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THE RISK EVALUATION OF BACTERIOLOGICAL CONTAMINATION IN A MEAT PRODUCT FACTORY UNIT

- SUMMARY OF PhD THESIS -

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SUMMARY

Following Romania's joining UE, meat producers from our country were confronted with a series of new problems, both legislative and infrastructural, regarding food safety. Due to these modifications, some wouldn't be able to face the new challenges; other tried to keep up by making big material efforts in order to remain on the market. It's obvious that, without these financial efforts either the legislative demanding or the customers claims whose nutrition tastes are becoming more and more refined, couldn't have been satisfied.

The researches of this master's degree thesis are motivated both by the necessity of implementation of new legislative framework in food safety domain and by the meat producers wish to obtain quality, salubrious, microbiological agents free products.

The thesis is divided into two parts: the bibliography part covering 70 pages and the personal research one covering 90 pages. The data presented in the thesis are sustained by a number of 30 tables, 5 charts, 30 figurative images representative for the conducted study.

The bibliographical lists 218 headlines from the national specialized and international literature, articles from the Internet, data from my own scientifically papers and food safety legislation.

First part – Bibliographical study contain the introduction that describes nutrition and the present stage of knowledge regarding the meat and meat products role as a source of microorganisms, the quality criteria of meat and meat products, and also the importance of implementation of food safety systems.

This first part is structured in a number of 4 chapters and 20 subchapters through which are presented information from the recent specialized literature and from the actual legislative framework regarding the subject of thesis, data that lay at the foundation of interpretation of the results obtained after the conducted researches.

In the first chapter "The importance of the meat in food" is presented data concerning the meat importance in people's nutrition, because it's the most important proteins source, releasing all the amino acids indispensable for our metabolism.

By its energetic and molding action, meat has a secondary role comparing with the vegetal organisms. Due to the low contain of glucides and mineral substances, meat is not a complex food. Besides that, it's an easy spoiling food, it may cause food poisonings but it still remains the most required produce in most of the countries. The progress recorded in the last decades regarding the processing, preserving and manipulating technology of meat didn't exclude only partially the possibilities of contamination with alteration or pathogen microorganisms.

The consumers' demands regarding the microbiological quality of meat have increased more and more. Buyers are more careful when buying meat, especially to alteration phenomena, to odor, to flavors, to color. We may say that nowadays the microbiological quality of meat has become a factor of great importance in its marketing.

The second chapter named "Meat and meat products role as a source of microorganism" emphasizes on the germs causing meat spoiling and on the main food poisonings produced after meat and meat products consumption.

Meat spoiling and bacteria causing diseases are controlled by observing the hygiene rules throughout the entire preparing and preserving process.

Industrial altering bacteria are represented mainly by microorganisms with best growing temperatures < 5°C, psihrotrophings. The alteration level dependents on the microorganism capacity of producing organoleptical modifications.

Food poisonings don't appear by simple touch of humans with the contaminated food only after its consumption. Living agents or their toxins contained in the food would cause illnesses to consumer only if they are in large quantities and only if they are introduced orally. In order to fulfill these conditions microbes must multiply on the contaminated food, must reach high concentrations or in some cases to produce large quantities of toxins and the consumer must eat large quantities of the contaminated food.

The 3rd chapter "Quality criteria in organoleptical, physic-chemical and microbiological control of meat and meat products" mentions aspects regarding the organoleptical analysis of physic-chemical indices for quality, the hygienical-sanitary and microbiological markers, aspects that are necessary in evaluation of meat and meat products quality.

Microbiological exams of food products establish the presence or the absence of their microbe toxicity for consumer and the preserving capacity of products under the given conditions. The exams have a sanitary and economical signification with great implications in actuality when the results are reflected on large quantities of products.

In order to assure the microbiological quality of food one must use surveillance and control system based on the application of the preventive measures. In many countries a series of measures is applied during the producing process that assures the obtaining of quality products. These measures are gathered into the so-called "Code of well practice and distribution" (GMP – Good Manufacturing and Distribution Practices) which was later included in HACCP.

