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To

Deolinda and Sacama's family.

"*Que a flecha do amor perfume bem no fundo do seu coração para que tudo tocar de hoje em diante mesmo azedo se torne doce…*"

Deolinda, 2009

Indice

Resume

In Mozambique, about 50 % of the tomato produce are wasted due to negligence, defective marketing system and lack of processing facilities. During the peak season tomato is sold at low prices due to larger supplies resulting in less return to growers. In addition, the demand for processed vegetable and fruit products is increasing progressively. Therefore, there is a potential market for processed tomatoes. Mozambique has got three agro ecology zones as Chokwe, Sussundenga, Nampula and Niassa district were produce tomato fruits, with production rate 63,6; 10,8; 7,2 and 18,4 %, respectively.

Foods processing as fruits and vegetable are one of prevention method from getting spoilt for a long period of storage. The color, taste and nutritive value of the food are also preserved.

Tomato (*Lycopersicon esculentum* Mill.) is considered as prize vegetable throughout the Worlds and extensively commercialized in fresh condition as well as for the preparation of various food products. It could be heat processing in order to get tomato products as tomato pulp, paste, juice, ketchup, sauce and puree.

The main objective of this work is to study the effect of heat-treatment on tomato quality (*Roma* variety) and measure rheological and chemical parameters. Tomato fruits were processed according reference methods.

First tomato fruits were washed with water contains sodium hypochlorite solution (65 ppm of active chlorine) and cleaned with potable water. It was color and physical state sorted, and then triturated in machine. The hot-break processing was done in range of temperature and time of inactivation of enzymes methylesterase and polygalacturonase. The juice is concentrated in two systems (vacuum evaporator and atmospheric evaporator), when the puree samples (15 % Brix) was already, it was pasteurized in water bath and water spray retort.

Quality parameters analysis was conducted according Reference Methods of Analysis (AOAC 2000 and UNE Standards) and the parameters were:

- Color,

- Lycopene concentration,
- Ascorbic acid concentration,
- Consistency,
- Brix degree,
- pH,
- Acidity.

In each step of process samples were taken to analysis and then stored in environment condition during 24 hours.

Data from this study concluded that processing of tomatoes in defined condition as pre-heating tomatoes fruits before triturating; heating tomato juice at 80,0 ºC, 16 minutes of hot-break; concentrate in vacuum evaporator, and pasteurize were optimal conditions to get tomato puree without considerable loss of components.

I. INTRODUCTION

1.1. Fruits and vegetables as natural antioxidants sources

Diet rich in fruits and vegetables are good for health and balances free radicals reaction. Most diseases can be reduced by consuming foodstuff rich in vitamins, phenolic compounds and minerals as fruits and vegetables (Metchell *et al*, 2012, Hall *et al*., 2010).

The tomato is native to the Andean of South America. In sixteenth century, the tomato was taken to Europe by the Spanish conquistadors and later introduced from Europe to southern eastern Asia, Africa and the Middle East. Tomato is annually plant, which can reach a height of over two meters. However, the some plants can be harvested for several years in succession. The first harvest is possible 45-55 days after flowering, or 90-120 days after sowing. The shape of the fruit differs per cultivar. The color range from yellow to red (Van-Dam *et al.*, 2005).

In example, "Tomato was produced at garden as ornament plant because British conquistadors identify it as superstition fruit. However, scurvy disease was critical in citizen". Cases of vitamin C deficiency disease (scurvy) are well documented throughout history. Although it is probable that many have suffered from the disease for centuries while on land, scurvy is most commonly associated with the extended sea travels in the 16th, 17th, and 18th centuries (Covey, 2012, Schlueter *et al*., 2011). This potentially fatal disease can be prevented with as little as 10 mg vitamin C/day (Rahman *et al*., 2006). William D. Clay said: "Eat fruits and vegetables at meals are good for balance nutritional necessity of body". (FAO, 2003)

1.2. Cultivate of Tomato in Mozambique

According the report of Agronomy Research Institute of Mozambique (Mazuze *et al*, 2006), describe four main agroecologies zone where tomato fruits are produced (Fig. 1). More than 63,6 % of tomatoes are from Chokwe district. The tomato is a part of commodity cultivated in Mozambique. The percentage of tomato produced in agro ecologies zones are:

- R_1 South (Chókwè) – 63,6 %;
- R_2 Center (Sussundenga) -10,8 %;
- R_3 North 1 (Nampula) – 7,2%;
- R_4 North 2 (Lichinga) – 18,4 %.

