eqn (1), after integration, yields to:
Zero order loss kinetic reaction:

$$C = C_0 - kt \quad (2)$$

First order loss kinetic reaction:

$$C = C_0\, exp(-kt) \quad (3)$$

A zero order reaction behaviour has been observed for the discolouration and the WHC of hamburger meat stored at five constant temperatures between -5°C and -22°C for up to seven months [Chen 1989]. The colour changes measured both instrumentally and visually of frozen shrimp stored at variable temperatures (from -5°C to -15°C) during 12 months are also described by a zero order reaction. Chen at al. [1989] reported a first order reaction for the oxidation of lipid in hamburger meat. First order reaction has also been found by Chung et al.

[1991] for both the texture and the lipid oxidation changes of frozen scallop meats stored at temperature varying from -5°C to -30°C during a few months.

The rate constant *k* is temperature dependant. The most common and generally valid assumption is that temperature dependence of the deterioration rate follows the Arrhenius equation:

$$k = k_0 \, exp\left(\frac{-Ea}{RT}\right) \tag{4}$$

where k_0 is a constant independent of temperature, *Ea* the activation energy, *R* the gas constant and *T* the absolute temperature. Kinetic data at several temperatures over a fairly large temperature range are required in order to test the applicability of the Arrhenius model. The k_0 constant and the activation energy are generally derived from the slope of the plot of *lnk* vs *1/T*, if it is a straight line. Chung at al. [1991] found activation energies of 43.6 and 37.9 kJ/mol respectively for the texture loss and the oxidation of lipids of frozen stored scallop meats (from -5°C to -30°C). Higher value of activation energy of lipid oxidation (96.1 kJ/mol) is found by Chen et al. [1989] studying quality changes in hamburger meat during storage at temperature from -5°C to -22°C. These authors reported activation energies of 82.3 kJ/mol for meat colour degradation and of 28.8 kJ/mol for WHC loss. These results indicate that the rate of change in WHC is less sensitive to storage temperature.

If the plot deviates from a straight line, either the test methods were poor, or other reactions begin to become critical above some temperature and influence the reaction rate being studied [Nelson 1994]. In case of deviation from the Arrhenius behaviour, other models have been suggested in order to study the effect of temperature on the apparent reaction kinetics. For example, at temperatures near the glass transition temperature of the considered product, the kinetics of reactions associated with quality losses can be described by the Williams-Landel-Ferry model [Williams 1955] developed for polymers and nowadays also applied to foods that undergo glass transition [Nelson 1994]. The model relates food stability to the difference between the storage temperature and the glass transition temperature.

Instead of the activation energy concept, many researchers in the food field have used the Q_{10} approach for temperature acceleration [Labuza 1984]. Q_{10} is the ratio of the reaction rate at temperature (T+10°C) to that at the temperature T as shown in eqn. (5).

$$Q_{10} = \frac{k(T+10)}{k(T)} \tag{5}$$

It describes the increase in kinetic rate for a 10°C temperature increase. By holding the Arrhenius equation, the relationship between Ea and Q_{10} is given by eqn. (6) as follows:

$$ln\, Q_{10} = \frac{10\, Ea}{RT(T+10)} \tag{6}$$

By using an arbitrary reference temperature T_{ref}, the equation for the shelf life plot may be written as:

$$t_s = t_{ref} \, exp\left[-b(T - T_{ref})\right] \tag{7}$$

Then substituting into eqn. (7) for shelf life at T and T+10, one gets:

$$Q_{10} = exp(10b) \tag{8}$$

where $b = Ea/RT(T+10)$ is the slope of the shelf life plot. The parameter Q_{10}, also called accelerating factor, is thus strongly dependant on temperature and so if it is reported the temperature range for which it applies should be mentioned. Chen at al. [1989] determined Q10 values of 5.68, 4.43 and 1.68 respectively for lipid oxidation, discolouration and WHC for frozen hamburger meat stored at -5°C to -22°C.