As soon as preventives principles are established and applied in food producing the microbiological exams becomes more efficient and well functional. The rigorous control of the raw material sources reduces a lot the fluctuations in microbiological quality of food and assures the uniformity of quality.

The 4th chapter named "Food safety systems" refers to the appreciation and the importance of food safety system implementation, emphasizing on HACCP, an efficient system for controlling and self controlling of food hygiene. This system is recommended world wide both, by the European legislation and by the national one. This preventive control system has as main goal the identification of the critical control points where the products may suffer depreciations, their remedy by special control measures and monitoring the maximal limits in every critical control point.

Food operators have the obligation to identify the source that has provided a food product relying on some producers, systems, recording and also to check the destination for products. This possibility to identify and to follow up through the entire production stages and distribution represents, in fact traceability. This traceability helps also the operator in case of a inadequate product and initiates the procedures of withdrawal of the product out of the market, informing about it. Although the final choice belongs to consumer, the producer's task is to inform correctly the consumer by a label contains all the identification elements of the product.

The Second part "Personal researches" has a number of 6 chapters with 24 subchapters where are presented the theme motivation, the research objectives, the used working material by observing the methodology and the working techniques.

The 5th chapter "Implementation of European legislation in analyzed unit" respectively the first chapter of personal research, describes the way of implementation of the provisions stipulated in "The New Legislative Packet" emphasizing on the structural aspects of the studied unit, on its endowment and necessary equipments for its activity, on the importance of supplying with quality raw materials and on hygiene control.

The used materials in analyzing the way of achievement the new legislative requirements, for the alignment of the studied unit to European standards, consisted of studying and modifying the initial unit project, respectively the actual one, and researching the archive documents that had showed the evolution of unit whose transformation was possible by obtaining European funds.

The safety standards apply both to food products made in UE and those originated in other countries. Legislation covers multiple areas, from fodder and nutrition, to animal, nonanimal food, food animal feeding etc. Thus, product trade may carry on freely, on condition that the products come from authorized units on communitarian trading and that the products are not under UE restrictions for a certain state member.

The communitarian legislative framework regarding food safety is common to all state members. In order for Romania to become a state member and to launch it on the sole European market, imposing radical measures to food industry was necessary. The measures were imposed by the National Veterinary Sanitary Agency for Food Safety. The measures contain reorganizing and modernizing programs that allow units to ally to European standards, either structural, or legislative on food safety background. As a result, Romania negotiated with UE previously from joining and obtained a transition period. At the end of this period, all units that have developed reorganizing and modernizing programs should fulfill the sanitary-veterinary conditions in order to be authorized to do communitarian trade.

The unit under study in this thesis is one that in 2006, year before joining, didn't accomplish the UE demands in a structure respect, but fulfilled the hygiene conditions to obtain safe products. The goal of unit was to be authorized in beef and pork and also in manufacturing small varieties of meat products.

After the closing of the slaughter activity at the end of 2006 by the sanitary-veterinary authorities because the unit didn't meet the sanitary conditions anymore, the managers analyzed the current situation. They performed a study and they asked for a transition period till the end of 2009 and for listing the unit in the "C" list, attaching the necessary documentation and the guarantee that these would be made by private financing.

For all the "C" category units the reorganizing and modernizing programs were approved also by the European Community and the units were enrolled on the "transition period list". They are monitorised by sanitary-veterinary authorities in Romania and are controlled by the European commissionaires during the visits conducted by the DG SANCO.

Following up the material efforts on the manager's side during the last years, the implementation, observing the new legislation and alignment to European standards has been achieved successfully in the analyzed unit. The fulfillment of the legislative requirements by implicating the legislative requirement by implicating the entire personnel and the accomplished modernizations has lead up to obtaining the sanitary-veterinary authorization for intercommunity trade. We must add that, for the moment, the unit isn't involved in intercommunity trade.