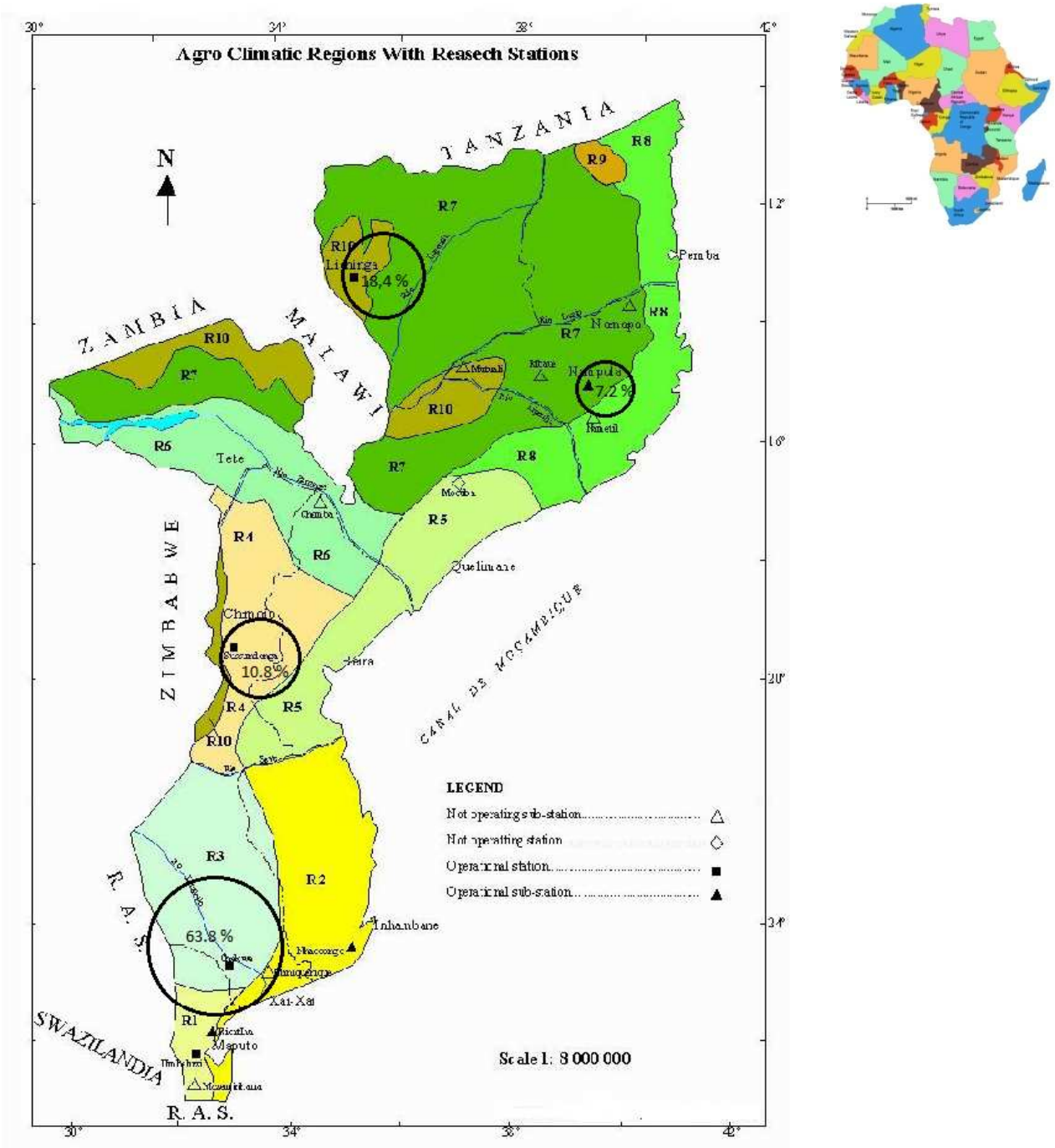


Figure 1. Agroecologies zones that cultivate tomato in Mozambique. Source: IIAM

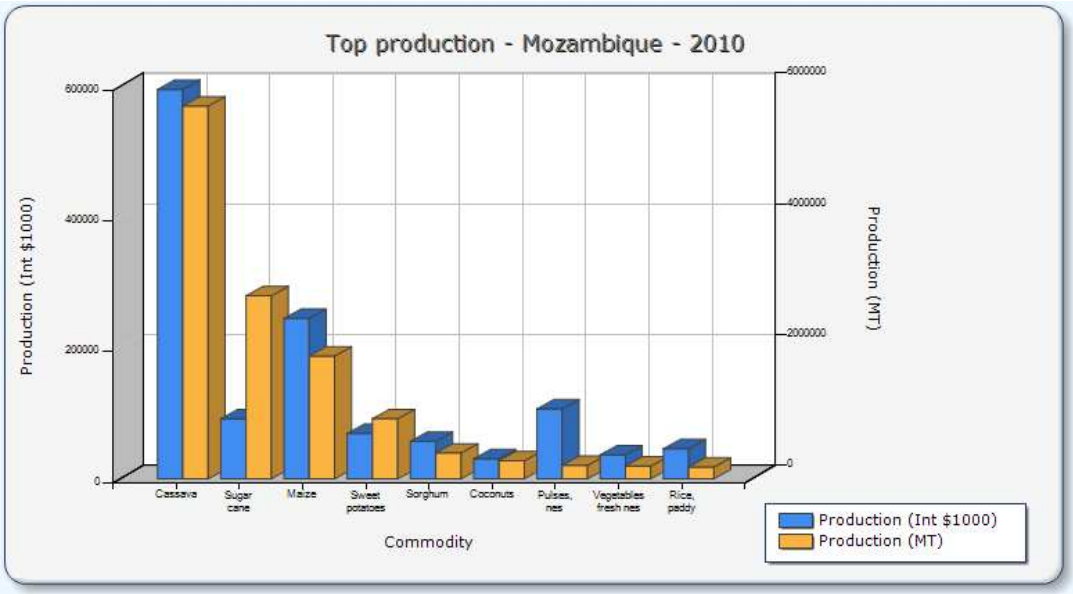


Figure 2. Mozambique commodity production. Source: FAOSTAT 2010.

1.3. Price of Tomato in Mozambique

According with FAOSTAT 2010 (Fig. 2), Mozambique produced 195000 tons of vegetables, and in 2011 tomato production were 93000 tons (PEDSA 2011-2020). In Mozambique, about 50 % of the tomato fruits are wasted due to negligence, defective marketing system and lack of processing facilities. During the peak season tomato is sold at low prices 0,25 to 0,33 €/kg, due to larger supplies resulting in less return to growers. In addition, the demand season the price varies 0,65 to 0,78 €/kg Fig. 3). Processed vegetable and fruit products are increasing progressively; therefore, there is a potential market for processed tomatoes.

Figure 3. Tomato harvesting and marketing in Mozambique. Source: PEDSA

When internal production falls down, tomato is imported from South Africa and Zimbabwe. For example, Maputo capital-city consumes 40 tons of tomato per day, and nowadays the national production is around 60 % of necessity. In rain season the production down to 25 or 30 %, (PEDSA 2011-2020).

1.4. Processing and Nutritional Importance of Tomato

Tomato (*Lycopersicon esculentum* Mill.) is important vegetable worldwide, both for the fresh and the processing products. It is available all year round and is rich in compounds including vitamin C, flavonoids, carotenoids and phenolic compounds and minerals (Metchell *et al*., 2012). Tomato puree is an important product and the consumption of tomato fruits and tomato products such as tomato juice, sauces, purees, paste and canned tomatoes is associated with a lower risk of cardiovascular and cancer diseases (Lavelli *et al*., 2011).