Fluctuation of temperature often occurs during food storage and needs to be taken into account by the modelling. Earlier studies in the field of the effects of temperature cycling on food quality loss during storage have been done with an erroneous consideration in the order of the

reactions. Indeed, these studies assuming zero order reactions, considered that a distribution could be divided into a set of constant temperature regions and then that the quality fraction consumed (defined as the amount of the measured quality index which has been used up to the amount of that quality which could be used up) can be summed up for each time period according to the Time-Temperature-Tolerance (TTT) method [Labuza 1984]. However, for first order reactions, since it involves a log function, the ratio is not the fraction of quality index consumed. On the basis of these earlier studies, Labuza [1979] developed kinetics models for both zero or first order reaction using the general kinetic equation in differential form :

$$\int_{C_0}^{C} \frac{-dC}{C^n} = \int_{t_0}^{t} k\, dt \qquad (9)$$

If the cycling pattern is not regular, eqn. (9) should be used and the quality loss summed up for each time-temperature segment as in the TTT method. Using this equation, the food kinetic group at the University of Minnesota has conducted studies of quality loss for a series of different food products stored for up to one year under square wave (eqn. 10) and sine wave temperature distributions (eqn. 11).

$$T = T_m \pm a_0 \qquad (10)$$

$$T = T_m + a_0 \sin\left(2\pi \frac{t}{t_p}\right) \qquad (11)$$

where T_m is the mean temperature of the cycle ($T_m = (T_{upper}+T_{lower})/2$), t_p is the period of the cycle and a_0 is the amplitude magnitude ($a_0 = (T_{upper}-T_{lower})/2$). The group developed the following equations for the theoretical rate constants.

Square wave:

$$k_{sq} = \frac{1}{2} k_{T_m} \left[exp(a_0 bZ) + exp\left(-a_0 bZ'\right) \right] \qquad (12)$$

Sine wave:

$$k_{sine} = k_{T_m} \left[1 + \frac{(a_0 b)^2}{2^2} + \frac{(a_0 b)^4}{2^2 4^2} + \frac{(a_0 b)^6}{2^2 4^2 6^2} \right] \qquad (13)$$

where $Z = (T_m+10)/(T_m+a_0)$, $Z' = (T_m+10)/(T_m-a_0)$ and b is the slope of the plot from eqn. (7).
in this case where the cycling temperature distribution is known, one can use the eqns. (2) and (3) according to the reaction order by substituting in them one of the above rate constants (k_{sq} or k_{sine}) for k. Chen et al. [7] applied this model to calculate the accumulated amount of deterioration of meat samples stored at fluctuating temperature following the square wave distribution (T_m = -13.84°C). They found smaller relative errors by comparison of the predicted and experimental results for Lipid oxidation (first order reaction) and colour and WHC change (zero order reaction).

The food supply chain includes various time/temperature histories for each storage and transportation period. The modelling must be achieved for a global prediction of the quality level of a product for the whole supply chain, or at a certain location in the distribution network. Considering an initial quality index C_0, and subsequent storage periods $i=1,\ldots,m$ with time interval t_i and degradation rate k_i depending on the temperature T_i, eqns. (2) and (3) lead to [Rong 2009] :

Zero order loss kinetic reaction:

$$C = C_0 - \sum_{i-1}^{m} k_i t_i \qquad (14)$$

First order loss kinetic reaction:

$$C = C_0 \, exp\left[\sum_{i=1}^{m} k_i t_i \right] \quad (15)$$

This kind of model can be used in the optimization of the supply chain combining food quality modelling in production and distribution with logistics models [Rong 2009].

In order to globally quantify the quality degradation of a food product during storage, Achour [Achour 2006] has proposed a single quality index. The concept is based on the definition of a Global Stability Index (GSI) varying between zero and one and taking simultaneously into account the time variations of all pertinent quality indices proper to the food product under study. This index is formulated as shown in eqn. (16):

$$GSI_j = 1 - \sum \alpha_i V_{ij} \quad (16)$$

where V_{ij} is a variation term defined as the ration of the variation of the quality criteria to the variation with the threshold value:

$$V_{ij} = \frac{C_{ij} - C_{i0}}{L_i - C_{i0}} \quad (17)$$

C_{ij} is the measured value of the criterion *i* at time *j* units; C_{i0} is the initial value of the criterion *i* at the start of the experiment (t=0); L_i is the threshold value of the criterion *i* set by regulations or common practices. As the *GSI* is closer to unity, the product is more stable, meaning its initial quality is being well preserved. The modelling approach described above can be applied here substituting the single quality index *C* by the *GSI*. This index enables one to judge the degradation of the microbiological, the physicochemical and the sensory quality of the food product during storage in a global fashion. This method have been successfully applied to the study of the stability of an orange-based carbonated beverage stored at various temperatures [Achour 2006] and to the quality assessment of butter lettuce during postharvest storage [Ansorena 2009].