The 6th chapter is "Following the drawing up and implementation of HACCP system". In this chapter the food safety system was analyzed mainly the one implemented in the unit, emphasizing on the correctness on the plan drawing out, on the correct associated risk evaluation, on the final product obtained and on its efficiency on keeping under control the physical, chemical, biological potential risks.

The HACCP system compels on the rigorous respecting of the preventive principles. Disobeying these principles in any of the producing process puts at risk the entire system.

The analyzed unit has started implementation of the HACCP system at the end of 2006 and obtained the certification in March, 2007.

The documentation was drafted by a consultancy firm, when the firm policy was reorientated and its managers aimed at the ISO 9001/2000, ISO 22000/2005 certification, besides obtaining of sanitary-veterinary authorization in intercommunity trading. Recertifying the system is to be made in 2010.

The materials we used in following up the implementation mainly consisted of studying the documentation elaborated by the unit together with drawing up of the system, and of comparing it with the HACCP demands.

Our analysis followed up aspects referring to:

- formulating quality policy of the firm;

- forming the necessary team to assure the functioning of the system;
- identification of the potential hazards and evaluation of the risks;
- determination of the critical control points and establishing their limits;
- establishing the necessary monitorization and the corrective actions to be applied in case of inadequacy.

The studied unit wishes to satisfy permanently the customer regarding the offered products and services. For this, the unit proposed itself a multidirectional policy that was shared and understood by the entire staff through periodical trainings. The minutes of the meetings and the well-established themes can certify that.

The policy of the studied unit is credible and is known by all the clients and suppliers; it's posted at the entrance to be read, so that the aimed objectives are well known to all who are interested.

From the system analysis of documentation we've observed that the HACCP team has identified correctly the product addressability, mentioning the sensitive consumer categories, indicated in the technical specifications of the final products. These details are not present for the time being on the labels.

In identification and evaluation of risks it was taken into consideration during stages of all the technological flow the impact of the raw material ingredients, drinking water, hygiene practices, fabrication processes in controlling the possible risks but also of the consumer categories of the final products.

HACCP team demonstrated that the risk analysis, the taken decisions and the control measures are justified by several procedures and detailed specifications that assures an effective implementation of the system.

In the analyzed unit I concluded that the potential risks are kept under control due to proper implementation of HACCP system by a correct management of food safety. This fact is demonstrated by the control reports and by the positive feed-back from the consumer's part. Communication within the team was an important factor in the good implementation and functioning of the HACCP system. This way, a permanent connection was assured between all the involved compartments for implementation action, as well as between team and HACCP team.

The working material for the last three chapters consisted of representative samples of minced meat, prepared meat and heat-treated meat products, sanitations tests of the working surfaces, tools, equipments and water drinking source.

Because the quality of water is very important, concerning the food poisoning microorganisms that can survive days even months in water, in the 7th chapter" Assessing the level of bacterial contamination in the drinking" I studied the level of bacterial contamination by watching the evolution of three microbiological criteria (total number of total aerobic count germs, coliform bacteria and E. coli, as well as intestinal enterococci).

These bacteriological analysis of water were conducted both on the public water network (used in all the technological process flow, including hygiene) and on the private water source (private well) used only in controlling the fire and washing the garbage platform.

Mezofilia germs, that develop at 37°C and represent about 30% from the total germs number, were chosen as a drinking mark because it is considered that between their number and the probability of pathogenically germ presence in humans and animals there is a positive correlation. They are counted in the whole as a total number of germs (colonies forming units) and are reported to 1 ml of water. In order to determine the total number of germs it was used the method according to reference standard SR EN ISO 6222/2004, colonies countering method by sowing at the agar surface. TNGM is a good mark, but it's also relative because there were situations when its values were good but infections (epidemics) appeared. Because of that, more precise markers were imposed, like feces pollution markers, because the majority of hydrical diseases have as etiological agents, germs eliminated through digestive duct. In order to detect

and count the coliform bacteria and E. coli it was used the method according to reference standard SR EN ISO 9308-1/2004 and the working technique is based on using the filtration system on Microfil membrane in microbe contamination control of liquid samples. Because it is considered that enterococci don't suffer a too big variability in time and are particular for humans and animals, they supply data on the polluting source. Enterococci are found in feces in a more reduced number than coliform bacteria. Still comparative to them, enterococci are more resistant in external environment and suffer to a lesser extent variability phenomena.