1.4.1. Tomatoes Harvest

The commercially cultivated forms of tomato belong to the variety *Lycopersicon esculentum,* Mill. Nevertheless, botanically the tomato is a *fruit* of the genus *Lycopersicon*. Most of the world's tomatoes, including processing tomatoes (cultivars used for canning, freezing, drying, etc.) are grown under non limiting light conditions for most of the year (Davies and Hobson, 1981).

The quality of raw tomatoes can be maintained or decreased depending on the harvesting methods used the handling of the raw product during harvesting, and the holding methods used. Destruction of quality can be ascertained in several ways: cracking; drosophila fly egg contamination; or the numbers of vegetative bacteria, spores, yeasts, and moulds. Tomatoes are transported from the field to the processing plant in hampers, lug boxes, plastic boxes, or bulk containers.

The attributes of fruit quality include not only the flavor, color, nutritional content and firmness, but also shelf life, processing qualities and resistance to preharvest and postharvest pathogens. Tomato fruit has a rather short post-harvest life. A large annual

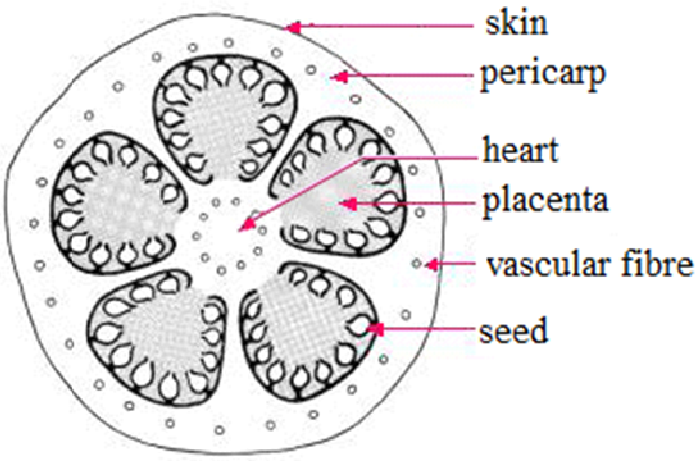
loss due to spoilage makes the ripening control a great economic importance. Although ripening makes fruit edible and tasty, it also initiates the gradual deterioration of fruit quality, especially in climacteric fruits such as tomato, in which the onset of ripening is considered to be initiated by endogenous ethylene (Ying *et al*., 2007).