4. Conclusion

The D. 3.2.4.2 is a review on chilled Ready to Eat pork meals and frozen pork meat. For chilled RTE pork meals, the safety attributes are "*Listeria monocytogenes* and spoilage lactic acid bacteria" and quality attributes are "texture and odor". For frozen pork meat, the quality attributes are "texture and odor". This review D 3.2.4.2 describes the large number of publications/data and models of pork meat and pork meat processed safety and quality indicators. The strong dependence of the safety and quality evolution of food on the time/temperature history was also identified.

This compilation will be completed with carefully designed experiments on the different foods using experimental facilities from partners ACTIA (ADRIA and Aérial) and works on model developments for FRISBEE in interaction with WP3 leader, partners and Coordinator Cemagref. ACTIA has identified necessary new data and/or models to develop inside the Frisbee project.

These experiences are necessary for the construction of a deterministic model, the aim **of which** is to quantify whether the chain cold may have an impact
-on the development of the bacterial composition and quality changes of chilled Ready to eat pork product
-on the quality changes of frozen pork meat.

The experimental design will be proposed by ACTIA and validated by WP3 leader before the beginning of the trials. Kinetic data at several temperatures over a fairly large temperature range are required in order to test the development and applicability of the models and in order to study the effect of temperature on the apparent reaction kinetics. The objective is to study the

degradation of the microbiological, the physicochemical and the sensory quality of the food product during storage (T°C/ duration).

Two food types have been identified as model systems relevant to chilled pork and RTE pork meals:

- Type 1 : raw, salted and smoked ham like bacon; packaging : modified atmosphere.
- Type 2: pasteurized ham and pasteurized pate; packaging : modified atmosphere.

The process of cooking has been demonstrated to be sufficiently destructive to the Gram-positive and Gram-negative foodborne pathogens and several major background flora like lactic acid bacteria. Industrially-produced cooked ham and pate products are exposed to the environment post-lethality and handled prior to final packaging. The risk of recontamination and subsequent growth must be addressed. Raw bacon has been shown to *contain L. monocytogenes* (Angelidis and al 2006; Uyttendaele and al 1999). Raw salted, smoked ham is retain because able to be consume, like a part of a chilled prepared sandwhich, deli-salad.

RTE pork meat:

Experiments (Autumn 2011) will be undertaken into static conditions of temperature (different levels of T°C ; range betwwen 2 and 8°C). **Kinetics data** will be acquired at each T°C level for:
* *L. monoctogenes, (contaminated artificially by challenge tests) and lactic acid natural flora*,

** relavent quality indicators : drip-loss, water holding capacity, colour, hardness.

pH, aw, atmosphere are fixed parameters and will be measured at 3 days during the duration of storage. For natural lactic acid flora, a relevant level of 5 log UFC/g of product will be the hypothesis of the potential spoilage of the product. Two dominant strains of natural lactic acid flora will be isolated during theses assays, representative of the two types of chilled processed pork meat. The cardinal values of parameter T°C will be acquired.

Frozen deboned pork meat:

Experiments (Autumn 2011) will be undertaken into static frozen conditions of temperature (4 different levels of T°C). Thawing control conditions will be applied with preservation of the sensorial characteristics. **Kinetics data** will be acquired at each T°C level for relevant quality indicators : lipid-oxydation, drip-loss, water holding capacity, colour, hardness.

Development of models which will describe the safety and quality of RTE pork meat and the quality of frozen pork meat will be realized. Cardinal type odel is favoured as a basis describing safety of RTE (*L. monocytogenes* and spoilage lactic flora behavior) for the further development of simulation model in the FRISBEE project. The review investigates a basis of several models of quality deterioration during storage: they constitute the basis which will be validate in FRISBEE context (identified experimental kinetic data, parameters estimation).

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