In order to identify and counting the intestinal enterococci it was used the method according to reference standard SR EN ISO 7899-2/2002.

For the network water the obtained values between January, 2007 and March, 2009, for all three markers, were relatively invariably and within reasonable limits that was expected because water comes from a drinking treated source.

The microbiological analysis results performed on the water from the well, between January, 2007 and March, 2009 the uniform increasing and decreasing of two analyzed sanitation markers coliform and enterococci were visibly influenced by the harvesting season, exceeding thus the acceptance limits.

Because nowadays consumers are becoming more and more aware of the hygiene aspects of food, it's becoming mandatory that all the food producers respect both technological demands and hygienic-sanitary ones. That's the reason why, in 8th chapter "Assessing the bacterial contamination level of surfaces" it was studied the hygiene in the meat products factory in order to determine if this could become a microbe contamination source for the products made within for the final consumers.

The necessary microbiological analysis for the establishing of the bacterial contamination level followed microbiological markers: total number of aerobe mezofili germs, the number of enterobacteria, the presence of the coliform bacteria.

The working method used in determinations of the total number of aerob mezofili germs was according to standard 4833/2003. In order to determine the number of enterobacteria it was used the method according to standard 8523/1991, and to determine the number of coliform bacteria it was used the method according to standard 4831/1992.

There were analyzed the working surfaces, the equipments and instruments with which raw material comes in contact during the technological process, the surfaces that can become an important contamination source of the obtained products if during the hygienically proceedings in the meat preparing units are superficially and wrongly performed. The accent laid also on the results of the sanitation analysis inside de refrigerating vehicles used to carry the meat products made inside the unit. The hands of the personnel working on the technological flow were also analyzed. Totally there were gathered and analyzed a total of 110 samples, of which 63 in 2007, 35 in 2008 and 18 in 2009. Of the 110 samples, 6 have shown values above the accepted limits for TNGMA, the rest of the analyzed samples having corresponding results.

Spice microbiology has a big influence on the quality of the final products, because they can contribute to the total microbial load, so that in the 9th chapter "Establishing the level of microbiological contamination in spices" 22 spice samples, were analyzed, spices for mici, sausages, burgers, salami, all samples harvested during the strategically programme. The samples were analyzed in the county laboratory. Intended microbiological markers were: yeasts and molds, coagulaza-positive, staphylococcus, Salmonella, E. coli and B. cereus.

Working methods were according to standards so, for the counting of viable yeast and molds in spices it was used the colonies counting technique, at 25°C, according to SR EN ISO 7954/2001, for the B. cereus determination it was used the technique according to SR EN ISO 7932/2005. Coagulaza-pozitive staphylococcus are bacteria that from tipical colonies on the selective agar medium with rabbit plasma fibrinogen, and the method used for counting coagulaza-pozitive staphylococcus was performed according to SR EN ISO 6579/2002. In order to determine E. coli marker in spices, in 2007, 2008, it was used the method according to SR EN ISO 7251/1996, in 2009 it was used the one according to SR EN ISO 16649-2/2001.

The obtain results after performing the analysis were equal to the above mentioned standards and the meaning of those values shows that in the analyzed unit there weren't any inconsequences of spices used in meat producing recipes, and that spices don't represent potential dangers for the final products.

