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THE EFFECT OF OREGANO ESSENTIAL OIL ON MICROBIAL LOAD, DRYING KINETICS AND SENSORY ATTRIBUTES OF DRIED MEAT

A Dissertation Presented

by

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DECLARATION

I, Helga Johana Hernández, declare that this thesis is my own work unless otherwise referenced or acknowledged, submitted for Ph.D., in the Faculty of Tropical AgriSciences of the Czech University of Life Sciences Prague.

In Prague, August 1st, 2016 Helga Johana Hernández, MSc

ABSTRACT

Microbial load can be controlled using either synthetic or natural preservatives. Particular interest has been focused on the potential application of plant essential oils as safer additives for meat. However, there is no published research on the use of essential oils during the meat drying process. This study was focused on enhancing the meat drying process. At first a value-added dried meat product by using oregano essential oil (OEO) to inhibit the growth of bacteria, and the sensorial response from assessors is presented. It was found that the application of the OEO in meat is effective in inhibiting *Salmonella enteritidis* and *Escherichia coli*. After 6 hours of drying at 55°C, 2 ml (0.038 ml l^{-1} air) and 1.5 ml (0.028 ml l^{-1} air) of OEO were considered the minimal inhibitory concentrations (MICs) against *S. enteritidis* and *E. coli*, respectively. Samples treated with 0.75 ml of OEO were more attractive for consumption compared with the control; at a higher concentration of OEO, the sensory quality of the food was affected.

Next, the effect of modified blanching treatments on the drying behaviour of beef meat was evaluated by determining moisture ratio versus time curves and the influence on sensory quality of the resulted product. The 3 treatments under investigation were (1) oil treatment (2) steam blanching and (3) hot air blanching with 3 doses of oregano essential oil (1) 1.5 ml (2) 3 ml and (3) 6 ml. Each treatment had an effect on the drying time of the beef samples, however, the dose of oregano essential oil applied did not affect the drying process. The results showed that steam blanching was very effective reducing the drying time. Meanwhile, 1.5 ml and 3 ml hot air blanching samples and 1.5 ml oil treatment samples were judged as better from sensory point of view and the respondents considered that adding oregano essential oil enriched the pleasantness of the smell.

In conclusion, a value-added dried meat product obtained by using oregano essential oil to enhance food safety received an acceptable sensorial response from consumers. Additionally, each modified blanching treatment tested influences the drying kinetics process, but the dose of oregano applied did not affect the drying process. In this sense, hot air blanching and oil treatment with the lowest dose had an acceptable sensorial response from consumers.

Keywords: oregano essential oil, meat drying, microbial load, meat sensory attribute, drying kinetics, modified blanching treatment.

PREFACE

Drying as a preservation food process has been used since ancient times. Drying is used in making a variety of processed meat products. The goal of drying meat is to boost its shelf life and its characteristic flavour is one of the important properties which are appreciated by consumers. However, the risk of food-borne illnesses from poorly dried meat such as salmonellosis and infestation by *E. coli*, presents a great hazard, which is a very important reason to control the microbial load in dried meats.

Microbial load can be controlled using either synthetic or natural preservatives. Besides, safety, consumers progressively demand use of natural products as alternative additives in foods for improving the product quality. Particular interest has been focused on the potential application of plant essential oils such as oregano essential oil instead of synthetic preservatives for meat safety and the developing of organoleptic properties. However, there is no applied research about the use of essential oils during the meat drying process.

At the same time, there is a lack of any detailed research and information in scientific literature on drying behaviour in combination with essential oil treatments of dried meat. In this context, there is an opportunity to fill a gap and increase meat value in the drying process. This investigation therefore presents a value-added dried meat product obtained by using oregano essential oil to enhance food safety and the influence of different modified blanching treatments by the application of oregano essential oil in the product. In addition, the use of oregano essential oil is limited by organoleptic criteria, therefore various doses of oregano essential oil were tested to evaluate how the different concentrations affect the sensory quality of the product.

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LIST OF ABBREVIATIONS

OEO: Oregano essential oil

MICs: Minimal inhibitory concentrations

S. enteritidis: Salmonella enteritidis

E. coli: Escherichia coli

VBNC: Viable But NonCulturable

EOCs: Essential oil components

MHB: Mueller hinton broth

MIC: Minimal inhibitory concentration

$NaNO_{2:}$Sodium Nitrite

GS/MS: Gas chromatography/ mass spectrometry

GC-FID: Gas chromatography with flame ionization detector

NaCl: Sodium chloride

CFU: Colony forming unit

HR: Relative humidity

HAB: Hot air blanching

SB: Steam blanching

OT: Oil treatment

MC_{wb}: dry matter content on a wet basis

MCdb: dry matter content on a dry basis

MR: Moisture ratio

M: Moisture content at any time (kg water/ kg dry mater)

Mo: Initial moisture content (kg water/ kg dry mater)

Me: Equilibrium moisture content (kg water/ kg dry mater)

1 INTRODUCTION

1.1 Role of dried meat

Dried meat is no comparable to fresh meat in terms of appearance and sensory and processing properties, the advantage of dried meat is the significant extension of the shelf-life. Under certain circumstances, in particular with the lack of refrigeration, these detriments have to be accepted, particularly where the alternative may be the loss of the product by spoilage. Most nutritional properties of meat, in particular the protein content, remain unchanged through drying (Heinz and Hautzinger, 2007).

Since 1960, global meat production has more than trebled. This is attributed partly to the rise in population, as well as to the increase of income in many countries (Delgado et al., 2001). Meat consumption in developing countries has been continuously increasing from a modest average annual per capita consumption of 10 kg in the 1960s to 26 kg in 2000 and will reach 37 kg around the year 2030 according to FAO projections. This forecast suggests that in a few decades, developing countries consumption of meat will move towards that of developed countries where meat consumption remains stagnant at a high level (Heinz and Hautzinger, 2007). The kinds of meat commonly consumed in different countries vary according to eating habits and the ability to rear the animals positively, which is influenced by local climate, geography, and economy (Higgs, J. and Pratt, 2003).

Drying is used in making a variety of special processed meats. Nowadays, there is a renewed interest in drying as a means of effective food preservation (Eklund et al., 2004; Mahmoud et al., 2006; Rahman et al., 2000). Some products are dried at ambient temperatures, e.g., country ham, and others, such as beef jerky, are dried at elevated temperatures (Aberle et al., 2001, pp. 151–153, 193–194; Burnham et al., 2008). Salting and drying were first used as common procedures for preserving meats. Nowadays, these salted, dried meats known as "*cesina*" or "*jerky*" made from almost any lean meat, including beef, pork, poultry, or game represent a great variety of products and their characteristic flavour is one of the key attributes for the consumer (Hierro et al., 2004).

Even that a jerky can be made from different animal species more than 70% of jerky is produced from beef meat (Hoffman and Wiklund, 2006). Jerky is one of the oldest forms

of preserved meat, relying on salting and drying to reduce water activity and hence retard microbial growth (Faith et al., 1998). According to the U.S. Department of Agriculture (1996) a jerky is classified as a heat-treated and shelf-stable ready-to-eat meat product. A moisture-to-protein ratio (M/Pr) of jerky is ≤ 0.75:1 and can be made from sliced (i.e. whole-muscle jerky) or ground (i.e. re-structured or formed jerky) portions of usually lean beef, pork, fish, chicken, turkey, and/or venison.

In United States, jerky is a popular snack item, a preserved cured meat, where safe preservation, flavour, and texture are important. The simplest method to make jerky is to cut meat into thin strips and dry it. More typically, spices or marinades are used to flavour the meat and curing or smoking may be used in combination with drying to prepare jerky (Nummer et al., 2004). In fact, a sale increase of this type of snack food in USA from 631.6 million dollars in 1994 to almost 2.7 billion dollars in 2004 and moreover it is estimated that 39% American families regularly buy meat snack foods (Konieczny et al.,2007).

The popularity and importance of dried meat is not just in USA. According to Nielsen Global Survey of Snacking (The Nielsen Company, 2014), which polled more than 30,000 consumers in 60 countries throughout Asia-Pacific, Europe, Latin America, the Middle East, Africa and North America; in the previous year of the survey, the fastest-growing snack categories were savoury snacks in which sales increased 21 percent in Latin America. Meat snacks grew 25 percent in the Middle East/Africa and 15 percent in North America.

1.2 The Drying Process

Drying is defined as the removal of moisture from a product (Barbosa-Cánovas, 1996) in order to reach the desired moisture content and is an energy intensive operation. The major objective of drying apart from extended storage life can also be quality improvement, ease of handling, further processing and sanitation and is probably the oldest method of food preservation practiced by humankind (Mujumdar and Huang, 2007).The removal of moisture prevents the growth and reproduction of microorganisms like bacteria, yeasts and molds causing decay and minimizes many of the moisture-mediated deteriorative reactions. It brings about considerable reduction in weight and volume, minimizing packing, storage, and transportation costs and enables storability of the product under ambient temperatures (Hii et al., 2012).

Drying in earlier times was done mainly in the sun, now many types of sophisticated equipment and methods are used to dehydrate foods. During the past few decades, considerable efforts have been made to understand some of the chemical and biochemical changes that occur during dehydration and to develop methods for preventing undesirable quality losses. The widespread among drying methods is convective drying, i.e. drying by blowing heated air circulating either over the upper side, bottom side or both, or across the products. Hot air heats up the product and transfers released moisture to atmosphere (Hii et al., 2012).

In technical terms, there are two processes that occur simultaneously during the thermal process of drying a wet solid (1) heat transfer to change the temperature of the wet solid and to evaporate its surface moisture and (2) the mass transfer of moisture to the surface of the solid and its subsequent evaporation from the surface to the surrounding atmosphere (Mujumdar, 2006).

1.2.1 The psychrometric charts

The commonly encountered terminologies in psychrometry and drying are briefly tabulated in Table 1.1.

Table 1.1 - Definition of commonly encountered terms in psychrometry and drying (Baker, 1997)

Term / symbol	Meaning
Adiabatic saturation temperature,Tas	Equilibrium gas temperature reached by unsaturated gas and vaporizing liquid under adiabatic conditions. (Note: for the air-water system only it is equal to the wet bulb temperature [Twb].)
Bound moisture	Liquid physically and/or chemically bound to solid matrix so as to exert a vapor pressure lower than that of pure liquid at the same temperature.
Constant rate drying period	Under constant drying conditions, drying period when evaporation rate per unit drying area is constant (when surface moisture is removed).
Dew point	Temperature at which a given unsaturated air-vapor mixture becomes saturated.

Term / symbol	Meaning
Dry bulb temperature	Temperature measured by a thermometer immersed in vapor-gas mixture.
Equilibrium moisture content, Xeq	At a given temperature and pressure, the moisture content of moist solid in equilibrium with the gas-vapor mixture (zero for non-hygroscopic solids).
Critical moisture content, Xc	Moisture content at which the constant drying rate first begins to drop (under constant drying conditions).
Falling rate period	Drying period (under constant drying conditions) during which the rate falls continuously with time.
Free moisture, X; X= Xt-Xeq	Moisture content in excess of the equilibrium moisture content (hence free to be removed) at given air humidity and temperature.
Humid heat	Heat required to raise the temperature of unit mass of dry air and its associated vapor through 1 degree (kJ kg^{-1}).
Humidity, absolute	Mass of water vapor per unit mass of dry gas (kg kg^{-1}).
Humidity, relative	Ratio of partial pressure of water vapor in gas-vapor mixture to equilibrium vapor pressure at the same temperature.
Unbound moisture	Moisture in solid which exerts vapor pressure equal to that of pure liquid at the same temperature.
Water activity,	Ratio of vapor pressure exerted by water in solid to that of pure water at the same temperature.
Wet bulb temperature, Twb	Liquid temperature attained when large amounts of air-vapor mixture are contacted with the surface. In purely convective drying, the drying surface reaches Twb during the constant-rate period.

Psychrometric charts are useful tools that translate air–water vapor data into convenient graphic form. They rely on the fact that in drying processes the rate at which water evaporation occurs depends on vapor concentration in the surrounding

air. In Europe and countries following the SI system, the charts are based on humidity-enthalpy coordinates with relative humidity and temperature as parameters. They are known as Mollier charts and are very useful for the prediction of drying (Belessiotis and Delyannis, 2011).

Psychrometric chart (*cf.*Figure 1.1) for the air-water system shows the relationship between the temperature (abscissa) and absolute humidity (ordinate, in g water per kg dry air) of humid air at 1 atmosphere total pressure over the range 0° to 180°C. Lines representing percent humidity and adiabatic saturation are drawn according to the thermodynamics definitions of these terms (Baker, 1997).

Table 1.2 summarizes the essential thermodynamic relationships for humid air. Equation for the adiabatic saturation and wet bulb temperature lines on the psychrometric chart are as follows (Geankoplis, 1993):

$$\frac{Y - Y_{as}}{T - T_{as}} = -\frac{c_s}{\lambda_{as}} = -\frac{1.005 + 1.88Y}{\lambda_{as}} \qquad (1)$$

and

$$\frac{Y - Y_{wb}}{T - T_{wb}} = -\frac{h/M_{air}k_y + 1.88Y}{\lambda_{wb}} \qquad (2)$$

where ratio ($h/M_{air}k_y$), termed the psychrometric ratio, lies between 0.96 and 1.005 for air-water vapor mixtures, thus is nearly equal to the value of humid heat c_s. If the effect of humidity is neglected, the adiabatic saturation and wet bulb temperatures (T_{as} and T_{wb}, respectively) are almost equal for the air-water system.

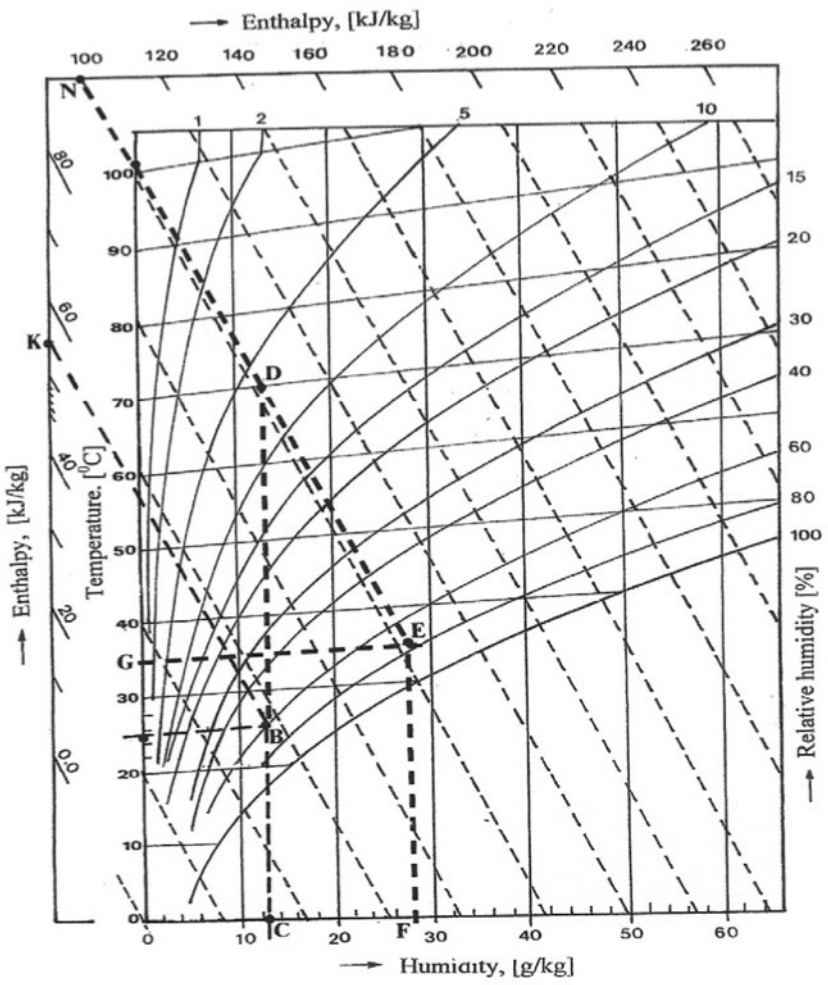


Figure 1.1 Psycometric chart for air-water vapour system

Adapted from Belessiotis and Delyannis (2011, p. 1688)

The adiabatic saturation temperature is a gas temperature and a thermodynamic entity. Plots of Y_{as} versus T_{wb} on psychrometric chart are straight lines and represent the path followed by the air in adiabatic drier. In contrast the wet-bulb temperature is a heat and mass transfer rate-based parameter and refers to the temperature of the liquid phase. Under constant drying conditions, the surface of the drying material attains the wet bulb temperature if heat transfer is by pure convection. The wet bulb temperature is independent of surface geometry as result of the analogy between heat and mass transfer (Baker, 1997).

Table 1.2 - Psychrometric equations for air-water vapor system (Geankoplis, 1993).

Parameter	Equation
Absolute humidity in kg H_2O per kg dry air	$Y = \frac{18.02}{29.97}\frac{p}{p_a - p}$
Saturation humidity	$Y = \frac{18.02}{29.97}\frac{p_w}{p_a - p_w}$
Percent humidity	$Y_p = 100\frac{Y}{Y_s}$
Relative humidity	$\psi = 100\frac{P}{P_w}$
Humid heat in kJ kg^{-1} dry air	$c_s = 1.005 + 1.88Y$
Total enthalpy in kJ kg^{-1} dry air	$H = (1.005 + 1.88Y)(T - T_r) + Y\lambda_r$ T_r is a reference temperature in K
Latent heat vaporization λ, kJ kg^{-1}	$\lambda = a_1(a_2 T)^{a_3}$ $a_1 = 267.155, a_2 = 374.2, a_3 = 0.38$ T in °C

1.2.2 Drying Kinetics

The mechanisms of moisture movement within the product can be summarized as (Barbosa-Cánovas, 1996): water movement under capillary forces; diffusion of liquid due to concentration gradients; surface diffusion; water vapor diffusion in air-filled pores; flow due to pressure gradient; and flow due to vaporization-condensation sequence. Capillary forces are responsible for water retention in porous solids of rigid construction, whereas osmotic pressure is responsible in aggregates of fine powders and on the surface of the solid.

The drying data are usually expressed as total weight of the wet material as a function of time during the drying process. Then, the data can be expressed in terms of rate of drying by recalculation of some values.