During fruit ripening, the most apparent cell-wall change is an increase in water-soluble polyuronide. The solubilization of polyuronide is generally attributed to the action of polygalacturonase, which appears in many fruits near the onset of ripening. Another change that occurs, in tomatoes, is a pronounced loss of galactose from the cell walls. Tomato color development during ripening and postharvest storage is influenced by many factors, as temperature and storage duration. Tomato fruit ripening, as indicated by red color (lycopene) (Pressey *et al*., 1983).

1.4.2. Tomato Shape and Anatomy

The shape of an individual tomato fruit depend on many factors as environmental and nutritional conditions. Tomato shape varies greatly, depending on the cultivars; commonly fruits are elongated or pearlike, oblate, or spherical. Typically, processing tomatoes are pear shaped; it is common to find globular and oblong fruits. The tomato is a fleshy fruit, specifically, a berry. It is composed by skin tissue and seeds (as the placenta and the pericarp). Pericarp includes the skin, peripheral pericarp, radial arms, and columella. Tomato “skin” or peel is composed of a thin layer of heavily cutinized epidermal cells and two or more layers of relatively small, flattened cells as shown at Figure 4 (Wayne *et al*., 1998).

a)



b)

Figure 4. Tomato composition a) and b)

1.4.3. Tomato components

The tomato fruit contain approximately 5 % of total solids and 93,41 % of water. The residues contain valuable nutritional compounds, mainly fibres (59 % of dry weight), proteins (19 % of dry weiht) and antioxidants, an important source of vitamins and minerals. In particular, tomatoes are rich in vitamins A, C, E and folate (vitamin B), potassium and iron. (Lavelli *et al*., 2011, Maroulis and Saravacos, 2003). They are also Additional compounds of interest from a health standpoint include phytonutrients such as lycopene, flavonoids, phytosterols and various trace elements. In general tomato components are given in average per 100 g of fresh tomato (Table 1).

Table 1. Components contained in tomato per 100 g of tomato.

Components	Quantity
Water	93,41 g
Energy	23,3 kcal
Protein	0,875 g
carbohydrate	3,5 g
Fiber	1,4 g
Total Wax	0,21 g
Calcium	10,6 mg
Iron	0,7 mg
Iodum	2,2 μg
Magnesium	8,3 mg
Zinc	0,16 mg
Sodium	9,0 mg
Selenium	0,985 μg
Potassium	242,0 mg
Phosphor	24,0 mg
Vitamin B_1 (Thiamine)	0,07 mg
Vitamin B_2 (Riboflavin)	0,04
Niacin	0,9 mg
Vitamin B_6 (Pyridoxine)	0,13 mg
Folic Acid	28,8 μg
Vitamin C (Ascorbic Acid)	26,6 mg
Carotenoid (β-carotene)	1302 μg
Vitamin A (Retinol)	217,0 μg
Vitamin E (Tocopherol)	0,89 μg

Font: http://alimentos.gratis.es/tomate/

In recent decades, the consumption of tomatoes has been associated with the prevention of several diseases mainly due to the content of antioxidants, including carotenes (lycopene as well as β-carotene), ascorbic acid, tocopherol, and phenolic compounds (Kamil *et al*., 2011).

The nutritional value of food products has become one of the top priorities for the food industry. With increased consumed demands for nutritious, safe and chemical free, high quality juice food processors are looking for various alternatives to conventional thermal preservation techniques. However, the nutritional quality of tomato juice is primarily related to the ascorbic acid content and presence of bioactive compounds lycopene is responsible for tomato juice color (Cullen *et al*, 2009, Esehaghbeygi, 2001, Kamil, 2011). Organic acids which influence its shelf-life and organoleptic properties such as color brightness and texture contribute to acid-base balance for the consumer (Safdar *et al*., 2010, Fernandes *et al*., 2010). Tomato fruit quality has been assessed by the content of chemical compounds such as Brix degree, acidity, single sugars, citric and other organic acids and volatile compounds (Romero *et al*., 2007).

In human nutrition, tomato fruits and tomato products are the most important sources of lycopene. Apart from lycopene, the presence and synergy of a multitude of nutrients found in tomatoes may have contributed to these benefits. Besides microbial safety (Stoforos *et al*, 2010), important quality aspects of tomato products are color, flavor, and consistency.

Several different parameters can be evaluated to determine quality loss. Degradation of ascorbic acid (AA) is an indicator of the severity of processing or duration and temperature of storage as AA is susceptible to thermal degradation (Barringer *et al*., 2001). Color, consistency and pH are important quality factors that are readily apparent to the consumer.

One of the important reasons for preserving foods is to take care of the excess produce. The second reason for preserving foods is that they add variety to our meals; Reaches areas where the food item is not grown and makes transportation and storage of foods easier preservation of foods usually reduces bulk. This makes their transportation and storage easier since it requires less space (Fernandes *et al*, 2010).