In the 10thchapter "Risk assessment for bacterial contamination of the finished products" safety criteria (used in evaluating the safety of the product or product lots) and the hygiene criteria (used to demonstrate that the producing process works accordingly) were studied.

In order to observe the NTGMA marker in minced meat produced in the unit, a total of 51 analyses were conducted, out of which 29 were conducted in 2008 and 22, in 2009.

For determining E. coli in minced and prepared meat, 51 samples were analyzed, representing 255 samples for each of two types of products.

The analyses conducted to determine *Salmonella* in minced meat involved harvesting and analyzing 60 samples. The meat products made in the unit that were subjected to heat-treatment during the technological flow were analyzed to determine *Salmonella*. Thus, 21 samples had been harvested, 11 of them in 2008 and 10, in 2009.

Out of meat products that were heat-treated during the technological flow to determine *Listeria monocytogenes*, 21 samples had been harvested, 11 of them in 2008 and 10, in 2009.

The working method used to determine NTGMA in minced meat was performed according to SR EN ISO 4866/2003, by deeply sowing of a defined medium, with a specified quantity of the sample. To determine *E. coli* in minced and prepared meat, it was used the method that establishes the number of β glucuronidase-positive *E. coli* by colonies counting technique, raised at 44°C using 5bromo2-cloro-3indol β-D-glucuronat, according to SR EN ISO 16649-2/2001. The principle of the method and the necessary endowments for isolating and identifying *Salmonella* germs types were the ones according to SR EN ISO 6579/2002. For the isolation and the identification of *Listeria monocytogenes* in meat products it was used the method according SR EN ISO 11290/2002 and the samples were harvested from the final product storage, labeled products ready to be delivered.

The monitorized and analyzed microbiological parameters have recorded normal values, the probed products being allowed for selling and consuming. As a consequence, on evaluating the final products obtained in the analyzed unit, we can conclude that they are not risk products for the consumers' health.

Also, germs with pathogen potential weren't identify during the analyses, so the final products are salubrious and don't endanger the consumers health.

General conclusions and recommendations

Romania's accession and, later, integration in the UE have brought a lot of obligations for the food processors, apart from the given opportunities. Due to these obligations, many processors embraced the challenge and started a series of modernizations that targeted building some units where the technological flow matches the European standards.

In the meat factory under study, in order to obtain the authorization for intercommunitary trading, reorganizing measures should have been taken concerning:

- the improvement of production infrastructure: buildings, access into unit, technological flows, water sources, refrigerating storages, means of transport for the final products etc;
- training the entire staff of the unit;
- implementation of food safety systems;
- starting the productive process in the new arranged areas, supervising and improving the system whenever it was necessary in order to obtain approved products.

The main goal of extension and modernization, by implementing the legislative demands was the one to adapt the already existent building considering all the legislative requirements in

the structural preoccupation was to prevent that the main contaminants was to contact with products, causing alteration to the final product.

Regarding the improvement of the producing infrastructure by alignment to the European standards, this was successfully performed. Thanks to the sustained efforts in the last years, the unit has been modernized with European funds.

All the modernizations and the fulfillment of the legislative demands have as a result the obtaining of the sanitary-veterinary authorization of the unit for intercomunitary trading. Although at present, the factory isn't doing such trade, it is committed to maintain the European standards by respecting the legislative framework. By doing that, the unit may trade, if financial opportunities may come.

The entire staff that work and manipulate food (inclusive the temporary personnel) is trained periodically, according to a schedule, regarding food hygiene, to a level that corresponds to the activities they have to perform in the section where they work. The beneficial effects of those trainings have brought their contribution to the favorable situation of the unit, meaning that there weren't any events that lead to food poisoning episodes.

In the analyzed unit the food safety system HACCP operates by good hygiene and productions rules, training procedures, appropriate professional training, discipline and diligence, a good communication, a correct and complete recording system and PCC monitoring.

The development procedures of HACCP plan in the analyzed unit are certified by the quality and safety assurance system, the unit having implementated the food safety system, ISO 22000, that relies on the previous HACCP programs combined with ISO 9001.