The moisture content is defined as the ratio of the amount of water in the food to the amount of dry solids, and is expressed as (Barbosa-Cánovas, 1996):

$$X_t = \frac{W_t - F_s}{F_s} \tag{3}$$

Where Wt is the total weight of the wet material at time t, F_s is the weight of the dry solids, and X_t is the moisture expressed as weight of water/weight of dry solids. An additional quantity that is important when designing a drying process is the free moisture content, X. The free moisture content can be evaluated by considering the equilibrium moisture content X_{eq}:

$$X = X_t - X_{eq} \tag{4}$$

The equilibrium moisture content X_{eq} refers to the moisture content when the vapor pressure exerted by the moisture of product equals vapor pressure of the nearby ambient air. This means that moisture desorption from the product is in dynamic equilibrium with the absorption of the environmental air moisture contain. Relative humidity at this point is known as the "equilibrium relative humidity", and is characterized by the curves of moisture content plots against equilibrium humidity known as moisture equilibrium isotherms (Belessiotis and Delyannis, 2011).

According to Barbosa- Cánovas (1996), the rate of drying, R, can be expressed proportional to the change in moisture content as a function of time $[t]$:

$$R \propto \frac{\mathrm{d}X}{\mathrm{d}t} \tag{5}$$

By convection, the drying rate R, is defined as (Mujumdar, 2006):

$$R = -\left(\frac{F_s}{\mathrm{A}}\right)\left(\frac{\mathrm{d}X}{\mathrm{d}t}\right) \tag{6}$$

where R is the drying rate and A is the surface area where the drying takes place.

In Barbosa- Cánovas (1996) the drying process of a product can be described as a series of steps in which the drying rate plays a key role. Figure 1.2 shows typical drying rate curve for a constant drying condition. Points A´ and A represent either a hot or a cold product, respectively. Point B represents an equilibrium temperature condition of the product surface. The period between points A (or A´) and B is usually short, and is not important in the analysis of drying times.

Section B to C of the curve, recognized as the constant rate period, represents the removal of free water from the product. The water acts as if the solid is absent. The surface of the product is very wet at the beginning and the water activity is approximately one. On porous solids, the removed water is supplied from the interior of the solid. The constant rate period continues only as long as the water is supplied to the surface as fast as it evaporates. As long as there is a film of free water present on the surface, the drying rate will remain in the constant period (Mujumdar, 2006). In general, the drying rate is determined by external conditions of temperature, humidity and air velocity (Aversa et al., 2007).

The falling rate period is reached when the drying rate starts to decrease, and the surface water activity falls to less than one. The rate of drying is governed by the internal flow of liquid or vapor (Barbosa-Cánovas, 1996). This point is represented by C in Figure 1.2.

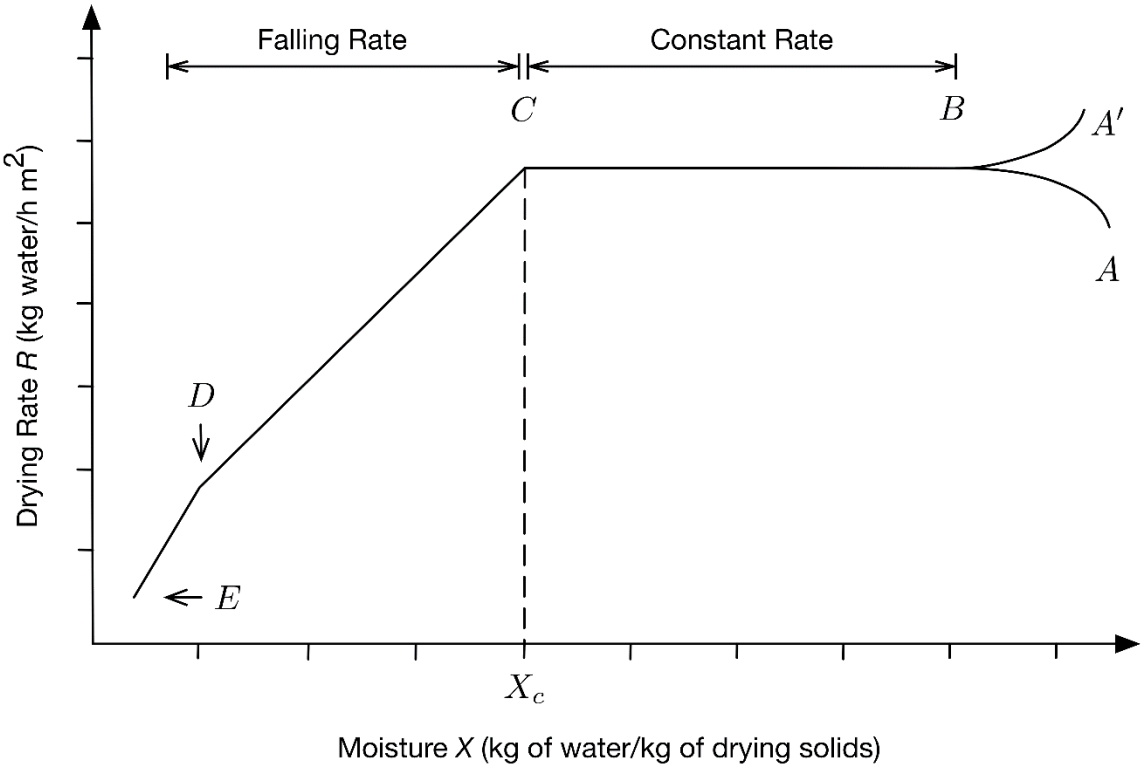


Figure 1.2 - Drying rate curve

Adapted from Barbosa-Cánovas (1996, p. 105)

At this point there is not enough water on the surface to maintain a water activity value of one. The falling rate period is divided in two stages. A first falling drying rate happens when wetted spots in the surface recurrently reduce until the surface is dried (point D), and a second falling rate period begins at point D when the surface is completely dry. The plane of evaporation recedes from the surface. Heat required for moisture removal is transferred through the solid into the air stream. Sometimes, there are no sharp differences between the first and second falling rates periods. The amount of water removed in this period may be relatively small, while the time required may be long because the drying rate is low (Barbosa-Cánovas, 1996).

1.3 Food-borne outbreaks

The risk of food-borne illnesses from poorly dried meat such as salmonellosis and infestation by *E. coli*, presents a great hazard, which is needed to be controlled. Previous studies have reported food-borne contamination attributed to meat jerky (Eidson et al., 2000; Govaris et al., 2010; Greig and Ravel, 2009; Keene et al., 1997; Rabsch et al., 2001). In many countries, the majority of the recorded food-borne outbreaks are attributed to salmonellosis (Govaris et al., 2010; Rabsch et al., 2001) and a wide range of food items have been implicated as vehicles of *S. enteritidis* infection in humans including meat (Greig and Ravel, 2009)

In 2003, at least 22 cases of salmonellosis were attributed to consumption of commercially produced beef jerky in New Mexico and investigators concluded that the very slow drying process under low humidity conditions allowed *Salmonella* organisms to dehydrate and become very resistant to the dry heat (Allen et al., 2007). In United States, a study by Levine et al. (2001) also suggested that foodborne pathogens can survive the moderate drying conditions (approximately 60°C) used by commercial jerky manufacturers. Their study informed a cumulative occurrence of *Salmonella* of 0.31% in jerky produced in federally inspected plants from 1990 to 1999, even though good manufacturing practices were tracked and Hazard Analysis Critical Control Point plans were in place.

E. coli O157:H7 outbreaks of foodborne illness due to ground dried-meat products during the mid-90s distinguished these products as likely vehicles for this pathogen (Faith et al., 1998; Nummer et al., 2004). In 1995, In United States, a study by Keene et al. (1997) investigated an outbreak of *E. coli* O157:H7 involving homemade jerky from deer meat. As many as 11 persons were infected. The home processor dried the

venison in a home-style dehydrator set between 51.7 and 57.2°C for 12 to 14 h. *E. coli* O157:H7 was recovered from the deer jerky, deer meat, and the deer carcass. After, jerky strips were inoculated with a single strain of *E. coli* O157:H7 and dried at temperatures up to 62.8°C for 10 h. Viable bacteria were recovered under all conditions tested. This report indicated that deer can be a reservoir for *E. coli* O157:H7. They determined that some traditional home-drying processes for jerky were insufficient for killing *E. coli* O157:H7 and suggested precooking deer meat to 74°C before drying.

1.4 Viable but nonculturable bacteria

Interestingly, in all the cases presented before, the risk is caused by pathogenic bacteria called as viable but nonculturable (VBNC). Both, *S. enteritidis* and *E. coli*, are species known to enter a VBNC state under stress conditions (Oliver, 2010, 2005) such as temperature changes and therefore once resuscitated, will become culturable (Khamisse et al., 2012) and may cause foodborne contamination.

Bacteria in the viable but nonculturable (VBNC) state fail to grow on the routine bacteriological media on which they would normally grow and develop into colonies, but are still alive (Oliver, 2010). In this sense, is a common response to stress factors by bacterial cells is their inability to develop into colonies on routine culture media, even though the cells may continue viable for long periods of time.

In conclusion, the VBNC state is a survival strategy and as it was suggested by Epstein (2009) that "dormancy" and "waking up" from this state, could be a method analogous to 'sending out scouts' to 'test the environment' for its suitability for growth of the entire population. In this situation, if the resuscitating cells notice that the previously stressful/adverse environment is now growth-permissive, they would indicate the remaining cells to revive.

When considering the role that the VBNC state plays, it is clear that a large number of non-spore-forming bacteria, most particularly a large number of human pathogens, are capable of entering this state, maintaining cellular structure and biology and continuing significant gene expression while otherwise nonculturable by standard laboratory methods (Oliver, 2010).

1.4.1 MIC

Minimum inhibitory concentrations (MICs) are defined as the lowest concentration of an antimicrobial that will inhibit the visible growth of a microorganism after overnight incubation (Andrews and Andrews, 2001). MICs are considered the "precious standard" for determining the susceptibility of organisms to antimicrobials. MICs are absolutely critical in diagnostic laboratories to confirm resistance of microorganisms to an antimicrobial agent, to reach a definitive answer when an unclear result is obtained by other methods of testing, and also to monitor the activity of new antimicrobial agents (Barros et al., 2006).

A Minimum inhibitory concentration is generally observed as the most basic laboratory measurement of the activity of an antimicrobial agent against an organism. Therefore, is very important to determine the MICs necessary to inhibit the growth of pathogenic bacteria such as *S. enteritidis* and *E. coli,* in this investigation.

1.5 Chemical additives Vs Natural additives

Besides drying, microbial load can be controlled using either synthetic or natural preservatives. Nowadays, meat industry uses chemical additives such as chlorides, nitrites, sulphites, lactic acid, ascorbic acid and sorbic acid (Dave and Ghaly, 2011) in several meat processes like curing where sodium nitrite is used to preserve meat (Nyachuba et al., 2007), and in refrigerated storage the synthetic antimicrobials added improve the shelf life of the product (Govaris et al., 2010). Concerns about the safety of chemical additives have arisen in recent years; consumers progressively demand use of natural products as alternative preservatives in foods. Particular interest has been focused on the potential application of plant essential oils instead of synthetic preservatives as safer additives for meat (Holley and Patel, 2005).

Preceding studies have stated that natural plant extracts have the potential to improve the overall quality and extend the shelf life of food products (Bussmann et al., 2010; Raman et al., 2009). Moreover, they can also be used in numerous food applications such as meat (turkey, beef, and chicken), seafood (Miladi et al., 2010), vegetable produce (spinach), probiotics, and packaging films (Cagri et al., 2003, 2001) along with other several technologies (bacteriocins, organic acids, temperature, and packaging) to enhance the microbial quality and safety of the food

products (Govaris et al., 2010; Nazzaro et al., 2009; Patra and Thatoi, 2011; Perumalla and Hettiarachchy, 2011; Serra et al., 2008).

On the other hand, the addition of antimicrobials to food products without adversely affecting the sensory attributes is still a challenge for researchers. The concentrations of plant essential oils that are necessary to ensure safety, up to 5 log CFU/g reductions in the most resistant pathogenic microorganism (Borowski et al., 2009) are several times higher than those accepted by consumers from sensory point of view (Burt and Reinders, 2003; Govaris et al., 2010). Consequently, new studies linking the use of antimicrobials with other methodologies of food preservation are essential to decrease the effect of these compounds on sensory properties.

1.5.1 Essential oils components

Essential oils, also named volatile or ethereal oils, are aromatic oily liquids obtained from plant materials (flowers, buds, seeds, leaves, twig bark, herbs, wood, fruits, and roots), which can be achieved by fermentation, extraction, or distillation. Distillation is the most common used method for the commercial production of essential oils (Burt, 2004).

Particularly, the antimicrobial compounds in plant materials are usually found in the essential oil portion of leaves (rosemary, sage, basil, oregano, thyme, and marjoram), flowers or buds (clove), bulbs (garlic and onion), seeds (caraway, fennel, nutmeg, and parsley), rhizomes (asafetida), fruits (pepper and cardamom), or other parts of plants (Gutierrez et al., 2008; Nychas et al., 2003).

Essential oils are constituted of a complex mix of compounds including terpenes, alcohols, cetones, phenols, acids, aldehydes, and esters (Ayala-Zavala et al., 2008; Burt, 2004). Chemical analysis of a range of essential oils revealed that the principal constituents of many include carvacrol, thymol, citral, eugenol (*cf.* Figure 1.3 for their chemical structure). Usually, the antimicrobial efficacy of essential oils is dependent on the chemical structure of their components as well as the concentration (Tiwari et al., 2009)

The use of phenolic compounds as antimicrobial agents provides additional aids such as preservation and health benefits. Knowing the antimicrobial effect of the phenolic compounds from several kinds of comestible plants on pathogenic microorganisms from foods, makes possible to search new strategies by the

combination of antimicrobial effects of phenolic compounds with their natural biological properties.

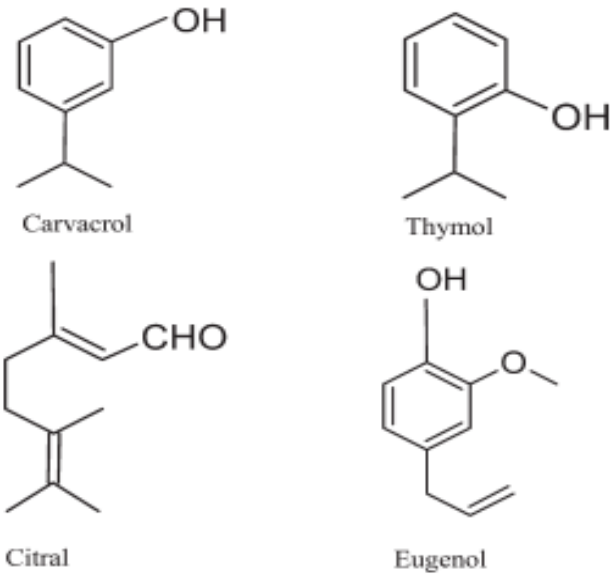


Figure 1.3 - Plant origin antimicrobial agents

Adapted from Tiwari et al. (2009)

The modes of action for phenolic compounds as antimicrobial agents can be concentration dependent (Juven et al., 1994). At low concentration, phenols affect enzyme activity, mainly those associated with energy production, while at high concentrations, they cause protein denaturation. The antimicrobial effect of phenolic compounds may be due to their ability to alter microbial cell permeability, consequently permitting the loss of macromolecules from the interior, such as ribose and Na glutamate (Rai and Chikindas, 2011). They could also interfere with membrane function (electron transport, nutrient uptake, protein, nuclein acid synthesis, and enzyme activity) (Rai and Chikindas, 2011) and interact with membrane proteins, causing deformation in structure and functionality (Kabara and Eklund, 1991; Rico-Munoz et al., 1987).

Essential oils are mainly used as food flavorings, in perfumes (fragrances and aftershaves), and as functional components in pharmaceuticals (Nychas et al., 2003). The individual essentials oils components (EOCs) are either extracted from plant material or are synthetically manufacture (Burt, 2004). The use of essential oils as food preservatives is often limited due to flavour considerations (Lambert et al., 2001).

1.5.1.1 Oregano Essential oil

One of the most important groups of bioactive natural compounds are essential oil components (EOCs). Since at least the middle ages these compounds have been widely used for their antimicrobial, antiparasitic and insecticidal properties in various applications (Cavanagh, 2007; Nedorostova et al., 2009; Tajkarimi et al., 2010). EOCs may be lethal to microbial cells or they might inhibit the production of secondary metabolites (e.g., mycotoxins)(Lazar, 2003). Plant essential oils are generally more inhibitory against Gram-positive than Gram-negative bacteria (Chorianopoulos et al., 2004; Gutierrez et al., 2008; Marino et al., 2001). Even though this is true for various essential oils, there are some agents that are effective against both groups, such as oregano, clove, cinnamon, and citral (Kim and Fung, 2004; Sivropoulou et al., 1996; Skandamis and Nychas, 2001).

EOCs, such as carvacrol and thymol, the two main phenols that constitute about 78-85% of oregano essential oil (OEO) (Kokkini et al., 1997). Both prevent the microbial and chemical deterioration when added to food, moreover their antimicrobial activity can be enhanced when combined with other natural preservatives (Burt, 2004; Mahmoud et al., 2006; Yamazaki et al., 2004). Carvacrol and thymol are reported to decrease the intracellular ATP (adenosine triphosphate) content of *E. coli* O157:H7 cells while simultaneously increasing extracellular ATP, indicating the disruptive action of these compounds on the plasma membrane (Helander et al., 1998).

Nowadays, oregano (*Origanum vulgare*), a herb of the *Lamiaceae* family, has been known as a flavouring agent for meat (Govaris et al., 2010). Other studies found the application of oregano essential oil effective in meats by inhibiting the spoilage microflora during fresh meat storage (Skandamis and Nychas, 2002, 2001; Tsigarida et al., 2000). However, there is no applied research about the use of plant essential oils during the meat drying process.