In tomato products, an important reaction during thermal processing is the degradation of the red pigment lycopene, originally in the *trans* form, due to isomerization to the *cis* form resulting in color changes. Moreover, in tomato juice products stabilized by thermal processing, the changes of color and flavor can also be caused by non enzymatic browning. Lycopene, the compound responsible for the red

color of tomato and representing approximately 80 % to 90 % of its total carotenoid content (Davies and Hobson 1981, Ishida *et al*, 2007).

During storage, changes occur in tomato puree, which affect the consistency after reconstitution. Concentrated puree is typically stored for 1 year or more, and this stable material is diluted for production of sauces, salsas and other value-added products. Many variations in the quality of the puree can be obtained depending on factors such as the cultivar of tomatoes used, the finisher screen size and the break temperature.

Consistency of tomato products (viscosity) is the ability of their solid portion to remain in suspension throughout the shelf life of the product. The consistency of tomato products is strongly affected by the composition of the pectins.

Pectin is a natural constituent of ripe tomato. It is formed between the microscopic cells which make up the fleshy red tissues, cementing these together. Pectins are polymers of α-δ- galacturonic acid linked 1-4. Like the majority of polysaccharides, pectins vary in chain length and hence molecular weight. They are also esterified to varying degrees with methyl groups. The low-ester pectins, those in which fewer than 50 % of the groups are esterified, are known as pectic acids; the higher-ester pectins are called pectinic acids.

Controlling the breakdown or retention of the pectins, and the enzymes that lead to changes in the pectins, is thus of great importance during processing.

Pectin methylesterase (PME) (Fig. 5a) and polygalacturonase (PG) (Fig. 5b) are involved in the breakdown organic structure of tomato and chance rheology properties. Polygalacturonase can further break down the long pectin chains into shorter ones. A third enzyme, pectin or pectinesterase, can move the methyl ester groups from the molecule, thus transforming pectinic to pectic acids. The tomato fruit is particularly rich in these enzymes. These enzymes can be classified more minutely by their actions:

- Endopolygalactunorase;
- Exopolygalacturonase;
- Endopolymethylgalacturonase, and
- Exopolymethylgalacturonase.

Another enzyme that can also act on tomato solids is cellulose which may be important in affecting texture.

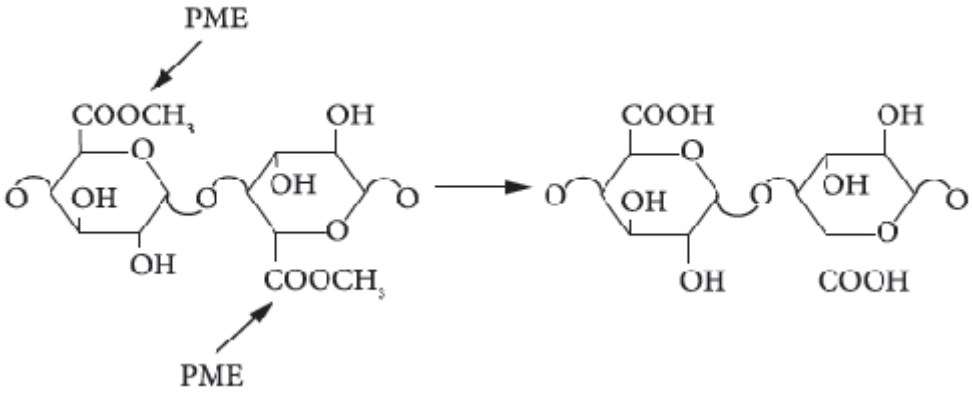


Figure 5a. PME act in the pectic substances. Source: Rosso *et al*., (2011).

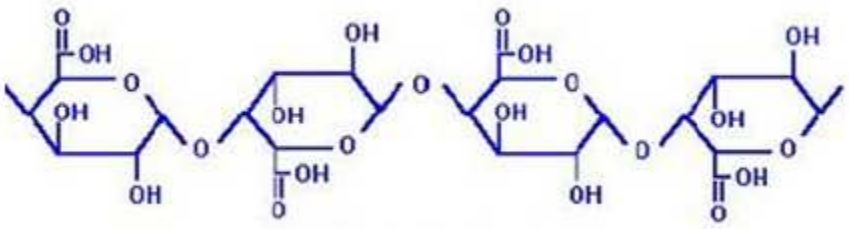


Figure 5b. PG pectin molecular structure.