Although at first the implementation of the system and the certifying process seemed to be simple, they required a lot of resources (human, financial, time), reorganizing and the most difficult thing was to keep this certification because this system of safety managing must be maintained and continuously improved.

Referring to risk evaluation of microbial contamination during the technological process, the efforts were directed to monitoring the water sources, the technological surfaces, the used spices and the final products obtained in the producing process.

The result of the microbiological analyses performed on the well water showed that the relatively uniform increasings and decreasings of two analyzed sanitary markers (coliforms and enterococci) were visibly influenced by the harvesting season, exceeding the admissible limits.

The appreciation of the microbiological contamination level by sanitation testing results of the surfaces that come in direct contact with the products showed that decontamination performed into the meat unit is able to keep under control the hygienically-sanitary markers.

The obtained result means that the spices haven't represented dangers for the final products by their use as ingredients in the composition of the products.

I would recommend a special attention to the following aspects (that don't influence negatively the quality and sanitation of the products, buy may become a real danger):

- Because the manipulation of the carcasses from the reception area to the storage zone is made manually, a special, attention must be granted to avoiding contact with walls, doors and poles. For this, every half pork or beef carcass must be handled individuality because the reception line is not an automatic one.
- The inclosing of the unit, made by a wooden fence, with a partial concrete pedestal, may allow digging of galleries beneath the fence, causing a possible disease spreading. We consider necessary the completing the concrete pedestal on the entire length of the fence.
- Till now, the implemented HACCP system has been monitorized constantly and observed periodically by an audit team from the organization that validated it. I recommend more attentions to be given regarding the legislative alterations in food sector, alterations that were made lately. In the same time, I recommend

maintaining and improving continuously the system in order to obtain safe final products.

- With only one drinking water source (from the public network), in case of malfunctions the unit may remain without water. Also, during summer time, when domestic consumption of the residents in the neighborhood is increased, the water debit inside the unit is low and it's difficult to assure the amount of water in order to carry on the technological process in good conditions. There is an undrinking water source, but this one is used only for putting out an eventual fire, for washing the sewerage, the garbage platform and the yard. It is necessary a private drinking water source, as a backup, to be used in case of an emergency. This source must provide water in sufficient quantities, either for carrying out the technological processes or for satisfying demands in certain moments (during cleaning the factory). At the moment, this aspect is performed by using the aquaPUR tank, with a capacity of only 750l.
- The procedure regarding staff access is not respected by the entire personnel. They don't always change the working equipment with normal clothes, before going to the cantina, as it is mentioned in the procedure, this may cause a crossed contamination.
- Because, by analyzing the recordings, we observed two unconformities regarding the disconnecting electrical supply, I recommend the acquirement of a generator to assure the energy source in case of malfunctions.
- The endowment of vehicles used in transportation of meat products allows only showing temperature not recording it as heat chart, fact that we strongly suggest to be improved in future.
- I also recommend creating a private hygienic and disinfection station for the transporting vehicles, because there is none at present. The hygiene of the refrigerating vehicles is carried out by a car wash that serves the unit. Operating on a contract, this car wash doesn't produce a disinfection of the means of transport, too.

The obtained results in this study are similar to ones published by different authors that conducted similar researches.

The results emphasize on the quality of the final products, they appreciate the importance of a hygienically way of processing, storage and transport in the meat units and they appreciate the necessity of implementation of a food safety management so that the obtained final products can be sold to the final consumers only after their quantity certification, mainly by the producer and secondarily by the state's authorities that have control and guidance tasks of a great importance.

In the thesis there are own data published in the four scientifically papers (two of them as an author, two in collaboration with a specialist in food microbiology and sanitary-veterinary expertise of food from the county Laboratory, Vâlcea), papers presented during scientifically meetings, throughout the symposiums organized by the University for Agricultural Sciences and Veterinary Medicine in Timisoara.