In this context, there is an opportunity to fill a gap and increase meat value in the drying process. Therefore, this study presents a value-added dried meat product obtained by using OEO to enhance food safety. However, the use of OEO is limited by organoleptic criteria. For this reason, it was essential to determine the MICs necessary to inhibit the growth of pathogenic bacteria, such as, *S. enteritidis* and *E. coli.*

1.5.2 Organic acids

Organic acids are organic compounds with acidic properties that are used in food preservation due to their antimicrobial effect on bacteria. The most common organic acids are lactic, acetic and propionic acids. Acetic acid is the strongest inhibitor and has a wide range of inhibitory activity, inhibiting yeasts, molds and bacteria (Blom and Mørtvedt, 1991), while propionic acid has been observed to perform a strong antimicrobial effect, in particular towards yeasts and molds (Suomalainen and Mäyrä-Makinen, 1999). Lactic acid is FDA (Food and drug administration) approved as an antimicrobial for application to animal carcasses. Its use in the meat industry is widespread and many researchers have documented its efficacy for the reduction of enteric pathogens on the surfaces of carcasses and derived cuts ((Baird et al., 2006; Bosilevac et al., 2006; Delmore Jr. et al., 2000; Mani-López et al., 2012).

The key basic principle on the mode of action of organic acids on bacteria is that non-dissociated (non-ionized) organic acids can penetrate the bacteria cell wall and disrupt the normal physiology of certain types of bacteria that we call *pH-sensitive*, meaning that they cannot tolerate a wide internal and external pH gradient. Among those bacteria are *E. coli* and *Salmonella* spp (Cetin-Karaca, 2011).

Upon passive diffusion of organic acids into the bacteria, where the pH is near or above neutrality, the acids will dissociate and lower the bacteria internal pH, conducting to situations that will decline or stop the growth of bacteria. On the other hand, the anionic part of the organic acids that cannot escape the bacteria in its dissociated form will accumulate within the bacteria and interrupt many metabolic functions, leading to osmotic pressure increase, incompatible with the survival of the bacteria (Cetin-Karaca, 2011).

An emerging potential issue is that organic acids have been detected to enhance survivability of acid sensitive pathogens exposed to low pH by induction of an acid tolerance response and that acid tolerance may be linked to increased virulence. Although this situation has implications regarding the use of organic acids, it may only apply to circumstances in which reduced acid levels have induced resistance and virulence mechanisms in exposed organisms. Evaluating effectiveness of organic acids for specific applications requires more understanding general and specific stress response capabilities of foodborne pathogens (Ricke, 2003).

1.5.3 Phytoalexins

Phytoalexins were first described as antifungal substances which are specifically formed when a plant is attacked by a fungus (Rai and Chikindas, 2011). Today, they defined as low molecular weight antimicrobial compounds that accumulate in plants as a result of infection or stress (Kuc, 1995). The rapidity and extend of their accumulation is determined by their release of immediate precursors from conjugates and/or de novo synthesis, as well as detoxification as a result of plant or microbial enzymes. Phytoalexins are only one component of the complex mechanisms for disease resistance in plants caused by fungi and bacteria (Kuc, 1995).

Phytoalexins are mainly produced by members of Leguminaceae and Rosaceae. Chemically, they have isoflavonoid structures classified as isoflavones, isoflavonones and isoflavans (Rai and Chikindas, 2011). Examples of these are isoflavonoids from Leguminosa as pisatin from garden pea (*Pisum savitum*) and phaseollin from beans (*Phaseolus vulgaris*). Others are rishitin from potatoes and tomatoes (Solanaceae), falcarindiol and 6-methoxy-mellein from carrots (J.R.L, 1994; Smid and Gorris, 1999).

1.5.4 Enzyme-released antimicrobial agents

Two major types of antimicrobials compounds activated by enzyme hydrolysis exist in edible plants. In the Allium family (garlic, onions, leek), sulfoxides are converted to pungent smelling sulphides such as dyallil disulfide upon tissue rupture (Baines and Seal, 2012). Most potent is garlic. It contains allinin (propenyl-cysteine sulfoxide), which is hydrolysed by the enzyme alliinase to allicin (2-propenyl-2-propenethiol sulfinate) (Ohlsson and Bengtsson, 2002). Various studies have shown these substances to be inhibitory to a wide range of microorganisms, but only at relatively high concentrations (Beuchat, 1994; Conner, 1993) and the main hindrance of use is sensory effects (Ohlsson and Bengtsson, 2002).

In plants of the Cruciferae family (cabbage, mustard, horse radish, Brussels sprout), glucosinolates are the substrates for hydrolytic enzymes. For example, sinigrin stored in mustard seeds is cleaved by myrosinase to yield allyl isothiocyanate (AITC), which is inhibitory against a wide range of bacteria such as *E. coli* O157:H7 and *S. enteritidis* and fungi (Delaquis and Sholberg, 1997). Applied through the gas

phase, the volatile AITC has proved effective in very low doses against food spoilage fungi (Ohlsson and Bengtsson, 2002).

1.6 Drying pre-treatments

Several pre-drying treatments has been studied in the drying of fruits and vegetables (An et al., 2013; Cheng et al., 2015; Dorantes et al., 2011; Kendall et al., 2012; Pangavhane et al., 1999; Ruiz-Ojeda and Peñas, 2013; Sotome et al., 2009; Tunde-Akintunde et al., 2005). The treatments tested in this study included steam blanching, oil treatment (oil dipping) and hot air blanching. Those are called modified blanching due to the application of OEO with different doses. However, the basic principles of some treatments, such as blanching and dipping were followed, and in this sense, it was applicable to the food system of this study. For the particular case of hot air blanching, previous studies were not found in the literature, thus the treatment is described in the chapter of materials and methods.

1.6.1 Blanching

Blanching is a pre-treatment operation whose main objective in ripe fruits and vegetables is to inactivate enzymes, and is usually applied before cutting, peeling, and freezing, in order to avoid browning and changes in texture. Besides inactivating enzymes such as polyphenoloxidase, peroxidase and pectinase, blanching may induce sensory and chemical changes that should be evaluated (Dorantes et al., 2011). Other objectives of blanching are to reduce the microbial load of products so as to improve its conservation, shorter cooking time, and to remove intracellular air to prevent oxidation (Ramesh et al., 2002; Shivhare et al., 2009). As a further consequence, blanching also lowers somewhat the mass of vegetables, so the process profitability can be affected by overtreatment (Ruiz-Ojeda and Peñas, 2013).

Blanching can be done in a number of ways which usually includes water, steam and oil blanching (Tunde-Akintunde et al., 2005). DiPersio (2005) evaluated the influence of traditional home-type drying treatments recommended on inactivation of *Salmonella* on carrot slices. Pre-drying treatments tested included steam or water blanching, immersion in NaCl solution, and post-drying oven heating (Kendall et al., 2012; Sagar and Suresh Kumar, 2010). It was concluded that modified treatments were necessary to enhance inactivation of *Salmonella* on dehydrated carrot slices.

Other reports about heat transfer and kinetics of thermal inactivation of enzymes in potato blanching has been studied and reported in detail by several researchers (Arroqui et al., 2002; Gonzalez-Martinez et al., 2004; Mukherjee and Chattopadhyay, 2007). In this studies, boiling water or saturated steam is usually used as a heating medium for potato blanching. However, softening of the tissue and unexpected quality changes are caused by water absorption into the potato tissue when these heating media are used.

As well, in studies carry out with pepper, pre-treatment of pepper before drying improves the quality of the dried pepper and increases its drying rate. Steam and water blanching as a form of pre-treatment has been reported to increase drying rate and improve the quality of dried products but there is not much information on other types of oil/water blanching methods (Tunde-Akintunde et al., 2005).

Blanching affects the distribution of soluble components within the tissues during drying. This also results in the leak of soluble components to the nearby environment (water) and loss of these solutes affects the rate of drying. Researchers have reported that blanching increases the drying rate of some fruits including peach slices (*Prunus persica* L.), red pepper (*Capiscum annum*) and carrot and pumpkins slabs (*Daucus carota* L.) (Akanbi et al., 2003; Arevalo-Pinedo and Xidieh Murr, 2007; Kingsly et al., 2007).

1.6.2 Oil dipping

Although many studies have been carried out on the effects of steam and water blanching on the drying rate of fruits and vegetables, not much has been done on other sorts of blanching methods, particularly oil blanching.

According to Tunde-Akintunde (2005), whom studied the effect of various blanching methods on the drying characteristics of bell pepper (*Capiscum annum*), the implementation of dipping in a homogenized mixture of palm oil or groundnut oil and water before the samples were placed in a hot air dryer reveals that the addition of oil to water used for blanching helps to replace the initial outer wax layer, which is helpful because of the protection it offers the fruit or vegetable from environmental and external factors. Furthermore, the presence of this layer and the well-known hydrophobicity of the used oils acted to reduce the moisture movement slightly, even so resulted in higher drying rate when compared to untreated pepper.

Similar effect of reduction of moisture movement due to surface coating was also observed during the drying of seedless grapes (Pangavhane et al., 1999). The treatment with cold dipping pretreatment (in which the solution is kept at ambient temperature), increase the drying rate, however less than that the hot dipping, but the raisins produced get an attractive golden brown colour without any cracks on the berries. In this context, the colour, taste, and texture of the raisins produced are important attributes for consumer acceptance and are judged by sensory evaluation.

1.7 Quality of dried product

In drying process, the continuous evaporation and weight losses cause changes of the shape of the meat through shrinkage. The meat pieces become smaller, thinner and to some degree wrinkled and darker in color. The texture also changes from soft to firm to hard. Dried meat is no comparable to fresh meat in terms of appearance and sensory and processing properties, the advantage of dried meat is the significant extension of the shelf-life. Under certain circumstances, in particular with the lack of refrigeration, these detriments have to be accepted, particularly where the alternative may be the loss of the product by spoilage. Most nutritional properties of meat, in particular the protein content, remain unchanged through drying (Heinz and Hautzinger, 2007).

According to Heinz and Hautzinger (2007) after the drying of fresh, untreated meat cut into strips, the product should meet the following quality criteria:

- The appearance of the dried meat should be as uniform as possible. The absence of large wrinkles and notches indicates the desired steady and uniform dehydration of meat.
- The color of the surface, as well as of the cross-cut should be uniform and dark red.
- The texture of properly dried meat must be hard, similar to frozen meat.
- Taste and flavor are very important criteria for the acceptance of dried meat by the consumer. Dried meat should possess a slight salty taste which is distinctive of naturally dried meat with no added spices. Off-odors must not occur. However, a slightly rancid flavor, which happens because of chemical changes during drying and storage, is commonly found in dried meat and is acceptable.

2 OBJECTIVES

The main objective of the thesis is to investigate the effect of the application of oregano essential oil on the microbial load, drying behaviour and sensory properties of dried meat.

The specific objectives are as follows:

- Enhance the meat drying process by using oregano essential oil to inhibit the growth of bacteria.
- Evaluation of drying behavior of beef dried meat with three modified blanching treatments: (1) oil treatment (2) steam blanching and (3) hot air blanching.
- Analysis of final sensory properties of the value-added dried meat product obtained.

3 MATERIALS AND METHODS

3.1 Meat samples

Fresh beef (steer, Bos Taurus, Fleckvieh Breed, 23 months old) from biceps femoris was purchased from a local butchery in Prague, Czech Republic. For the microbial load analysis, the outer surface of each beef muscle was sterilized by immersion in 70% (v/v) ethanol, then the outer surface was aseptically removed with a knife in a laminar safety cabinet (Faster, Italy). In the case, of the drying kinetics analysis the outer surface of each beef muscle was washed with distillated water.

After that, the beef muscles were stored at -6°C for 1 day and then each frozen muscle was thawed at 4°C for few hours and was sliced into 0,5 cm thickness by a meat cutter SILVER (Kalorik, Fl, USA), then cut into 5 x 2.5 cm small rectangular samples (*cf.*Figure 3.1). Meat slices were packaged in plastic bags and stored at 6°C for later use.

Figure 3.1 - Preparation of meat samples

3.2 Preparation of inoculum and inoculation procedure

Two bacterial strains were used to inoculate the meat samples in this study: *Salmonella enteritidis* ATCC 13076 and *Escherichia coli* ATCC 25922. Strains were purchased from Fluka Analytical (Sigma Aldrich, USA) and Oxoid (Brno, CZ), respectively. Bacterial inoculum in concentration 1.5×10^7 CFU g^{-1} were prepared into Mueller Hinton Broth (MHB), from 24 hours old cultures cultivated in MHB at

37°C. Subsequently, raw meat slices were inoculated with 800 µl of selected strain and distributed on the surface (400 µl per each side), and let dry for 20 minutes. All procedures were done in a laminar safety cabinet (Faster, Italy).

3.3 Drying procedure

The first part of the drying was carried out in a standard laboratory drier (Memmert drier UFE 400, Germany, *cf.* Figure 3.2a). The second part of the experiments for the application of different treatments with oregano essential oil, the drier used was a climate box (Pol-Eko climate box KKP 115 Top+, Poland) which were connected to two external balances and is referred as a laboratory dryer (*cf.* Figure 3.2b).

3.3.1 Drying procedure and OEO application for calculation of microbial load

The drying was carried out in a standard laboratory drier (Memmert drier UFE 400, Germany) equipped with two trays (*cf.* Figure 3.2c). Before each drying experiment the drier was preheated to appropriate temperature. Samples were dried for 6 hours at 55°C, with drying air relative humidity values ranging from 30 to 45%. Oregano essential oil (OEO) (64.5% carvacrol, 5.2% p-cymene and 2.9% thymol), was purchased from commercial vendor Biomedica (Prague, CZ), the chemical composition was analyzed by GC/MS and GC-FID as described elsewhere by Kloucek et al. (2012) For the application of the vapours a filter paper soaked with essential oil and placed in front of the fan inside the dryer was used. The concentration of essential oil was expressed as volume of OEO per volume of the drier (ml l^{-1} air). To reach the minimal inhibitory concentration of OEO for each pathogen, doses of 1.5 ml (0.028 ml l^{-1} air), 2 ml (0.038 ml l^{-1} air) and 3 ml (0.057 ml l^{-1} air) were used for *S. enteritidis* and doses of 0.75 ml (0.014 ml l^{-1} air), 1.5 ml (0.028 ml l^{-1} air) and 3 ml (0.057 ml l^{-1} air) were tested for *E. coli*. The immersion in sodium nitrite ($NaNO_2$) was used to compare the OEO with synthetic preservative. Sodium nitrite was purchased from Lach-Ner (Neratovice, CZ) and after inoculation was applied to the raw samples by immersion in concentration 1 g l^{-1} to avoid toxicity (Hospital et al., 2014; RJ et al., 1992) in both cases, against *S. enteritidis* and *E. coli.* These samples were drying using the same procedure.

Memmert drier UFE 400

(a) closed

Pol-Eko climate box KKP 115 Top+

(b) closed

(c) opened

(d) opened

Figure 3.2 - Laboratory driers

3.3.2 Preparation of modified blanching treatments

Meat samples were treated by three different modified blanching treatments by the application of oregano essential oil (OEO) with three different doses 1.5 ml, 3 ml and 6 ml. Oregano essential oil (OEO) (64.5% carvacrol, 5.2% p-cymene and 2.9% thymol), was obtained from commercial vendor Biomedica (Prague, CZ), the chemical composition was analyzed by GC/MS and GC-FID as described elsewhere by Kloucek et al.(2012). The first treatment, hot air blanching (HAB) see Figure 3.3a. consisted in the use of a filter paper soaked with each different concentration of OEO and placed in front of the fan inside the drier (Memmert drier UFE 400, Germany) equipped with two trays, then the meat sample was introduced into the drier and was dried for 10 min at 35°C to generate vapours of OEO. For the steam blanching (SB) treatment see Figure 3.3b, the meat sample was placed in a metal strainer inside a pan which contains 1 l of distilled hot water with the addition of each concentration of OEO and blanched at 90°C for 2 min and then cooled immediately in ice cubes of distilled water. In the third method, oil treatment (OT) see Figure 3.3c, the sample was subjected to dipping for 10 min in 20 ml of sunflower oil in combination with each OEO concentration at ambient temperature.

3.3.3 Drying procedure for modified blanching

The drying was carried out in a climate box (Pol-Eko climate box KKP 115 Top+, Poland) equipped with two small trays which were connected to two external balances (Radwag electronical balance PS 1200.R2, Poland) and those were connected to Pomiar Win computer program for the extraction and collection of the weight loss data (*cf.* Figure 3.2d). Before each drying experiment the climate box was programed to start about 1 h before the drying experiments to achieve steady-state conditions of drying temperature and air relative humidity (RH). Per drying run, two meat samples (control and treated sample) were placed in the two trays and dried for 6 h at 55°C with drying RH ranging from 20 to 30%. Temperature, RH and the weight loss of the samples were monitored and collected by the program system every minute throughout drying. The control sample was untreated and it was dried in the climate box per drying run. At the end of each drying test the meat samples were collected and dry matter content was estimated by the oven method at 105°C for 24 h (Memmert drier UFE 400, Germany). The experiments were conducted per treatment and concentration by triplicate.

Equation (7) was used to estimate dry matter content on a wet basis MC_{wb} (Kg per Kg of mixture), where W is mass of water (kg) and W_d is mass of dry solid (kg) (Belessiotis and Delyannis, 2011).

$$MC_{wb} = \frac{W(kg)}{W(kg) + W_d\ (kg)} \qquad (7)$$

The most convenient way to express moisture for mathematical calculations is on dry basis. Equation (8) was used to estimate dry matter content on a dry basis MC_{db} (Kg of water per Kg of dry material), where W is mass of water (kg) and Wd is mass of dry solid (kg) (Belessiotis and Delyannis, 2011). Each punctual value was used for further calculations:

$$MC_{db} = \frac{W(kg)}{W_d\ (kg)} \qquad (8)$$

(a) HAB

(b) SB

(c) OT

Figure 3.3 - Modified blanching treatments

3.3.3.1 Calculation of kinetics parameters

To predict the drying curves of the meat samples, the moisture content data in dry basis was transformed into the dimensionless moisture ratio (MR) as described in Eq.(9) (Radhika et al., 2011). Where M is the moisture content at any time (kg water/ kg dry mater), M_o is the initial moisture content (kg water/ kg dry mater), and M_e (kg water/ kg dry mater) is the equilibrium moisture content. Since the values of M_e may be relatively small compared to M and M_o, the equation was simplified.

$$MR = \frac{M - M_e}{M_o - M_e} = \frac{M}{M_o} \qquad (9)$$

The moisture ratio was used to plot the drying curves instead of the moisture content due to the initial value for all the drying experiments which is 1, is a uniform value (Afolabi et al., 2015). The drying curves of the present study were performed using the software MATLAB R2015a and we expressed the moisture ratio as a function of drying time to compare the different treatments.