The rheological properties of fluid tomato products like sauces and ketchup are important quality parameters. The flow properties of the whole juice, referred to as the gross viscosity or the consistency, are typical evaluated using a Bostwick consistometer. In this measurement, the distance the juice flows in a trough in 30 s is measured. More viscous juices flows shorter distances; thus, a higher Bostwick value indicates a lower consistency. Other devices such as efflux pippetes and rheometers have also been used to measure the apparent viscosity of whole juice. Concentrating tomato juice to puree during the tomato season allows for preservation and long-term storage, but subsequent dilution for formulation of value-added products is known to result in a loss of consistency. During tomato puree production, Bostwick values for each batch of tomato puree are routinely determined on the day the puree is produced (Anthon *et al*., 2010).

The effect of heating on consistency is due to a softening of the tomato juice prior to finishing in addition to the preservation of the pectin in the serum. Microscopic study indicated consistency to be due primarily to the number and shape of suspended particles. Hand and co-workers have pointed out that the consistency of whole tomato juice (gross viscosity) depends upon the viscosity of the serum (tomato juice free of suspended particles) and upon the viscous character of the suspended particles.

Consistency measurements were made on the whole tomato juice samples, since tomato juice is a non-Newtonian liquid (Gallegos *et al.*, 2003). Wherein stress is not proportional to rate of shear, all single gross viscosity measurement is actually "apparent viscosity" values as presented in this paper. Depending upon the temperature reached and rate of heating, heat treatment prior to finishing may either stimulate or destroy pectic enzymes.

Two different processes are commonly used in the production of tomato products. In the hot-break process, tomatoes are rapidly heated to 70,0–90,0 ^{o}C immediately after homogenization. This process is believed to inactivate enzymes rapidly, particularly those involved in pectin degradation, and give a product with high consistency. In the cold-break process, the homogenized tomatoes are heated only to around 60,0 ^{o}C. This is believed to have benefits in the production of products such as juice. The lower temperature reduces the amount of thermal abuse of the product, giving a greater retention of color and flavor components and reducing production of undesirable compounds. The lower temperature also does not entirely inactivate the enzymes PME and PG and allows these enzymes to break down of some of the pectins reducing the viscosity of the juice (Hsu *et al*., 2008).

Effects of heat processing on the tomato puree quality had been widely investigated. The volatile components and vitamin C of canned tomato juice can be reduced by heat-treatments. The color of tomato juice degraded more rapidly with increasing temperature. On the other hand, heat-treatments would increase the overall antioxidant potential of the tomato juice coincide with the positive effect of the temperature on the extractability of lycopene. Besides, lycopene in tomato is relative resistant to thermal degradation, whereas antioxidants such as (ascorbic acid, tocopherol and b-carotene) degrade more rapidly by thermal processing.

1.4.5. Microorganisms in Tomato Products

Generally, microorganisms are associative with nutrients groups in food. The microorganisms destruction or survive depends of heat treatment submitted or post contamination of canning food. Tomato products are acidic food with pH = 3,7 - 4,5. Characteristics microorganisms in that range are spores and no spores' bacteria's, yeasts and moulds.

Yeasts and moulds as *Byssochlamys fulva* is pectinic matters decomposition responsible and optimal multiply temperature is 30,0 – 37,0 ^{o}C. Others species of moulds in canned vegetables are *Byssochlamys nívea*, *Penicillium*, *Aspergillus*, *Rhizopus nigricans* and *Rhizopus stolonifer*.

The mechanisms of thermal destruction of microorganisms consist in proteins and enzymes metabolism inactivation. Pasteurization process is submitting canned food at temperature around 100,0 ^{o}C in atmospheric pressure or water spray retort. This process is applied for acidic and more acidic foods.

1.4.6. Color of Tomato

The color of tomato fruit measurement is frequently used in the tomato industry to predict the color of finished tomato products, which is an important quality index and determines the maturity and tomato post harvest life. Color or pigment changes during tomato ripening are characterized by a decrease in chlorophyll and a rapid accumulation of carotenoids, particularly lycopene which is the predominant pigment of carotenoid family in tomato and imparts the attractive redness (Camara *et al*., 2010).

When the color is measured at colorimeter instrument provide an objective, nondestructive and rapid technique that enables to obtain a series of parameters in a few seconds, and it is a useful tool for food quality control. Fruit chromaticity can be evaluated by the color space coordinates which are presented in alphanumeric form as three parameters: L*, a measure of lightness on a scale of 0 (black) to 100 (white); a*, which denotes greenness when negative and redness when positive; and b*, denoting

blueness when negative end yellowness when positive. Therefore, general parameter to characterize color quality in tomato is L* and ratio a*/b*.

1.4.7. Lycopene in Tomato

Lycopene is a hydrocarbon carotenoid, $C_{40}H_{56}$, with molecular weight of 537. In nature, it is most abundantly found as the red pigment of tomato. It present in one of two isomeric forms, *cis* and *trans* isomers. In the fresh tomato, 95,4% of the lycopene is in the *trans* isomer configuration. Processing often induces isomerization of carotenoids to the *cis* isomer. This is true for ß-carotene, and the *cis* isomer of that compound is not as biologically active. In the case of lycopene, there does not appear to be as much processing-induced isomerization. Indeed, Nguyen and Schwartz (1998) found lycopene to be relatively heat resistant, and found less than 10 % *cis* isomer in the processed tomato products analyzed. Lycopene have got antioxidant property which enable neutralize free radical formed in human body.