3.4 Microbial analysis

Inoculated raw samples were tested for the presence of *S. enteritidis* and *E. coli* and compared with non-inoculated control samples prior to the drying to see inoculation efficiency. For $NaNO_2$ and each OEO concentration two inoculated dried samples were compared with two dried non-inoculated control samples. After the drying the samples were checked for viable *S. enteritidis* and *E. coli*, respectively. The pre-enrichment of each meat sample suspended in a flask with peptone water (8.5 g NaCl, 1 g peptone, 1 g Tween 80 l^{-1}) in ratio 1:10 (w/v) and incubated at 37°C for 4 and 6 hours, was used to determine the presence/absence of bacteria. After each period, resulting solution was serially diluted (1:9) in sterile peptone water. Sample dilutions (0.1 ml) were cultivated by direct planting (Roberts and Greenwood, 2003) on selective agar; S.S. for *S. enteritidis* (Fluka Analytical, CZ) and MacCONKEY agar (OXOID Laboratories, CZ) for *E. coli*. The number of colonies (CFU g^{-1} of dried meat) was evaluated after 24 hours cultivation in 37 °C. Prior to meat inoculation with pathogen, meat samples were also examined for any contamination, they were suspended in a flask with peptone water with the same characteristics as described

before with the difference that they were not pre-incubated for 4 and 6 hours and the sample dilutions (0.1 ml) were also cultivated by direct planting.

3.5 Sensory analysis

The aim of the sensory evaluation was to determine whether the addition of OEO affected the sensory properties of the dried meat. The sensory analysis was divided in two parts. The first part was carried out for the treatment and the concentrations tested for the calculation of microbial load. The second part of the sensory analysis was applied to different modified blanching treatments by using oregano essential oil with different concentrations.

3.5.1 First sensory analysis

Sensory analysis of beef samples treated with OEO was performed by 51 trained assessors. The sensory panellists comprised of students and employees of the Department of Quality of Agricultural Products from the Faculty of Agrobiology, Food and Natural Resources of Czech University of Life Sciences Prague who were selected and trained according to ISO 8586: 2012. All samples were prepared 1 day prior to the sensory evaluation and were held in zippered plastic bags to prevent the loss of oregano flavour. For salt treatment, samples were immersed in salt solution (38 g NaCl l^{-1}) for ten minutes and dried as described before.

Dried meat samples were coded using three-digit, randomly generated numbers. Six samples were offered to each panellist: 1. Control, 2. Control + salt, 3. Treated 0.75 ml + salt, 4. Treated 0.75 ml no salt, 5. Treated 1.5 ml no salt and 6. Treated 1.5 ml + salt. Ethanol 40% was provided to assessors as neutralizer between samples. The panel was instructed how to evaluate the samples and fill in the form prior the beginning of the evaluation. Linear unstructured graphical 100 mm scales oriented by description at both ends were used for sensory profile evaluation of fifteen descriptors (Table 3.1). Sensory analysis was accomplished in 4 sessions (in separate days) within 2 weeks.

3.5.2 Second sensory analysis

Beef samples were evaluated by 16 trained assessors. The sensory panellists consisted of students and employees of the Czech University of Life Sciences Prague, who were experienced panellists. All samples were prepared 1 day prior to the sensory evaluation and were vacuum-packaged (MAGIC VAC Champion, Elaem Nuova, Italy) in bags at ambient temperature to prevent the loss of oregano flavour. For salt treatment, all samples were immersed in salt solution (12 g NaCl l^{-1}) for ten minutes and dried as described before.

Dried meat samples were coded using three-digit, randomly generated numbers. Seven samples were offered to each panellist: 1, Control; 2, HAB 1.5 ml; 3, OT 1.5 ml; 4, SB 1.5 ml; 5, HAB 3 ml; 6, OT 3 ml; 7, SB 3 ml. Unsalted bread, room temperature tap water and Ethanol 40% were provided to assessors to clean the palate between samples. The panel was instructed how to evaluate the samples and fill in the form previous the beginning of the evaluation. Linear unstructured graphical 100 mm scales oriented by description at both ends were used for sensory profile evaluation of 16 descriptors (Table 3.2). Sensory analysis was accomplished in 2 sessions (in separate days) within 1 week.

Table 3.1 - Sensory descriptors and their scales orientations I

Sensory descriptor	Scale orientation
Appearance:	
general appearance	0 %= very bad, 100 % = excellent
Smell:	
general pleasantness of the smell	0 % = very bad, 100 % = excellent
general intensity of the smell	0 % = imperceptible, 100 % = very strong
intensity of oregano smell	
Texture:	
general pleasantness of the texture	0 % = very bad, 100 % = excellent
juiciness	0 % = dry, 100% = juicy
chewiness	0 % = easy, 100% = difficult
Taste:	
general pleasantness of the taste	0 % = very bad, 100 % = excellent
general intensity of the taste	0 % = imperceptible, 100 % = very strong
Intensity of partial tastes:	
oregano taste, salty, bitter, astringent, pungent	0 % = imperceptible, 100 % = very strong
Overall evaluation of the sample:	0 % = very bad, 100 % = excellent

Table 3.2 - Sensory descriptors and their scales orientations II

Sensory descriptor	Scale orientation
Appearence:	
general appearence	0 % = very bad, 100 % = excellent
Smell:	
general pleasantness of the smell	0 % = very bad, 100 % = excellent
intensity of oregano smell	0 % = imperceptible, 100 % = very strong
Color:	
general likableness of the smell	0 % = dislike, 100 % = like
general intensity of the color	0 % = extremely light, 100 % = extremely dark
Texture:	
general pleasantness of the texture	0 % = very bad, 100 % = excellent
juiciness	0 % = dry, 100% = juicy
chewiness	0 % = easy, 100% = difficult
Taste:	
general pleasantness of the taste	0 % = very bad, 100 % = excellent
general intensity of the taste	0 % = imperceptible, 100 % = very strong
Intensity of partial tastes:	
oregano taste, salty, bitter, astringent, pungent	0 % = imperceptible, 100 % = very strong
Overall evaluation of the sample:	0 % = very bad, 100 % = excellent

3.6 Statistical analysis

3.6.1 Data analyzed after de microbial load calculations

Three independent experiments (replications) were conducted with two samples per treatment and per pathogen resulting in 6 observations per mean. Microbial load values were converted to log CFU g^{-1} and subjected to analysis of variance ANOVA for main effects of treatment. The Tukey adjustment was performed for multiple means comparisons, significant differences were determined using the statistical package STATISTICA 12 (Statsoft Inc, 2013, Oklahoma). Sensory data were also analyzed by one – way ANOVA for the main effects of treatment by groups and Tukey test was performed for separation of mean differences.

3.6.2 Data analyzed for modified blanching treatments by using OEO

Three independent experiments (replications) were conducted per treatment and per concentration resulting in 360 observations of the experimental unit per replication, due to the weight loss data was taken every minute during 6 hours of drying and then the moisture ratio was calculated. The moisture ratio variable was subjected to analysis of repeated measures factorial ANOVA, because multiple measurements were taken serially in time on the experimental unit. The Tukey test was performed for multiple means comparisons, significant differences were determined using the statistical package STATISTICA 13 (Statsoft Inc, 2013, Oklahoma). Sensory data were analysed by one – way ANOVA for the main effects of treatment by groups and Tukey post hoc test was used for comparison of means. Differences were considered significant at $P < 0.05$.

4 RESULTS AND DISCUSSION

4.1 Effect of OEO application on microbial load

As stated before *S. enteritidis* and *E. coli* are examples of those bacteria strains known for the ability to enter a VBNC state as a survival strategy. Preliminary experiments in this study indicated, that *E. coli* and *S. enteritidis* enter the VBNC state during drying, because there were no cultivable bacteria immediately after the drying process. Hence, executing pre-enrichment was necessary. In periods shorter than 4 hours, there were still very low numbers of growing cells, finally after 4 and 6 hours we were able to count these strains. Therefore, all the results are expressed after pre-enrichment with exception of raw inoculated controls (Table 4.1 and Table 4.2). In general, after the experiments were carried out with dose of 3 ml (0.057 ml l^{-1} air) OEO for both strains, the bacteria were not detected after the treatments. Consequently, the application of 3 ml of OEO was effective against both strains and thus lower concentrations were tested to define the MICs against *S. enteritidis* and *E. coli*, respectively.

Table 4.1 presents the survival of *S. enteritidis* on beef samples after drying subjected to pre-enrichment. Raw samples after inoculation contained on average 4.88 log CFU g^{-1} of *S. enteritidis*. After drying, not treated (NoEO) and $NaNO_2$ treated meat samples in both pre-enrichments had significantly higher numbers ($P < 0.05$) in population of *S. enteritidis* compared to the population of samples treated with 2 ml, showing a slight inhibition by sodium nitrite, similarly, Hospital et al.(2014) concluded that nitrite has a little effect for controlling *Enterobacteriaceae,* including *Salmonella* in commercially prepared foods. At the same time, at 6 hours of pre-enrichment there was a significant difference ($P = 0.039$) between the samples treated with 1.5 ml and 2 ml, where *S. enteritidis* has been reduced to a level of 1.24 log CFU g^{-1} for the higher dose. Therefore, a significant reduction of VBNC *S. enteritidis* ($P = 0.001$) after 6 hours of drying (55°C) with a concentration 0.038 ml l^{-1} vapours of the OEO was observed and it is considered as the minimal inhibitory concentration (MIC) against *S. enteritidis*. These results are consistent with the fact that the oregano essential oil possess strong antimicrobial properties (Botsoglou et al., 2003; Exarchou et al., 2002; Govaris et al., 2010) and that the application of the OEO in minced meats was found effective in inhibiting *S. enteritidis* (Barbosa et al., 2009).

Actually, there are no reported effects of an application of OEO during meat drying in the literature. However, in minced sheep meat was found that the addition of OEO at 0.6% (v/w) provided antimicrobial activity against *S. enteritidis* during storage at 4°C and 10°C (Govaris et al., 2010), indeed this higher percentage of OEO compare to the concentration used could be attributed to the effect of meat drying and therefore the concentration of OEO needed to eliminate the pathogen is lower. Additionally, Penalver et al.(2005) estimated the MIC for OEO against poultry origin strain of *S. enteritidis* as 0.25% (v/v) by *in vitro* assay where OEO showed higher antimicrobial activity compare to other types of essential oils

Table 4.2 shows the survival of *E. coli* on beef samples after 6 hours of drying subjected to pre-enrichment. Raw samples after inoculation contained on average 4.84 log CFU g^{-1} of *E. coli.* After drying, no significant differences ($P > 0.05$) were found between not treated (NoEO) and $NaNO_2$ treated meat samples for both pre-enrichments, presenting the synthetic antimicrobial agent as not very effective, likewise was presented in a previous study where nitrite had a little inhibitory effect against gram-negative enteric pathogens (Hospital et al., 2014). At the same time, no significant differences ($P > 0.05$) were observed between the counts of non-treated samples (NoEO) and the samples treated with 0.75 ml for both pre-enrichments. This result exhibited a failed performance against *E. coli* of the treatment conducted with 0.75 ml. In samples treated with 1.5 ml, the *E. coli* has been almost eliminated to a mean value of 1.161 log CFU g^{-1}. Hence, a significant reduction of VBNC *E. coli* ($P=0.03$) after 6 hours of drying (55°C) with a concentration 0.028 ml l^{-1} vapours of the OEO was found and was indicated as the MIC against *E. coli.* This reduction can be explained by the fact that OEO contains carvacrol, which is well known for its antimicrobial activity and is one of the most active against strains of *E. coli* (Burt and Reinders, 2003; Smith-Palmer et al., 1998).

Previous studies suggest that foodborne bacteria are able to survive drying (Levine et al., 2001). Some researchers studied the combination of drying with spice marinates (Nummer et al., 2004), but it is rather difficult to compare the results with the present study due to the fact, that no quantification of the essential oil content in the marinades was done. Application of OEO has been tested against *E. coli in vitro* rather than meat, regarding to Burt and Reinders (2003) study OEO was effective at 0.06 % (v/v) (0.625 ml l^{-1}) against *E. coli,* and the proposed mechanism causes the destruction of the cell membrane, which allowed them to pass easily through the pores of the membrane compared to the untreated control cells and inhibit the

growth of the microorganism, which finally resulted to the collapse of *E. coli* cells treated with OEO (Burt and Reinders, 2003). This fact is consistent with the one stated before by Lambert et al. (2001) where the carvacrol and thymol (principal constituents of OEO) render bacterial cell membranes permeable.

4.2 Effect of modified blanching by OEO on drying kinetics

Drying curves were used to determine the effect of OEO modified blanching treatments on the drying time. Figure 4.1, shows the experimentally determined moisture ratios of beef samples for oil treatment (OT), steam blanching (SB) and hot air blanching (HAB) versus drying time with 3 different doses 1.5 ml, 3 ml and 6 ml. The MR decreased exponentially with time in in all the used treatments which is consistent with the results reported in former studies (Afolabi et al., 2015; Cheng et al., 2015; İsmail et al., 2015; Kucerova et al., 2015). In Figure 4.1a, it might be seen that between the OT treated samples the dose of OEO affects the drying time, and even for 6 ml OEO the drying rate was slightly higher as compared to the rest of the doses in the initial period of drying, but no significant differences ($P > 0.05$) were found compared to samples treated with the other doses and the untreated control, which means that the OT itself slowed down the drying process and was not affected by the OEO dose, this is consistent with results from (Tunde-Akintunde et al., 2005) where combining oil/water blanching in drying kinetics of bell pepper presented lower drying rates.

Figure 4.1b, indicates a non-organized effect on the drying time due to different doses of OEO, instead the SB method itself was affecting the drying time because it was perceived that the drying rate was higher compared to the not treated control samples at the first 2 h of the drying. This fact can be explained by the effect of softening and partial cooking of the tissues, rendering the cell membranes more permeable to moisture transference (Brenndorfer et al., 1987). From Figure 4.1c, it might be concluding that HAB treatment was not affected by the different doses of OEO applied, moreover no statistical differences ($P > 0.05$) were found between treated samples and even no significant difference ($P > 0.05$) compared to the untreated control sample.

Table 4.1 - Mean (standard deviation) counts of Salmonella Enteritidis ATCC 13076 (log CFU g-1) of beef samples dried at 55°C for 6 hours in conventional oven subjected to pre-enrichment treatments for 4 hours and 6 hours and the control named as Raw.

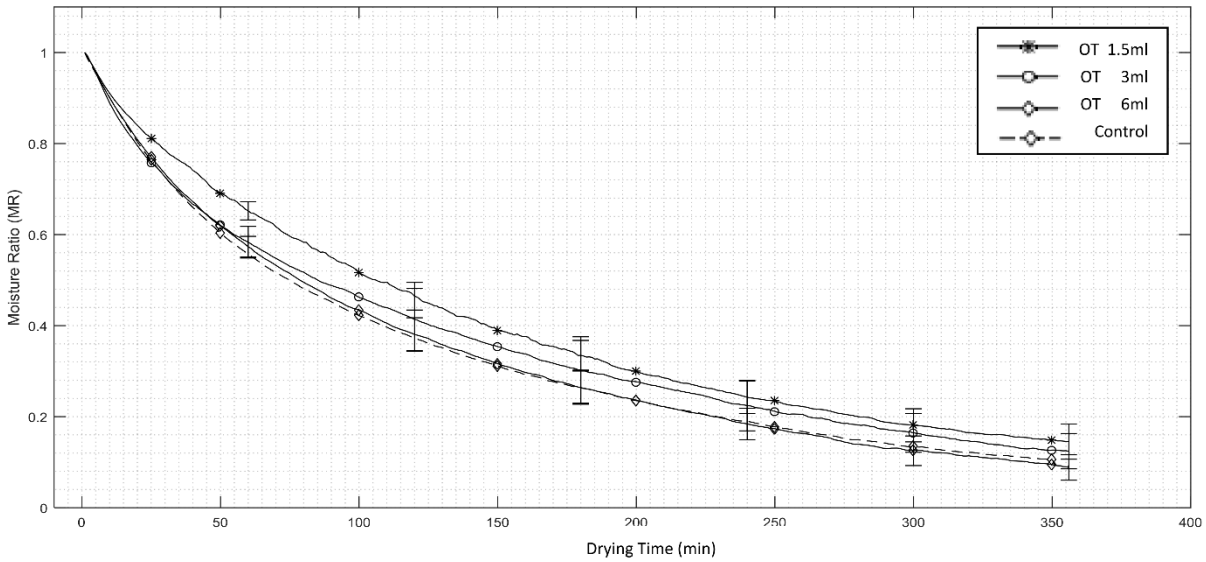
	Sample Type		
Dose of OEO	**PE 4H**	**PE 6H**	**Raw**
No EO	3.386 (1.00)^{c}	5.621 (0.88)^{c}	4.545 (0.16)^{a}
$NaNO_2$ (0,1 g)	2.694 (0.28)^{bc}	4.631 (0.48)^{bc}	5.722 (0.17)^{a}
1.5 ml	1.738 (1.85)^{abc}	3.446 (1,51)^{b}	4.709 (0.33)^{a}
2 ml	ND^{a}	1.24 (1.37)^{a}	4.613 (0.63)^{a}
3 ml	ND^{a}	ND^{a}	4.794 (0.23)^{a}

Dose of OEO, dose of oregano essential oil; NoEO, no essential oil; NaNO2 (1 g l-1), sodium nitrite; PE 4H, pre-enrichment 4 hours; PE 6H, pre-enrichment 6 hours; Raw, control of the inoculation efficiency; ND, not detected. Different letters as superscripts in the same column indicate significant difference (P<0.05).