One form of the oxidative degradation of carotenoids is loss of color during processing and storage of foods. The extended conjugated double bond system of lycopene compounds is an important feature in the carotenoids responsible for their attractive colors because it forms the light absorbing chromophore. The existence of visible color in these compounds requires at least seven conjugated double bonds. The greater the number of conjugated double bonds, the higher a wavelength value for maximum absorption is observed (Ismail *et al*., 2010).

Lycopene is an unsaturated acyclic carotenoid with 11 linear conjugated and two non-conjugated double bonds (Fig. 6). Lycopene occurs naturally as all *trans* form and its chain containing seven double bonds that can be isomerized to mono-*cis* or poly-*cis* due to the exposure to high temperatures, light, oxygen, acids, catalyst and metal ions. Lycopene is a lipophilic compound with hydrophobic characteristics due to its acyclic structure and 11 linear conjugated double bonds that make it more soluble in organic solvents such as chloroform, hexane, benzene, methylene chloride, acetone and petroleum ether (Ismail *et al*., 2010).

Lycopene concentration in fresh tomatoes may vary from 5,0 to 50,0 mg/kg, depending on the cultivar, ripening stage, and temperature during crop growth (Choudhary *et al*., 2009).

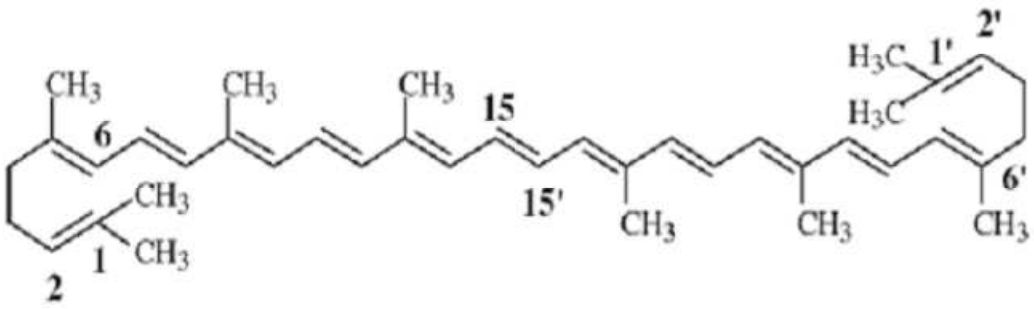


Figure 6. Structural formula of all-trans lycopene.

According studies carried out with Ismail *et al*., (2010), total world consumption of lycopene was tripled to 15000 tons in 2004 compared to 5000 tons in 1995. Thus, alternative sources for the production of natural lycopene are warranted.

1.4.8. Vitamin C (L-Ascorbic Acid)

Vitamin C (L-Ascorbic Acid) is present in all animal and plant cells, mostly in free form, and it is probably bound to protein as well. Vitamin C loss during storage of vegetables from winter through late spring can be as high as 70 %.

The oxidation of ascorbic acid to dehydroascorbic acid (Fig. 7) and its further degradation products depends on a number of parameters such oxygen partial pressure, pH, temperature and the presence of heavy metal ions are of great importance. Metal-catalyzed destruction proceeds at a higher rate than noncatalyzed spontaneous autoxidation. Traces of heavy metal ions, particularly Cu^{2+} and Fe^{3+}, result in high losses.

Fruits, vegetables, and organ meats are generally the best sources of ascorbic acid; muscle, meats and most seeds do not contain significant amounts of ascorbic acid. The amount of ascorbic acid in plants varies greatly, depending on such factors as the

variety, weather, maturity, in postharvest storage conditions of the fruit or on the environmental factors and on the applied agricultural techniques (Vermeir *et al*., 2008).

Vitamin C is one of the most important vitamins for human nutrition and plays a crucial role in several biochemical processes in the human body that is supplied by fruits and vegetables. L-Ascorbic acid (AA) is the main biologically active form of vitamin C. Ascorbic acid represents a redox system consisting of 2 L-isomers: ascorbic acid (vitamin C) in the reduced state and dehydroascorbic acid (DHA) in the oxidized state, which also exhibits biological activity (Khan *et al*., 2007**)**. Most of the vitamin's functionality in the human body is related to the role of vitamin C as an electron donor; hence, vitamin C is the active, stable form of vitamin C in tissues.