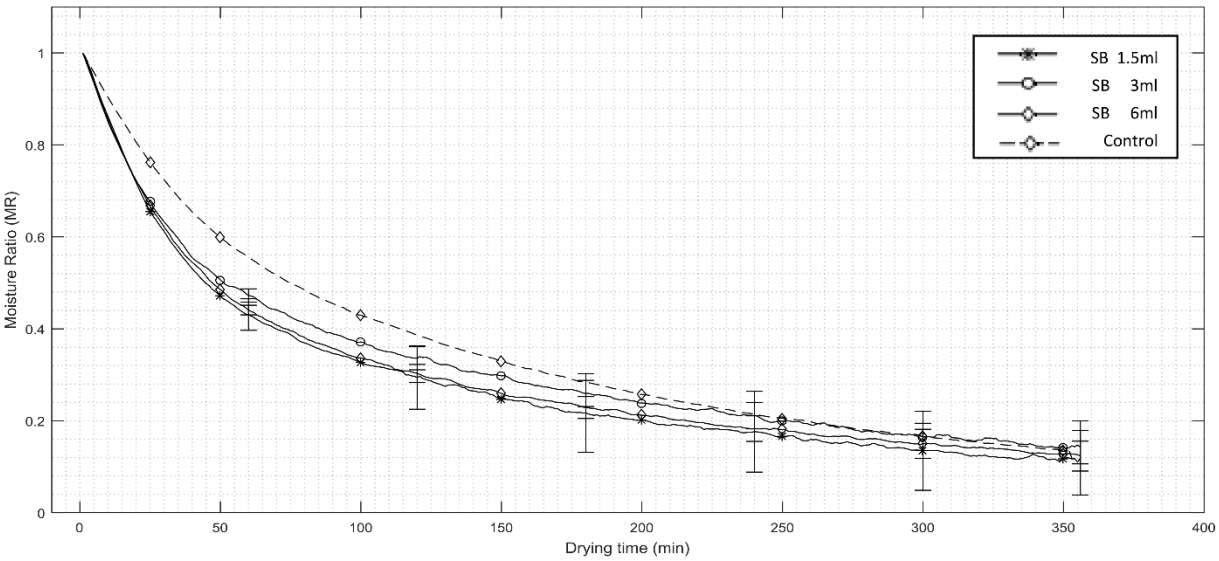
Table 4.2 - Mean (standard deviation) counts of Escherichia Coli ATCC 25922 (log CFU g-1) of beef samples dried at 55°C for 6 hours in conventional oven subjected to pre-enrichment treatments for 4 hours and 6 hours and the control named as Raw.

	Sample Treatment		
Dose of OEO	**PE 4H**	**PE 6H**	**Raw**
No EO	4.324 (1.29)^{c}	4.478 (1.39)^{c}	4.733 (0.45)^{a}
$NaNO_2$ (0,1 g)	1.872 (1.36)^{abc}	3.903 (1.49)^{bc}	5.037 (0.68)^{a}
0.75 ml	1.667 (1.19)^{abc}	3.478 (1.44)^{bc}	4.636 (0.50)^{a}
1.5 ml	0.851 (2.08)^{a}	1.161 (2.84)^{a}	5.126 (0.77)^{a}
3 ml	ND^{a}	ND^{a}	4.675 (0.09)^{a}

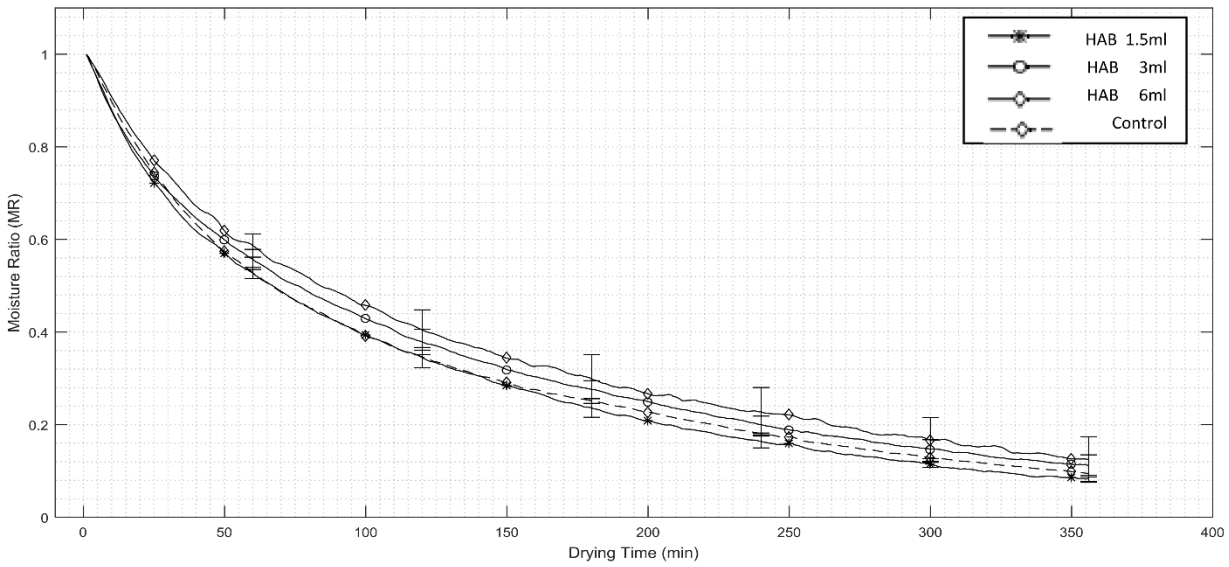
Dose of OEO, dose of oregano essential oil; NoEO, no essential oil; $NaNO_2$ (1 g l^{-1}), sodium nitrite; PE 6H, pre-enrichment 6 hours; Raw, control of the inoculation efficiency; ND, not detected. Different letters as superscripts in the same column indicate significant difference (P<0.05).



(a) Oil treatment (OT)



(b) Steam blanching (SB)



(c) Hot air blanching (HAB)

Figure 4.1 - Experimental moisture ratio of beef samples dried in climate box as a function of drying time with standard deviations at different oregano essential oil doses

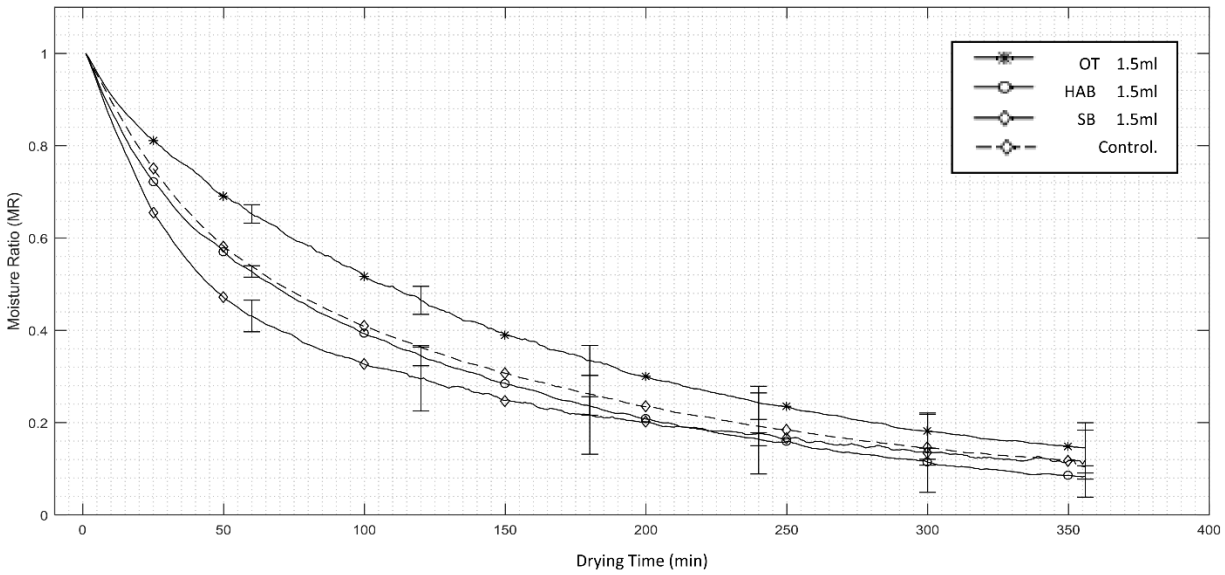
Figure 4.2, presents the drying curves (time versus MR) of the beef samples with all the used modified blanching treatments OT, SB and HAB during 6 hours of drying per dose of OEO. As indicated in these curves, the moisture ratio decreased continuously with drying time for all doses. As can be seen in Figure 4.2a, the drying rate of steam blanched samples for dose of 1.5 ml was found to be significantly higher ($P < 0.05$) during initial stage of drying compared to OT treated samples. Also after the preliminary phase of drying, the total time required for the drying beef control samples to reach the MR of 20% at 55°C is about 250 min while for steam blanched ones only 200 minutes are needed. Therefore, SB treatment can reduce the drying time with approximately 14% to reach the same moisture content at that temperature with dose of 1.5 ml OEO, which is very similar with results reported by (Cheng et al., 2015) in drying characteristics of blanched cherry tomato prior to drying where the drying time was reduced due to the effect of the pre-treatment.

In Figure 4.2b and Figure 4.2c, this tendency of faster drying rate caused by SB compared to the control is not repeated in the latter stages of drying, furthermore, at 20% of MR not significant differences ($P > 0.05$) were found between controls and SB samples with doses of 3 ml and 6 ml OEO, respectively, and for those doses the drying time needed to achieve the 20% of MR was higher, compared to 1.5 ml OEO

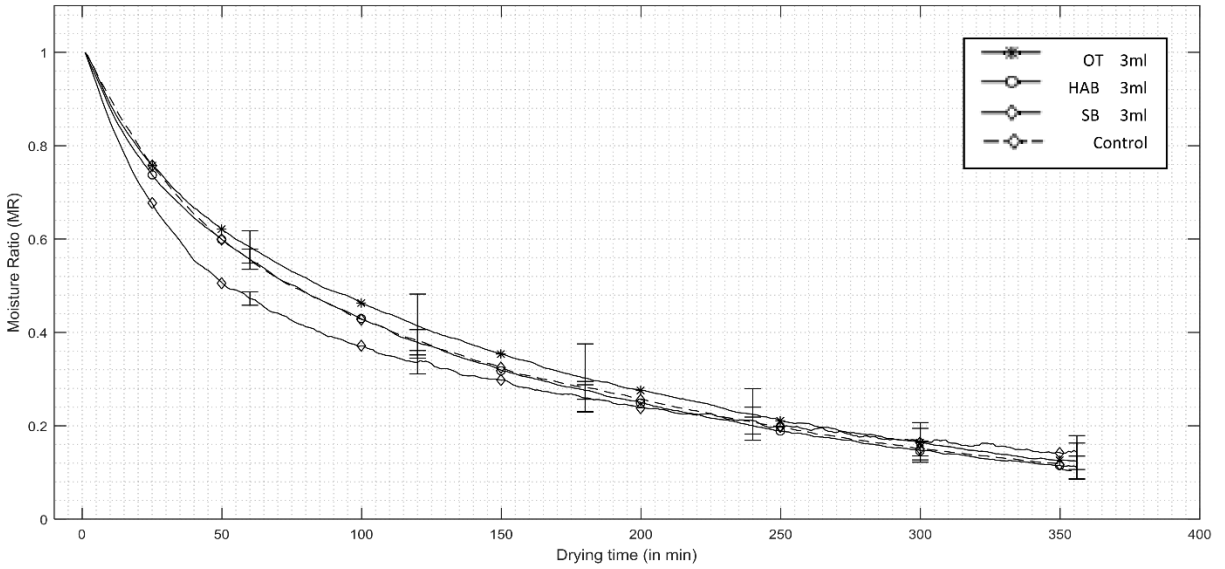
probably due to the increase of the essential oil concentration. These findings are in agreement with results reported in previous studies whereas the drying time increases, the effects of blanching on drying time are becoming smaller for the cherry tomatoes (Cheng et al., 2015).

OT resulted to be the treatment which delays the drying process for all doses at the initial period of drying and likewise at the falling period (*cf.*Figure 4.2), these findings correspond to the results of a similar dipping study of bell pepper in palm oil where the coating property of the surface is due to the wax-like oil. The presence of this coating and the effect of the hydrophobicity of the palm oil, in present case the sunflower oil acted to reduce the moisture movement slightly (Tunde-Akintunde et al., 2005) and both cause a lower drying rate throughout the drying process. In Figure 4.2b and Figure 4.2c, HAB treatment showed a similar behaviour as OT decreasing the drying rate, in addition no significant differences ($P > 0.05$) were presented between the samples treated with 3 ml and 6 ml of OEO for both treatments and also no significant differences ($P > 0.05$) between control samples for both concentrations. Summing up, the results showed that steam blanching treatment was more effective itself in reducing the beef drying time compared to the other OEO modified blanching treatments.

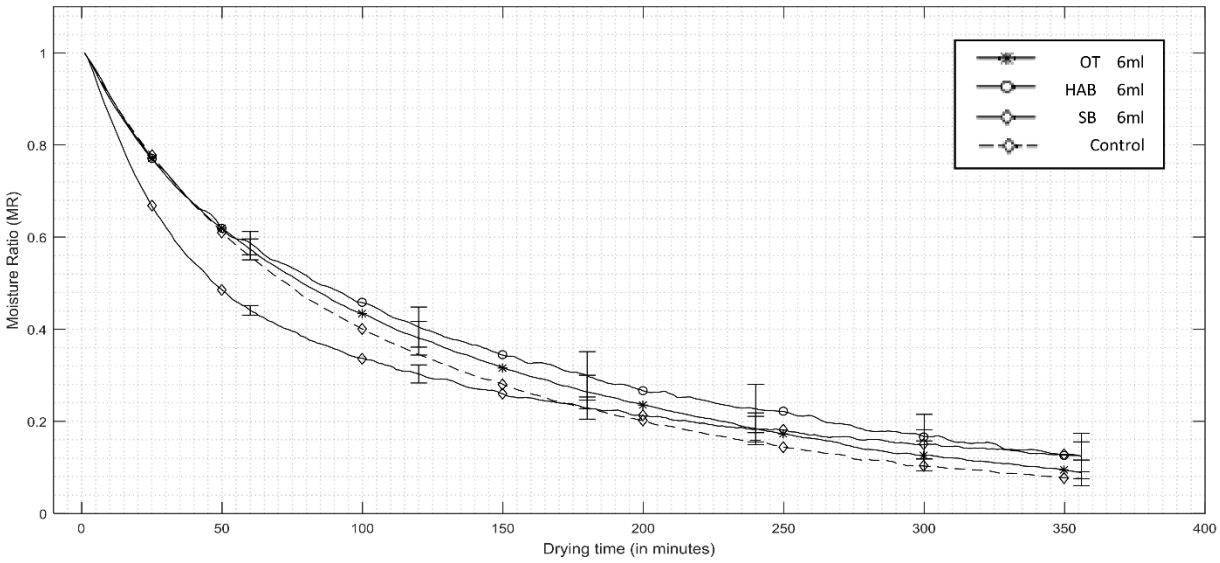
In general, according to the overall progress of the drying curves for all treatments it is possible to observe higher drying rates at the primary stages of the drying and dropping drying rates at the latter stages of the drying when the process entered the falling rate period. This behaviour is consistent with other studies and particularly in the meat drying process, where the incidence of lower drying rates at the final stage of the drying could be explained by the action of denatured proteins which exposed to heat form a gel matrix causing difficult movement of water from the interior part of the meat (Kucerova et al., 2015; Nathakaranakule et al., 2007).



(a) Dose of 1.5 ml



(b) Dose of 3 ml



(c) Dose of 6 ml

Figure 4.2 - Experimental moisture ratio of beef samples dried in climate box as a function of drying time with standard deviations for OT, SB and HAB treatments with oregano essential oil and control

4.3 Sensory analysis

The first part of the analysis was recollected for the different treatments applied for the calculation of microbial load by using oregano essential oil and different concentrations. The second part of the present analysis was practiced to different modified blanching treatments by using oregano essential oil with different concentrations.

4.3.1 First sensory analysis applied

In Table 4.3 the summary of general sensory attributes for dried meat samples is presented. Panellists ranked the 0.75 ml and 0.75 ml + salt treated samples as superiors ($P < 0.05$) to the control in terms of general pleasantness of the smell and as more intensive concerning general intensity of the smell, intensity of the oregano smell and general intensity of the taste. According to Hulankova et al.(2013) the odour of cooked meat samples treated with higher than 0.2 % (v/w) OEO was found as too strong and no longer pleasant. In this case with 0.75 ml OEO (0.014 ml l^{-1} vapours), the odour was more intensive than the control and the addition of oregano improved the pleasantness of the smell (*cf.* Figure 4.3). Moreover, there are more studies which stated better sensory properties of minced beef or sheep meat treated with 0.8 - 1 % of OEO in comparison to control (Govaris et al., 2010; Skandamis and Nychas, 2001; Tsigarida et al., 2000).

Importantly, the 0.75 ml and 0.75 ml + salt treated samples were scored as superiors ($P < 0.05$) to the 1.5 ml and 1.5 ml + salt treated samples regarding to general pleasantness of the taste. Written comments indicated that it was perceived a sweet flavour imparted by oregano essential oil, and reduced the perceived saltiness of the jerky. Consequently, the two samples treated with 1.5 ml were scored lower ($P < 0.05$) than all the other samples in relation to general pleasantness of the taste and were described as "unusual" or metal and sour flavour. These results are in agreement with those obtained by Govaris et al. (2010) where the taste of minced sheep meat treated with a lower concentration, OEO at 0.6% (v/w), was scored as significantly higher compare to control as well as samples treated with OEO at 0.9% (v/w) throughout the refrigerated storage. According to the results, no significant differences were presented concerning general appearance and general pleasantness of the texture between samples and both attributes showed scores close to the control value which is not significantly different in sensory terms.

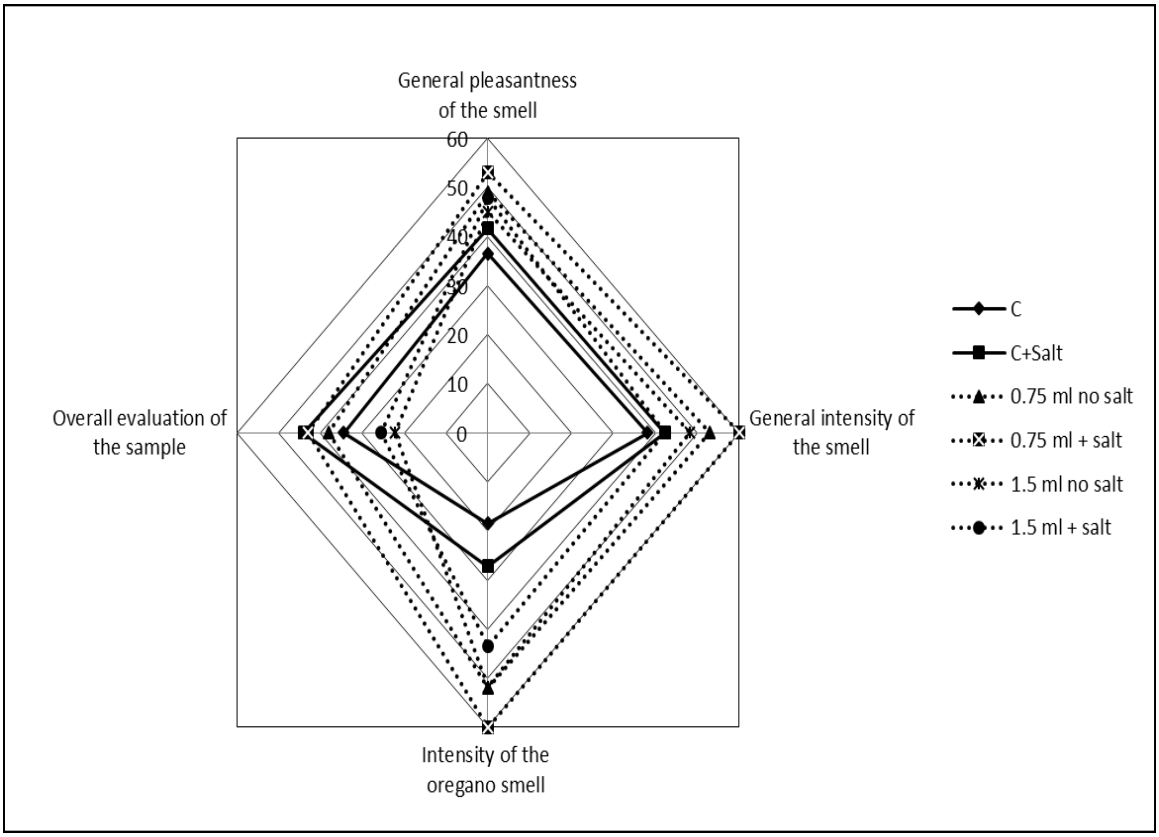


Figure 4.3 - Spider diagram of sensory profiling of the meat dried samples regarding to the odour descriptors and the overall evaluation of the sample

C=Control, C + salt= Control + salt, 0.75 ml no salt= treated 0.75 ml no salt, 0.75 ml + salt= treated 0.75 ml + salt, 1.5 ml no salt= treated 1.5 ml no salt and 0.75 ml + salt= treated 0.75 ml + salt.

Both samples treated with 1.5 ml (*cf.* Table 4.4) showed higher values for bitter, astringent and pungent attributes ($P < 0.05$) compared with the rest of the samples and also presented higher values ($P < 0.05$) than the control samples in terms of oregano taste, which could explain the reason why the samples treated with 1.5 ml were judged as bitter, astringent and pungent. With respect to the 0.75 ml and 0.75 ml + salt treated samples (*cf.* Table 4.4) there were rated as intensive in case of oregano taste ($P < 0.05$) compared to control, and judged as more intensive for juiciness, which is one essential descriptor for the texture attribute.

In terms of chewiness there was no significant difference between all the samples, intriguingly the mean values could indicate that more difficult chewiness is linked to the application of essential oil, due to the presence of higher values in samples

treated with 0.75 ml and 1.5 ml OEO (*cf.* Table 4.4). According to Konieczny et al. (2007) ensuring chewiness is one of the most important sensory attributes in jerky type snack foods and regarding the present results limits the consumer acceptability in terms of general pleasantness of the texture for dried meat samples (*cf.* Table 4.3). With respect to overall evaluation of the sample (*cf.* Table 4.4), samples treated with 1.5 ml OEO were significantly less acceptable ($P < 0.05$) compared to samples treated with 0.75 ml OEO and the control. These results are similar to those presented by Govaris et al.(2010) where compared to the control, the scores of overall acceptability of minced sheep meat treated with a higher concentration in that particular case: OEO at 0.9% (v/w), were lower up to the 6^{th} day of storage. The present findings are in agreement with the fact, that oregano is common spice used for meat marination (Friedman et al., 2007) and also preceding observations that OEO may enhance the organoleptic properties of meat (Burt, 2004; Govaris et al., 2010; Skandamis and Nychas, 2001).

Table 4.3 - Mean in % (standard deviation in %) general sensory attributes scores by dried meat sample.

Sensory Attribute	Sample Type					
	Control	Control + salt	Treated 0.75 ml	Treated 0.75 ml + salt	Treated 1.5 ml	Treated 1.5 ml + salt
General Appearance	38 (17)[a]	41 (17)[a]	47 (19)[a]	47 (20)[a]	40 (19)[a]	48 (21)[a]
General pleasantness of the smell	37 (14)[a]	42 (17)[b]	49 (18)[bc]	53 (21)[c]	45 (15)[bc]	48 (15)[bc]
General pleasantness of the texture	38 (20)[a]	41 (19)[a]	38 (17)[a]	40 (18)[a]	33 (17)[a]	34 (17)[a]
General pleasantness of the taste	34 (22)[ab]	41 (24)[b]	36 (16)[b]	43 (19)[b]	24 (15)[a]	26 (15)[a]
General intensity of the smell	38 (20)[a]	42 (21)[ab]	53 (22)[c]	60 (18)[c]	48 (19)[bc]	42 (20)[b]
Intensity of the oregano smell	18 (16)[a]	27 (23)[a]	52 (26)[b]	60 (21)[c]	52 (19)[b]	43 (20)[b]
General intensity of the taste	46 (17)[a]	53 (15)[ab]	56 (19)[b]	62 (15)[c]	63 (20)[c]	63 (19)[c]

Means in the same row followed by the same letter are not significantly different (P > 0.05). Linear unstructured graphical oriented 100 mm scales were used for evaluation of general appearance, general pleasantness of the smell, general pleasantness of the texture, general pleasantness of the taste (0 % = very bad, 100% = excellent), general intensity of the smell, intensity of oregano smell and general intensity of taste (0 % = imperceptible, 100 % = very strong).

Table 4.4 - Mean in % (standard deviation in %) scores for taste, texture and the overall liking sensory attributes by dried meat sample.

Sensory Attribute	Sample Type					
	Control	Control + salt	Treated 0.75 ml	Treated 0.75 ml + salt	Treated 1.5 ml	Treated 1.5 ml + salt
Oregano taste	19	28	55	59	64	63
	(15)^{a}	(19)^{a}	(22)^{b}	(19)^{b}	(19)^{b}	(17)^{b}
Salty	26	34	25	32	25	31
	(14)^{a}	(18)^{a}	(18)^{a}	(20)^{a}	(18)^{a}	(21)^{a}
Bitter	17	14	30	26	45	42
	(19)^{a}	(17)^{a}	(26)^{b}	(23)^{ab}	(29)^{c}	(30)^{c}
Astringent	13	13	24	21	38	38
	(17)^{a}	(15)^{a}	(25)^{ab}	(21)^{a}	(32)^{b}	(30)^{b}
Pungent	12	13	23	21	45	44
	(17)^{a}	(18)^{a}	(22)^{b}	(20)^{ab}	(29)^{c}	(30)^{c}
Juiciness	24	29	31	37	24	24
	(18)^{a}	(19)^{ab}	(19)^{bc}	(21)^{c}	(18)^{b}	(17)^{b}
Chewiness	50	48	55	56	57	60
	(22)^{a}	(22)^{a}	(23)^{a}	(23)^{a}	(24)^{a}	(23)^{a}
Overall evaluation of the sample	34	44	38	43	22	25
	(22)^{b}	(24)^{c}	(21)^{b}	(19)^{bc}	(14)^{a}	(15)^{a}

Means in the same row followed by the same letter are not significantly different (P > 0.05). Linear unstructured graphical oriented 100 mm scales were used for evaluation of overall evaluation of the sample (0 % = very bad, 100% = excellent), intensity of oregano, salty, bitter, astringent and pungent tastes (0 % = imperceptible, 100 % = very strong, juiciness (0 % = dry, 100 % = juicy) and chewiness (0 % = easy, 100 % = difficult).

4.3.2 Second sensory analysis applied

In Table 4.5 the summary of general sensory attributes for dried meat samples is presented. Panellists ranked OT 1.5 ml sample as superior ($P < 0.05$) to SB 1.5 ml and SB 3 ml samples in terms of general appearance, general pleasantness of the smell and general likableness of the colour and as more intensive concerning general intensity of the taste. The appearance of the meat products determine how consumers perceive their quality (Resurreccion, 2004), in this sense, the intensity of the dark reddish colour and brightness appearance from the HAB and OT treated samples may have positively contributed to their higher acceptance, and in SB treated samples, the brownish colour and a course texture seemed to contribute to its lower appearance scores.

Samples treated with OT 3 ml were scored as more intensive regarding intensity of the oregano smell comparing to the control and SB 1.5 ml samples. These findings are in agreement with Hulankova et al. (2013)where the odour of cooked meat samples treated with higher than 0.2 % (v/w) OEO was found as firmly strong. In this study, for HAB and OT treatments with 1.5 ml and 3 ml OEO doses, respectively, the odour was more intensive than the control and adding OEO enriched the pleasantness of the smell (*cf.* Table 4.5). c. (Govaris et al., 2010; Skandamis and Nychas, 2001; Tsigarida et al., 2000).

Notably, the control and HAB 3 ml samples were scored as superiors to the other treated samples in relation to general pleasantness of the taste, where OT 3 ml and SB 1.5 ml samples were the lower scored in this matter. In contrast, results proposed by Govaris et al. (2010) where the taste of minced sheep meat treated with a lower OEO concentration: 0.6% (v/w), was recorded as significantly higher compare to samples treated with OEO concentration of 0.9% (v/w) throughout the refrigerated storage. Writing comments described OT 3 ml sample as "artificial" or metal flavour and for SB 1.5 ml sample no taste was perceived probably due to the aggressive effect of the SB treatment which beat the OEO taste, this is consistent with the effect on the flavour loss explained by Sotome et al. (2009) for potato blanching with saturated steam caused by the dissolution of a portion of solid content into the water steam from the potato. According to Al-Khusaibi & Niranjan (2012), the study of the effect of blanching potatoes on the oil uptake of fried potato slices, indicated that the oil uptake of blanched slices is lower that unblanched ones, since the cellular

structure breakdown does not facilitate oil absorption which could be the same effect occurred in meat samples of this study and resulting on taste loss of the SB sample.

OT 3 ml samples (*cf.*Table 4.6) indicated higher values for bitter, astringent and pungent attributes ($P < 0.05$) compared with the rest of the samples and also pointed to higher values ($P < 0.05$) than the control samples in terms of oregano taste, which justify why the samples treated with OT 3 ml were judged as bitter, astringent and pungent. With respect to the SB 1.5 ml treated samples (*cf.* Table 4.6) there were rated as less intensive in case of oregano taste compared to other treated samples, and referred as less intensive for juiciness, which is absolutely critical descriptor for the texture attribute.

In terms of chewiness there was no significant difference between all the samples, interestingly the mean values draw a conclusion that the application of essential oil is not linked with easy chewiness (*cf.* Table 4.5) which is in agreement with results from the previous part: sensory analysis of microbial load, in samples treated with HAB 1.5 ml and lower doses. Ensuring chewiness is one of the most critical sensory qualities in jerky type snack foods (Konieczny et al., 2007) and regarding the present results limited the consumer acceptability in terms of general pleasantness of the texture for SB 1.5 ml treated samples which were significantly less pleasantness ($P < 0.05$) compared to HAB 3 ml, OT 1.5 ml and OT 3 ml treated samples (*cf.* Table 4.5).

With respect to overall evaluation of the sample (*cf.* Table 4.6), the control, HAB 3 ml and OT 1.5 ml treated samples were significantly high acceptable ($P < 0.05$) compared to samples treated with SB 1.5 ml. These results contrasted those presented by Hernández et al. (2016) where HAB 1.5 ml salted and unsalted treated samples scored lower on overall acceptability of the sample compared to the control and samples treated with less dose of OEO, these variances may be explained by the wide offer of treatments in the current study which could be perceived different by panellists in sensory terms and their inclinations to a more intensive oregano taste. At the end, the present findings are in agreement with reported studies that OEO could enhance the organoleptic properties of meat (Burt, 2004; Govaris et al., 2010; Skandamis and Nychas, 2001).

Table 4.5 - Mean (standard deviation) general sensory attribute scores (%) by dried meat sample.

Sensory Attribute	Sample treatment type						
	Control	HAB 1.5 ml	OT 1.5 ml	SB 1.5 ml	HAB 3 ml	OT 3 ml	SB 3 ml
General Appearance	66 (15)^{b}	68 (23)^{b}	78 (10)^{b}	33 (21)^{a}	67 (15)^{b}	71 (19)^{b}	42. (24)^{a}
General pleasantness of the smell	57 (17)^{abc}	62 (15)^{bc}	71 (11)^{c}	46 (11)^{a}	61 (18)^{abc}	63 (14)^{bc}	52 (16)^{ab}
General likableness of the color	72 (20)^{b}	69 (21)^{b}	83 (10)^{b}	23 (24)^{a}	73 (15)^{b}	75 (20)^{b}	46 (27)^{a}
General pleasantness of the texture	53 (21)^{ab}	50 (20)^{ab}	59 (22)^{b}	37 (21)^{a}	60 (18)^{b}	59 (21)^{b}	43 (18)^{ab}
General pleasantness of the taste	67 (15)^{c}	54 (19)^{abc}	49 (23)^{abc}	39 (22)^{ab}	61 (16)^{bc}	37 (26)^{a}	55 (23)^{abc}
Intensity of the oregano smell	23 (20)^{a}	40 (21)^{ab}	40 (25)^{ab}	25 (17)^{a}	40 (24)^{ab}	49 (22)^{b}	33 (22)^{ab}
General intensity of the color	49 (22)^{a}	74 (13)^{b}	69 (13)^{b}	60 (26)^{ab}	71 (12)^{b}	75 (14)^{b}	69 (16)^{b}
General intensity of the taste	59 (23)^{b}	49 (10)^{ab}	60 (15)^{b}	32 (19)^{a}	57 (18)^{b}	57 (26)^{b}	47 (18)^{ab}

Control, untreated sample; HAB 1.5 ml; OT 1.5 ml; SB 1.5 ml; HAB 3 ml; OT 3 ml; SB 3 ml. Means in the same row followed by the same letter are not significantly different (P > 0.05). Linear unstructured graphical oriented 100 mm scales were used for evaluation of general appearance, general pleasantness of the smell, general pleasantness of the texture, general pleasantness of the taste (0 % = very bad, 100% = excellent), general likableness of the color (0 % = dislike, 100% = like), intensity of oregano smell, general intensity of taste (0 % = imperceptible, 100 % = very strong) and general intensity of the color (0 % = extremely light, 100 % = extremely dark).

Table 4.6 - Mean (standard deviation) taste, texture and the overall liking sensory attribute scores (%) by dried meat sample.

Sensory Attribute	Sample treatment type						
	Control	HAB 1.5 ml	OT 1.5 ml	SB 1.5 ml	HAB 3 ml	OT 3 ml	SB 3 ml
Oregano taste	32 (29)^{a}	46 (22)^{ab}	54 (18)^{ab}	35 (22)^{ab}	38 (19)^{ab}	56 (26)^{b}	40 (19)^{ab}
Salty	29 (22)^{a}	20 (18)^{a}	25 (19)^{a}	16 (18)^{a}	24 (17)^{a}	22 (18)^{a}	22 (18)^{a}
Bitter	10 (16)^{a}	15 (16)^{a}	28 (24)^{ab}	14 (17)^{a}	16 (21)^{a}	43 (27)^{b}	15 (20)^{a}
Adstringens	8 (13)^{a}	11 (13)^{a}	26 (25)^{ab}	12 (16)^{a}	16 (17)^{a}	43 (30)^{b}	17 (22)^{a}
Pungent	13 (17)^{a}	15 (15)^{a}	27 (20)^{ab}	12 (17)^{a}	14 (11)^{a}	45 (28)^{b}	18 (23)^{a}
Juiciness	34 (26)^{abc}	42 (24)^{bc}	39 (16)^{bc}	14 (13)^{a}	48 (21)^{b}	45 (29)^{bc}	24 (18)^{ac}
Chewiness	51 (23)^{a}	42 (27)^{a}	43 (18)^{a}	45 (31)^{a}	58 (19)^{a}	51 (26)^{a}	32 (24)^{a}
Overall evaluation of the sample	69 (17)^{c}	51 (16)^{abc}	55 (23)^{bc}	33 (18)^{a}	62 (17)^{c}	41 (22)^{ab}	50 (22)^{abc}

Control, untreated sample; HAB 1.5 ml; OT 1.5 ml; SB 1.5 ml; HAB 3 ml; OT 3 ml; SB 3 ml. Means in the same row followed by the same letter are not significantly different (P > 0.05). Linear unstructured graphical oriented 100 mm scales were used for evaluation of overall evaluation of the sample (0 % = very bad, 100% = excellent), oregano taste, salty, bitter, astringent and pungent tastes (0 % = imperceptible, 100 % = very strong, juiciness (0 % = dry, 100 % = juicy) and chewiness (0 % = difficult, 100 % = easy).

5 CONCLUSIONS

The results obtained from this thesis reveal that the application of the oregano essential oil (OEO) in meat effectively inhibits *Salmonella enteritidis* and *Escherichia coli*, due to significant reduction of *E. coli* and *S. enteritidis* viable counts after 6 hours of drying at 55°C with concentrations of 2 ml (0.038 ml l^{-1} air vapours) and 1.5 ml (0.028 ml l^{-1} air vapours) of OEO, respectively. On the other hand, the samples treated with 0.75 ml (0.014 ml l^{-1} air) of OEO were judged as better from sensory point of view and the respondents considered that the addition of oregano improved general pleasantness of smell, consequently there were more attractive for consuming compare to the original one. Additionally, according to the results a higher concentration of OEO influence the quality of the product, therefore for a highest consumer acceptability it is recommended a concentration between 0.75 and 1.5 ml OEO to be tested in combination with other natural preservatives such as organic acids, other types of essential oils, lysozyme, nisine, conventional preservatives or physical methods according to the hurdle technology concept.

This is the first report that exhibits that the application of OEO may be a highly effective alternative to synthetic preservatives, particularly sodium nitrite, for reducing microbial load in dried meat and presents a value-added meat product by using OEO for enhancement of the food safety and an acceptable sensorial response from consumers. For further research, the dose optimization of the application and combination with other methods are recommended in terms of promoting the industrial scale production.

The drying kinetics of the beef samples shows that modified blanching affects the drying time but the application of OEO doses does not affect the drying process. In the case of OT, this treatment itself slowed down the drying process and was not affected by the OEO dose. For HAB method, the results showed a similar tendency as OT, where the treatment had an effect of decreasing the drying rate during the drying, although it was not highly significant as OT. Whereas, SB presented a non-organized effect on the drying time by the application of different OEO doses and instead SB itself was affecting the drying process, shortening the drying time at the first part of the drying.

On the other hand, samples treated with OT 1.5 ml where judged as superior to SB 1.5 ml and SB 3 ml samples in terms of general appearance, general pleasantness of

the smell, general pleasantness of the texture and general likableness of the colour and as more intensive concerning general intensity of the taste. Samples treated with HAB 3 ml were scored as superiors to the other treated samples in relation to general pleasantness of the taste, where OT 3 ml and SB 1.5 ml samples were the lower scored. In conclusion, HAB 1.5 ml, HAB 3 ml and OT 1.5 samples were judged as better from sensory point of view and the respondents considered that adding OEO enriched the pleasantness of the smell.

Since a higher concentration of OEO has not a highly significant effect on the drying kinetics, but it highly influences the quality of the product, it is recommended to test HAB and OT treatments in dose of 1.5 ml OEO for further research in combination with other SB technologies, such as, superheated steam and hot water microdroplets, which will help to the efficiency of the drying process and aid with a less aggressive influence on sensory properties than the current SB treatment.

The further trend, is towards the use of treatments which bring a positive effect to the drying kinetics of dried meat samples with the application of plant essential oils, and deliver products that are less severe preserved, have higher quality, are more natural, free from additives and nutritionally healthier. To the best of our knowledge, this study brings a new knowledge about the drying behaviour of beef samples, presenting a value-added meat product by using OEO with acceptable sensorial response from consumers which might be important for promoting the industrial scale production.

REFERENCES

Aberle, E.D., Forrest, J.C., Gerrard, E.D., Mills, W.E., 2001. Principles of meat science. Kendall Hunt.

Afolabi, T.J., Tunde-Akintunde, T.Y., Adeyanju, J. a, 2015. Mathematical modeling of drying kinetics of untreated and pretreated cocoyam slices. J. Food Sci. Technol. 52, 2731–40. doi:10.1007/s13197-014-1365-z

Agriculture, U.S.D., 1996. Standards and Labeling Policy Book. Food Safety and Inspection Service, Meat and Poultry Inspection Technical Services, Standards and Labeling Division, Washington D.C.

Akanbi, C.T., Olumese, A.O., Taiwo, K.A., Ojo, A., Akinwande, B.A., 2003. Effect of blanching medium on drying and storage characteristics of pepper. Niger. Dry. Symp. Ser. 1, 95–107.

Al-Khusaibi, M.K., Niranjan, K., 2012. The Impact of Blanching and High-Pressure Pretreatments on Oil Uptake of Fried Potato Slices. Food Bioprocess Technol. 5, 2392–2400. doi:10.1007/s11947-011-0562-2

Allen, K., Cornforth, D., Whittier, D., Vasavada, M., Nummer, B., 2007. Evaluation of high humidity and wet marinade methods for pasteurization of jerky. J. Food Sci. 72. doi:10.1111/j.1750-3841.2007.00458.x

An, K., Li, H., Zhao, D., Ding, S., Tao, H., Wang, Z., 2013. Effect of Osmotic Dehydration with Pulsed Vacuum on Hot-Air Drying Kinetics and Quality Attributes of Cherry Tomatoes. Dry. Technol. 31, 698–706. doi:10.1080/07373937.2012.755192

Andrews, J.M., Andrews, J.M., 2001. Determination of minimum inhibitory concentrations. J. Antimicrob. Chemother. 48 Suppl 1, 5–16. doi:10.1093/jac/48.suppl_1.5

Arevalo-Pinedo, A., Xidieh Murr, F.E., 2007. Influence of pre-treatments on the drying kinetics during vacuum drying of carrot and pumpkin. J. Food Eng. 80, 152–156. doi:10.1016/j.jfoodeng.2006.05.005

Arroqui, C., Rumsey, T.R., Lopez, A., Virseda, P., 2002. Losses by diffusion of ascorbic acid during recycled water blanching of potato tissue. J. Food Eng. 52, 25–30. doi:10.1016/S0260-8774(01)00081-4

Aversa, M., Curcio, S., Calabr??, V., Iorio, G., 2007. An analysis of the transport

phenomena occurring during food drying process. J. Food Eng. 78, 922–932. doi:10.1016/j.jfoodeng.2005.12.005

Ayala-Zavala, J.F., Oms-Oliu, G., Odriozola-Serrano, I., González-Aguilar, G.A., Álvarez-Parrilla, E., Martín-Belloso, O., 2008. Bio-preservation of fresh-cut tomatoes using natural antimicrobials. Eur. Food Res. Technol. 226, 1047–1055. doi:10.1007/s00217-007-0630-z

Baines, D., Seal, R., 2012. Natural food additives, ingredients and flavourings, Elsevier. ed.

Baird, B.E., Lucia, L.M., Acuff, G.R., Harris, K.B., Savell, J.W., 2006. Beef hide antimicrobial interventions as a means of reducing bacterial contamination. Meat Sci. 73, 245–248. doi:10.1016/j.meatsci.2005.11.023

Baker, C., 1997. Industrial drying of foods. Chapman & Hall.

Barbosa, L.N., Rall, V.L.M., Fernandes, A.A.H., Ushimaru, P.I., da Silva Probst, I., Fernandes, A., 2009. Essential oils against foodborne pathogens and spoilage bacteria in minced meat. Foodborne Pathog. Dis. 6, 725–728. doi:10.1089/fpd.2009.0282

Barbosa-Cánovas, G. V., 1996. Dehydration of Foods, Chapman And Hall. doi:10.1136/bmj.2.4260.254

Barros, L., Calhelha, R.C., Vaz, J. a., Ferreira, I.C.F.R., Baptista, P., Estevinho, L.M., 2006. Antimicrobial activity and bioactive compounds of Portuguese wild edible mushrooms methanolic extracts. Eur. Food Res. Technol. 225, 151–156. doi:10.1007/s00217-006-0394-x

Belessiotis, V., Delyannis, E., 2011. Solar drying. Sol. Energy 85, 1665–1691. doi:10.1016/j.solener.2009.10.001

Beuchat, L.R., 1994. Natural antimicrobial systems and food preservation, in: Antimicrobial Properties of Spices and Their Essential Oils. pp. 167–179.

Blom, H., Mørtvedt, C., 1991. Antimicrobial substances produced by food associated microorganisms. Biochem. Soc. Trans. 19, 694–698.

Borowski, A.G., Ingham, S.C., Ingham, B.H., 2009. Validation of ground-and-formed beef jerky processes using commercial lactic acid bacteria starter cultures as pathogen surrogates. J. Food Prot. 72, 1234–1247.

Bosilevac, J.M., Nou, X., Barkocy-Gallagher, G. a, Arthur, T.M., Koohmaraie, M., 2006. Treatments using hot water instead of lactic acid reduce levels of aerobic

bacteria and *Enterobacteriaceae* and reduce the prevalence of *Escherichia coil* O157:H7 on preevisceration beef carcasses. J. Food Prot. 69, 1808–13.

Botsoglou, N.A., Govaris, A., Botsoglou, E.N., Grigoropoulou, S.H., Papageorgiou, G., 2003. Antioxidant activity of dietary oregano essential oil and alpha-tocopheryl acetate supplementation in long-term frozen stored turkey meat. J. Agric. Food Chem. 51, 2930–2936. doi:10.1021/jf021034o

Brenndorfer, B., Kennedy, L., Mrema, G., 1987. Solar dryers: their role in post-harvest processing. Commonwealth Secretariat.

Burnham, G.M., Hanson, D.J., Koshick, C.M., Ingham, S.C., 2008. Death of Salmonella serovars, Escherichia coli O157 : H7, Staphylococcus aureus and Listeria monocytogenes during the drying of meat: A case study using Biltong and dro??wors. J. Food Saf. 28, 198–209. doi:10.1111/j.1745-4565.2008.00114.x

Burt, S., 2004. Essential oils: Their antibacterial properties and potential applications in foods - A review. Int. J. Food Microbiol. doi:10.1016/j.ijfoodmicro.2004.03.022

Burt, S. a, Reinders, R.D., 2003. Antibacterial activity of selected plant essential oils against Escherichia coli O157:H7. Lett. Appl. Microbiol. 36, 162–7. doi:10.1046/j.1472-765X.2003.01285.x

Bussmann, R.W., Glenn, A., Sharon, D., 2010. Antibacterial activity of medicinal plants of Northern Peru - can traditional applications provide leads for modern science? Indian J Tradit. Knowl. 9, 742–753.

Cagri, A., Ustunol, Z., Osburn, W., Ryser, E.T., 2003. Inhibition of Listeria monocytogenes on hot dogs using antimicrobial whey protein-based edible casings. J. Food Sci. 68, 291–299. doi:10.1111/j.1365-2621.2003.tb14155.x

Cagri, A., Ustunol, Z., Ryser, E.T., 2001. Antimicrobial, Mechanical, and Moisture Barrier Properties of Low pH Whey Protein-based Edible Films Containing p-Aminobenzoic or Sorbic Acids. J. Food Sci. 66, 865–870. doi:10.1111/j.1365-2621.2001.tb15188.x

Cavanagh, H.M.A., 2007. Antifungal Activity of the Volatile Phase of Essential Oils: A Brief Review. Nat. Prod. Commun. 2, 1297–1302.

Cetin-Karaca, H., 2011. Evaluation of natural antimicrobial phenolic compounds against foodborne pathogens. University of Kentucky.

Cheng, L.-S., Fang, S., Ruan, M.-L., 2015. Influence of Blanching Pretreatment on the Drying Characteristics of Cherry Tomato and Mathematical Modeling. Int. J.

Food Eng. 11, 265–274.

Chorianopoulos, N., Kalpoutzakis, E., Aligiannis, N., Mitaku, S., Nychas, G.-J., Haroutounian, S. a, 2004. Essential oils of Satureja, Origanum, and Thymus species: chemical composition and antibacterial activities against foodborne pathogens. J. Agric. Food Chem. 52, 8261–8267. doi:10.1021/jf049113i

Conner, D.E., 1993. Naturally occurring compounds, in: Antimicrobials in Food. New York, pp. 441–468.

Dave, D., Ghaly, A.E., 2011. Meat spoilage mechanisms and preservation techniques: a critical review. Am. J. Agric. Biol. Sci. 6, 486–510.

Delaquis, P.J., Sholberg, P.L., 1997. Antimicrobial Activity of Gaseous Allyl Isothiocyanate. J. Food Prot. 60, 943–947.

Delgado, C., Rosegrant, M.W., Steinfeld, H., Ehui, S., Courbois, C., 2001. Livestock to 2020: the next food revolution. Outlook Agric. doi:10.5367/000000001101293427

Delmore Jr., R.J., Sofos, J.N., Schmidt, G.R., Belk, K.E., Lloyd, W.R., Smith, G.C., 2000. Interventions to reduce microbiological contamination of beef variety meats. J Food Prot 63, 44–50.

DiPersio, P.A., Yoon, Y., Sofos, J.N., Kendall, P.A., 2005. Inactivation of Salmonella during Drying and Storage of Carrot Slices Prepared Using Commonly Recommended Methods. J. Food Sci.

Dorantes, L.A., Jaramillo, E.F., González, K., Martinez, R., Parada, L., 2011. Blanching peppers using microwaves. Procedia Food Sci. 1, 178–183. doi:10.1016/j.profoo.2011.09.028

Eidson M, Sewell C.M., Graves G, O.R., 2000. Beef jerky gastroenteritis outbreaks. J. Environ. Health 62, 9–13.

Eklund, M.W., Peterson, M.E., Poysky, F.T., Paranjpye, R.N., Pelroy, G. a, 2004. Control of bacterial pathogens during processing of cold-smoked and dried salmon strips. J. Food Prot. 67, 347–351.

Epstein, S.S., 2009. Microbial awakenings. Nature 457, 1083. doi:10.1038/4571083a

Exarchou, V., Nenadis, N., Tsimidou, M., Gerothanassis, I.P., Troganis, A., Boskou, D., 2002. Antioxidant activities and phenolic composition of extracts from Greek oregano, Greek sage, and summer savory. J. Agric. Food Chem. 50, 5294–5299. doi:10.1021/jf020408a

Faith, N.G., Le Coutour, N.S., Alvarenga, M.B., Calicioglu, M., Buege, D.R., Luchansky, J.B., 1998. Viability of Escherichia coli O157: H7 in ground and formed beef jerky prepared at levels of 5 and 20\% fat and dried at 52, 57, 63, or 68 C in a home-style dehydrator. Int. J. Food Microbiol. 41, 213–221.

Friedman, M., Henika, P.R., Levin, C.E., Mandrell, R.E., 2007. Recipes for antimicrobial wine marinades against Bacillus cereus, Escherichia coli O157:H7, Listeria monocytogenes, and Salmonella enterica. J. Food Sci. 72. doi:10.1111/j.1750-3841.2007.00418.x

Geankoplis, C.J., 1993. Transport Processes and Unit Operations. Prentice-Hall Int.

Gonzalez-Martinez, G., Ahrne, L., Gekas, V., Sjoholm, I., 2004. Analysis of temperature distribution in potato tissue during blanching and its effect on the absolute residual pectin methylesterase activity. J. Food Eng. doi:10.1016/j.jfoodeng.2004.02.003

Govaris, A., Solomakos, N., Pexara, A., Chatzopoulou, P.S., 2010. The antimicrobial effect of oregano essential oil, nisin and their combination against Salmonella Enteritidis in minced sheep meat during refrigerated storage. Int. J. Food Microbiol. 137, 175–180. doi:10.1016/j.ijfoodmicro.2009.12.017

Greig, J.D., Ravel, A., 2009. Analysis of foodborne outbreak data reported internationally for source attribution. Int. J. Food Microbiol. 130, 77–87. doi:10.1016/j.ijfoodmicro.2008.12.031

Gutierrez, J., Rodriguez, G., Barry-Ryan, C., Bourke, P., 2008. Efficacy of plant essential oils against foodborne pathogens and spoilage bacteria associated with ready-to-eat vegetables: antimicrobial and sensory screening. J. Food Prot. 71, 1846–54.

Heinz, G., Hautzinger, P., 2007. Meat processing technology for small to medium scale producers, RAP Publication (FAO).

Helander, I.M., Alakomi, H.-L., Latva-Kala, K., Mattila-Sandholm, T., Pol, I., Smid, E.J., Gorris, L.G.M., von Wright, A., 1998. Characterization of the Action of Selected Essential Oil Components on Gram-Negative Bacteria. J. Agric. Food Chem. 46, 3590–3595. doi:10.1021/jf980154m

Hernández, H., Fraňková, A., Sýkora, T., Klouček, P., Kouřimská, L., Kučerová, I., Banout, J., 2016. The effect of oregano essential oil on microbial load and sensory atributes of dried meat. J. Sci. Food Agric. n/a–n/a. doi:10.1002/jsfa.7685

Hierro, E., De La Hoz, L., Ordóñez, J.A., 2004. Headspace volatile compounds from

salted and occasionally smoked dried meats (cecinas) as affected by animal species. Food Chem. 85, 649–657. doi:10.1016/j.foodchem.2003.07.001

Higgs, J. and Pratt, J., 2003. MEAT | Nutritional Value. Encycl. Food Sci. Nutr.

Hii, C.L., Jangam, S. V., Ong, S.P., Mujumdar, A.S., 2012. Solar drying: Fundamentals, applications and innovations. TPR Group Publication, Singapore.

Hoffman, L.C., Wiklund, E., 2006. Game and venison - meat for the modern consumer. Meat Sci. 74, 197–208. doi:10.1016/j.meatsci.2006.04.005

Holley, R.A., Patel, D., 2005. Improvement in shelf-life and safety of perishable foods by plant essential oils and smoke antimicrobials. Food Microbiol. doi:10.1016/j.fm.2004.08.006

Hospital, X.F., Hierro, E., Fern??ndez, M., 2014. Effect of reducing nitrate and nitrite added to dry fermented sausages on the survival of Salmonella Typhimurium. Food Res. Int. 62, 410–415. doi:10.1016/j.foodres.2014.03.055

Hulankova, R., Borilova, G., Steinhauserova, I., 2013. Combined antimicrobial effect of oregano essential oil and caprylic acid in minced beef. Meat Sci. 95, 190–194. doi:10.1016/j.meatsci.2013.05.003

İsmail, O., Kantürk Figen, A., Pişkin, S., 2015. Effects of open-air sun drying and pre-treatment on drying characteristics of purslane (Portulaca oleracea L.). Heat Mass Transf. 51, 807–813. doi:10.1007/s00231-014-1452-8

ISO, 2012. ISO 8586:2012. Sensory Analysis – General Guidelines for the Selection, Training and Monitoring of Selected Assessors and Expert Sensory Assessors.

J.R.L, W., 1994. Antimicrobial compounds in food plants, in: Natural Antimicrobial Systems and Food Preservation. Wallingford, UK, pp. 181–204D.

Juven, B.J., Kanner, J., Schved, F., Weisslowicz, H., 1994. Factors that interact with the antibacterial action of thyme essential oil and its active constituents. J. Appl. Bacteriol. 76, 626–631. doi:10.1111/j.1365-2672.1994.tb01661.x

Kabara, J.J., Eklund, T., 1991. Organic acids and esters. Food Preserv. 44–71.

Keene, W.E., Sazie, E., Kok, J., Rice, D.H., Hancock, D.D., Balan, V.K., Zhao, T., Doyle, M.P., 1997. An outbreak of Escherichia coli O157:H7 infections traced to jerky made from deer meat. JAMA 277, 1229–1231. doi:10.1001/jama.277.15.1229

Kendall, P., DiPersio, P., Sofos, J., 2012. Drying Vegetables. Food Nutr. Ser. 9.308.

Khamisse, E., Firmesse, O., Christieans, S., Chassaing, D., Carpentier, B., 2012. Impact of cleaning and disinfection on the non-culturable and culturable bacterial loads of food-contact surfaces at a beef processing plant. Int. J. Food Microbiol. 158, 163–168. doi:10.1016/j.ijfoodmicro.2012.07.014

Kim, S., Fung, D.Y.C., 2004. Antibacterial effect of water-soluble arrowroot (Puerariae radix) tea extracts on foodborne pathogens in ground beef and mushroom soup. J. Food Prot. 67, 1953–1956.

Kingsly, R.P., Goyal, R.K., Manikantan, M.R., Ilyas, S.M., 2007. Effects of pretreatments and drying air temperature on drying behaviour of peach slice. Int. J. Food Sci. Technol. 42, 65–69. doi:10.1111/j.1365-2621.2006.01210.x

Kloucek, P., Smid, J., Frankova, A., Kokoska, L., Valterova, I., Pavela, R., 2012. Fast screening method for assessment of antimicrobial activity of essential oils in vapor phase. Food Res. Int. 47, 161–165. doi:10.1016/j.foodres.2011.04.044

Kokkini, S., Karousou, R., Dardioti, A., Krigas, N., Lanaras, T., 1997. Autumn essential oils of Greek oregano. Phytochemistry 44, 883–886. doi:10.1016/S0031-9422(96)00576-6

Konieczny, P., Stangierski, J., Kijowski, J., 2007. Physical and chemical characteristics and acceptability of home style beef jerky. Meat Sci. 76, 253–257. doi:10.1016/j.meatsci.2006.11.006

Kuc, J., 1995. Phytoalexins, stress metabolism, and disease resistance in plants. Annu. Rev. Phytopathol. 33, 275–297. doi:10.1146/annurev.py.33.090195.001423

Kucerova, I., Hubackova, A., Rohlik, B.-A., Banout, J., 2015. Mathematical Modeling of Thin-Layer Solar Drying of Eland (Taurotragus oryx) Jerky. Int. J. Food Eng. doi:10.1515/ijfe-2014-0227

Lambert, R.J.W., Skandamis, P.N., Coote, P.J., Nychas, G.J.E., 2001. A study of the minimum inhibitory concentration and mode of action of oregano essential oil, thymol and carvacrol. J. Appl. Microbiol. 91, 453–462. doi:10.1046/j.1365-2672.2001.01428.x

Lazar, T., 2003. Food Microbiology: Fundamentals and Frontiers (2nd edn). Trends Food Sci. Technol. doi:10.1016/S0924-2244(03)00047-5

Levine, P., Rose, B., Green, S., Ransom, G., Hill, W., 2001. Pathogen testing of ready-to-eat meat and poultry products collected at federally inspected establishments

in the United States, 1990 to 1999. J. Food Prot. 64, 1188–93.

Mahmoud, B.S.M., Yamazaki, K., Miyashita, K., Kawai, Y., Shin, I.S., Suzuki, T., 2006. Preservative effect of combined treatment with electrolyzed NaCl solutions and essential oil compounds on carp fillets during convectional air-drying. Int. J. Food Microbiol. 106, 331–337. doi:10.1016/j.ijfoodmicro.2005.09.010

Mani-López, E., García, H.S., López-Malo, A., 2012. Organic acids as antimicrobials to control Salmonella in meat and poultry products. Food Res. Int. 45, 713–721. doi:10.1016/j.foodres.2011.04.043

Marino, M., Bersani, C., Comi, G., 2001. Impedance measurements to study the antimicrobial activity of essential oils from Lamiaceae and Compositae. Int. J. Food Microbiol. 67, 187–95. doi:10.1016/S0168-1605(01)00447-0

Miladi, H., Chaieb, K., Ammar, E., Bakhrouf, A., 2010. Inhibitory effect of clove oil (Syzium Aromaticum) against Listeria Monocytogenes cells incubated in fresh-cut salmon. J. Food Saf. 30, 432–442. doi:10.1111/j.1745-4565.2010.00217.x

Mujumdar, A.S., 2006. Book Review: Handbook of Industrial Drying, Third Edition, Drying Technology. doi:10.1080/07373930701399224

Mujumdar, A.S., Huang, L.X., 2007. Global R&D needs in drying. Dry. Technol. 25, 647–658.

Mukherjee, S., Chattopadhyay, P.K., 2007. Whirling bed blanching of potato cubes and its effects on product quality. J. Food Eng. 78, 52–60. doi:10.1016/j.jfoodeng.2005.09.001

Nathakaranakule, A., Kraiwanichkul, W., Soponronnarit, S., 2007. Comparative study of different combined superheated-steam drying techniques for chicken meat. J. Food Eng. 80, 1023–1030. doi:10.1016/j.jfoodeng.2006.04.067

Nazzaro, F., Caliendo, G., Arnesi, G., Veronesi, A., Sarzi, P., Fratianni, F., 2009. Comparative content of some bioactive compounds in two varieties of capsicum annuum l. sweet pepper and evaluation of their antimicrobial and mutagenic activities. J. Food Biochem. 33, 852–868. doi:10.1111/j.1745-4514.2009.00259.x

Nedorostova, L., Kloucek, P., Kokoska, L., Stolcova, M., Pulkrabek, J., 2009. Antimicrobial properties of selected essential oils in vapour phase against foodborne bacteria. Food Control 20, 157–160. doi:10.1016/j.foodcont.2008.03.007

Nummer, B. a, Harrison, J. a, Harrison, M. a, Kendall, P., Sofos, J.N., Andress, E.L.,

2004. Effects of Preparation Methods on the Microbiological Safety of Home-Dried Meat Jerky. J. Food Prot. 67, 2337–2341.

Nyachuba, D.G., Donnelly, C.W., Howard, A.B., 2007. Impact of nitrite on detection of Listeria monocytogenes in selected ready-to-eat (RTE) meat and seafood products. J. Food Sci. 72. doi:10.1111/j.1750-3841.2007.00455.x

Nychas, G., Skandamis, P., Tassou, C., Roller, S., 2003. Antimicrobials from herbs and spices. Nat. Antimicrob. minimal Process. foods.

Ohlsson, T., Bengtsson, N., 2002. Minimal Processing Technologies in the Food Industry, Food Manufacture. doi:10.1201/9781439823132

Oliver, J.D., 2010. Recent findings on the viable but nonculturable state in pathogenic bacteria. FEMS Microbiol. Rev. doi:10.1111/j.1574-6976.2009.00200.x

Oliver, J.D., 2005. The viable but nonculturable state in bacteria. J. Microbiol. 43 Spec No, 93–100. doi:2134 [pii]

Pangavhane, D.R., Sawhney, R.L., Sarsavadia, P.N., 1999. Effect of various dipping pretreatment on drying kinetics of Thompson seedless grapes. J. Food Eng. 39, 211–216. doi:10.1016/S0260-8774(98)00168-X

Patra, J.K., Thatoi, H.N., 2011. Metabolic diversity and bioactivity screening of mangrove plants: A review. Acta Physiol. Plant. doi:10.1007/s11738-010-0667-7

Peñalver, P., Huerta, B., Borge, C., Astorga, R., Romero, R., Perea, A., 2005. Antimicrobial activity of five essential oils against origin strains of the Enterobacteriaceae family. APMIS 113, 1–6. doi:10.1111/j.1600-0463.2005.apm1130101.x

Perumalla, A.V.S., Hettiarachchy, N.S., 2011. Green tea and grape seed extracts - Potential applications in food safety and quality. Food Res. Int. doi:10.1016/j.foodres.2011.01.022

Rabsch, W., Tschäpe, H., Bäumler, A., 2001. Review non-typhoidal salmonellosis: emerging problems. Microbes Infect. 3, 237–247.

Radhika, G., Satyanarayana, S., Rao, D., 2011. Mathematical model on thin layer drying of finger millet (Eluesine coracana). Adv. J. Food Sci. Technol.

Rahman, M.S., Guizani, N., Al-Ruzeiki, M.H., Khalasi, A.S. Al, 2000. Microflora Changes in Tuna Mince During Convection Air Drying. Dry. Technol. 18, 2369–2379. doi:10.1080/07373930008917847

Rai, M., Chikindas, M., 2011. Natural Antimicrobials in Food Safety and Quality, Natural Antimicrobials in Food Safety and Quality. doi:10.1079/9781845937690.0000

Raman, B.V., Ramkishore, A.S., Uma Maheswari, M., Radhakrishnan, T.M., 2009. Antibacterial activities of some folk medicinal plants of Eastern Ghats. J. Pure Appl. Microbiol. 3, 187–194.

Ramesh, M.N., Wolf, W., Tevini, D., Bogna?r, a, 2002. Microwave blanching of vegetables. J. Food Sci. 67, 390–398.

Resurreccion, A.V.A., 2004. Sensory aspects of consumer choices for meat and meat products. Meat Sci. 66, 11–20. doi:10.1016/S0309-1740(03)00021-4

Ricke, S., 2003. Perspectives on the use of organic acids and short chain fatty acids as antimicrobials. Poult. Sci. 82, 632–639. doi:10.1093/ps/82.4.632

Rico-Munoz, E., Bargiota, E.E., Davidson, P.M., 1987. Effect of selected phenolic compounds on the membrane-bound adenosine triphosphatase of Staphylococcus aureus. Food Microbiol. 4, 239–249. doi:10.1016/0740-0020(87)90006-2

RJ, E., PB, A., Warthesen JJ, 1992. Nitrite in Meat [WWW Document]. Univ. Minnesota Ext. Serv.

Roberts, D., Greenwood, M., 2003. Practical Food Microbiology, Blackwell. doi:10.1002/9780470757512

Ruiz-Ojeda, L.M., Peñas, F.J., 2013. Comparison study of conventional hot-water and microwave blanching on quality of green beans. Innov. Food Sci. Emerg. Technol. 20, 191–197. doi:10.1016/j.ifset.2013.09.009

Sagar, V.R., Suresh Kumar, P., 2010. Recent advances in drying and dehydration of fruits and vegetables: A review. J. Food Sci. Technol. doi:10.1007/s13197-010-0010-8

Serra, A.T., Matias, A.A., Nunes, A.V.M., Leit??o, M.C., Brito, D., Bronze, R., Silva, S., Pires, A., Crespo, M.T., San Rom??o, M. V., Duarte, C.M., 2008. In vitro evaluation of olive- and grape-based natural extracts as potential preservatives for food. Innov. Food Sci. Emerg. Technol. 9, 311–319. doi:10.1016/j.ifset.2007.07.011

Shivhare, U.S., Gupta, M., Basu, S., Raghavan, G.S. V, 2009. Optimization of blanching process for carrots. J. Food Process Eng. 32, 587–605.

doi:10.1111/j.1745-4530.2007.00234.x

Sivropoulou, A., Papanikolaou, E., Nikolaou, C., Kokkini, S., Lanaras, T., Arsenakis, M., 1996. Antimicrobial and Cytotoxic Activities of Origanum Essential Oils. J. Agric. Food Chem. 44, 1202–1205. doi:10.1021/jf950540t

Skandamis, P.N., Nychas, G.J.E., 2002. Preservation of fresh meat with active and modified atmosphere packaging conditions, in: International Journal of Food Microbiology. pp. 35–45. doi:10.1016/S0168-1605(02)00177-0

Skandamis, P.N., Nychas, G.J.E., 2001. Effect of oregano essential oil on microbiological and physico-chemical attributes of minced meat stored in air and modified atmospheres. J. Appl. Microbiol. 91, 1011–1022. doi:10.1046/j.1365-2672.2001.01467.x

Smid, E.J., Gorris, L.G., 1999. Natural antimicrobials for food preservation, in: Food Science and Technology. Marcel Dekker, New York, pp. 285–308.

Smith-Palmer, A., Stewart, J., Fyfe, L., 1998. Antimicrobial properties of plant essential oils and essences against five important food-borne pathogens. Lett. Appl. Microbiol. 26, 118–122. doi:10.1046/j.1472-765X.1998.00303.x

Sotome, I., Takenaka, M., Koseki, S., Ogasawara, Y., Nadachi, Y., Okadome, H., Isobe, S., 2009. Blanching of potato with superheated steam and hot water spray. LWT - Food Sci. Technol. 42, 1035–1040. doi:10.1016/j.lwt.2009.02.001

Suomalainen, T.H., Mäyrä-Makinen, A.M., 1999. Propionic acid bacteria as protective cultures in fermented milks and breads. Lait 79, 165–174. doi:10.1051/lait:1999113

Tajkarimi, M.M., Ibrahim, S.A., Cliver, D.O., 2010. Antimicrobial herb and spice compounds in food. Food Control. doi:10.1016/j.foodcont.2010.02.003

The Nielsen Company, 2014. Nielsen Global Survey of Snacking.

Tiwari, B.K., Valdramidis, V.P., O'Donnell, C.P., Muthukumarappan, K., Bourke, P., Cullen, P.J., 2009. Application of natural antimicrobials for food preservation. J. Agric. Food Chem. doi:10.1021/jf900668n

Tsigarida, E., Skandamis, P., Nychas, G.-J., 2000. Behaviour of Listeria monocytogenes and autochthonous flora on meat stored under aerobic, vacuum and modified atmosphere packaging conditions with or without the presence of oregano essential oil at 5 °C. J. Appl. Microbiol. 89, 901–909.

Tunde-Akintunde, T.Y., Afolabi, T.J., Akintunde, B.O., 2005. Influence of drying

methods on drying of bell-pepper (Capsicum annuum). J. Food Eng. 68, 439–442. doi:10.1016/j.jfoodeng.2004.06.021

Yamazaki, K., Yamamoto, T., Kawai, Y., Inoue, N., 2004. Enhancement of antilisterial activity of essential oil constituents by nisin and diglycerol fatty acid ester. Food Microbiol. 21, 283–289. doi:10.1016/j.fm.2003.08.009

BIBLIOGRAPHY

Research papers

Hernández, H., Fraňková, A., Sýkora, T., Klouček, P., Kouřimská, L., Kučerová, I., Banout, J., 2016. The effect of oregano essential oil on microbial load and sensory atributes of dried meat. J. Sci. Food Agric. n/a–n/a. doi:10.1002/jsfa.7685

Hernández,H., Claramount, D., Kučerová,I. and Banout,J., 2016. The effect of modified blanching and oregano essential oil on drying kinetics and sensory attributes of dried meat. J. J. Food Process. Preserv. Status: In Press.

Poster in International Conference

Using of Natural Preservatives for Meat Drying and Storage. Tropentag 2014. http://www.tropentag.de/2014/proceedings/node375.html#9192

ANNEX A – FORM FOR MICROBIAL LOAD SENSORY

ANALYSIS

Sensory profile of dried meat

Name:..... Surname: ..... sample no.: ……

Health: ..... Date and hour: ……

Please, mark with the cross the answer:

The oregano smell: I like ☐ I do not like ☐ I do not mind ☐

Task: Please taste the given meat sample and focus on the evaluation of its taste smell and consistency. For evaluation use the graphical scale.

APPEARANCE

GENERAL APPEARANCE: _____
very bad excellent

SMELL

GENERAL PLEASANTNESS OF THE SMELL: _____
very bad excellent

GENERAL INTENSITY OF THE SMELL: _____
extremely bland extremely intense

INTENSITY OF THE OREGANO SMELL: _____
extremely bland extremely intense

TEXTURE (after 10 chews)

GENERAL PLEASANTNESS OF THE TEXTURE: _____
very bad excellent

JUICINESS: _____
dry juicy

CHEWINESS: _____
easy difficult

TASTE

GENERAL PLEASANTNESS OF THE TASTE: _____
very bad excellent

GENERAL INTENSITY OF THE TASTE:

extremely bland — extremely intense

INTENSITY OF PARTIAL TASTES

OREGANO TASTE:

extremely bland — extremely intense

SALTY:

extremely bland — extremely intense

BITTER:

extremely bland — extremely intense

ADSTRINGENS:

extremely bland — extremely intense

PUNGENT:

extremely bland — extremely intense

OTHER (SPECIFY): (_____)

extremely bland — extremely intense

OVERAL EVALUATION OF THE SAMPLE:

very bad — **excellent**

NOTES:

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ANNEX B – FORM FOR MODIFIED BLANCHING

SENSORY ANALYSIS

Sensory profile of dried meat

Name:..... Surname: ..... sample no.: ……

Health: ..... Date and hour: ……

Task: Please taste the given meat sample and focus on the evaluation of its appearance, smell, color, taste and consistency. For evaluation use the graphical scale.

APPEARANCE

GENERAL APPEARANCE: _____
very bad excellent

SMELL

GENERAL PLEASANTNESS OF THE SMELL: _____
very bad excellent

INTENSITY OF THE OREGANO SMELL: _____
extremely bland extremely intense

COLOR

GENERAL LIKABLENESS OF THE COLOR: _____
like dislike

GENERAL INTENSITY OF THE COLOR: _____
extremely light extremely dark

TEXTURE (after 10 chews)

GENERAL PLEASANTNESS OF THE TEXTURE: _____
very bad excellent

JUICINESS: _____
dry juicy

CHEWINESS: _____
difficult easy

TASTE

GENERAL PLEASANTNESS OF THE TASTE: very bad — excellent

GENERAL INTENSITY OF THE TASTE: extremely bland — extremely intense

INTENSITY OF PARTIAL TASTES

OREGANO TASTE: extremely bland — extremely intense

SALTY: extremely bland — extremely intense

BITTER (HOŘKÁ): extremely bland — extremely intense

ASTRINGENS (TRPKÁ) : extremely bland — extremely intense

PUNGENT (ŠTIPLAVÝ): extremely bland — extremely intense

OTHER (SPECIFY): (_____) extremely bland — extremely intense

OVERALL EVALUATION OF THE SAMPLE:

very bad — **excellent**

NOTES:

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