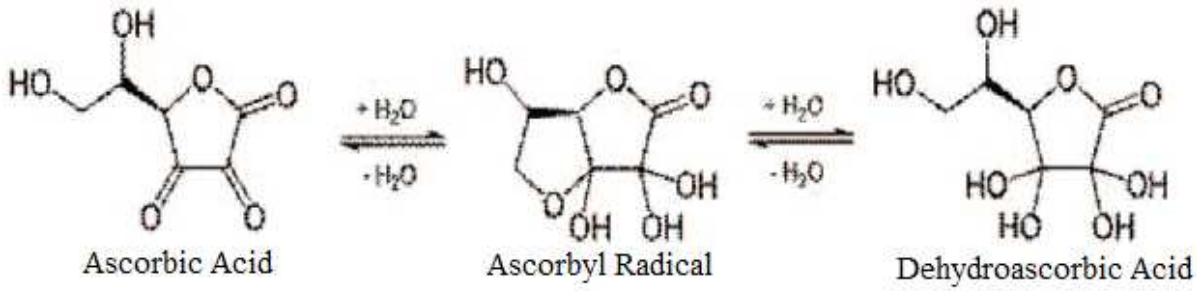


Figure 7. Chemical transformation of Ascorbic Acid

1.4.9. Consistency of Tomato juice and puree

According to Bourne 1982, the textural properties of a food are the "group of physical characteristics that arise from the structural elements of the food are sensed by the feeling of touch, are related to the deformation, disintegration, and flow of the food under a force, and are measured objectively by functions of mass, time, and distance". Knowledge of the rheological properties of fluid and semisolid foodstuffs is important in the design of flow processes in the quality control, storage and the processing stability, and in understanding and designing texture.

The rheological properties of fluid tomato products like puree are important quality parameters. The flow properties of the whole juice, referred to as the gross viscosity.

The consistency is the result of the solutes present, particularly the polymeric material, which in tomato juice consists mostly of pectins. It can be quantify by Bostwick values which are routinely determined in quality control evaluations during tomato puree production (Anthon and Barrett, 2010).

Concentrating tomato juice to puree during the tomato season allows for preservation and long-term storage. The consistency occurs on multiple stage evaporators on processing plant. Moreover, tomato puree exhibits complex rheological behavior, i.e. it is a non-Newtonian (fluids that show a small thixotropy), shear-thinning and time-dependent fluid that shows an apparent yield stress.

A number of studies have been conducted on the rheological behavior of tomato products at low concentrations, resulting in evidence that many factors play a role in determining the consistency of tomato products, including the degree of maturity, particle size and particle interactions, content of solids as well as temperature of processing. Thus different temperatures might affect its rheological properties (Bayod *et al*, 2008)

Consistency is an important quality attribute of tomato products arising from the retention of pectin and destruction of pectin methylesterase and polygalacturonase during hot-break treatment. Inactivation of pectolytic enzymes by heat results in significantly higher values of serum and efflux viscosity (Hayes *et al.* 1998, Vasiljevic, 2009).

Devices used for quality control of tomato products are the Bostwick consistometer and the Brookfield viscometer. The former allows for an empirical measurement of the distance that a specific volume of fluid can flow under its own weight in a known interval of time. The greatest contributor to the consistency of tomato products are the insoluble solids (Wayne *et al*., 1998).

1.4.10. Brix Degree (Total Soluble Solids)

Usually sugars are the soluble solids that are in the largest quantity in fruit, so measuring the soluble material in samples of the juice can give a reliable measure of its sugar content. The measurement of sugars in the fruit can provide an indication of the stage of ripeness or maturity of that fruit. Tomato solids are important because they affect the yield and consistency of the finished product.

The Brix degree is a parameter to measure soluble sugars glucose and fructose is the largest contributor to the total soluble solids in the juice of samples of tomato fruit. Sugars in tomato fruit change between the first and last harvests, storage time and processing condition and the sugar content reaches a peak in tomatoes when the fruit is fully ripe. The soluble solids are usually expressed as °Brix or % Brix (percent of sucrose by weight), measured with refractometers. (Maroulis *et al*., 2003). Soluble solids such as sugars, proteins, and lipids exert a protective action against pectinesterase inactivation by heat (Rahman, 2007 and Thompson, 2003).

During heat treatment, the reducing sugar content decreases due to caramelization, the Maillard reaction, and the formation of 5-hydroxymethyl furfural. The amount of sugar lost depends on the process. Studies have reported as much as a 19 % loss in processed tomato juice (Hui *et al*., 2004).

1.4.11. pH and Acidity

Sweetness is a function of sugar concentration and sourness is a function of acidity, and their ratio determines the flavor of fruits because consumers perceive sweetness or sourness in terms of sugar:acid ratio. Sugar concentration is usually estimated by measuring the percentage of soluble solids (% Brix) using a refractometer and acidity by titration against a standard base (solution of NaOH 0,1 N). The Brix-to-acid ratio (BAR) is used to assess the relative sweetness or sourness for most of the fruits for processing purposes.

One must consider the acidity of the fruit or its pH value. Control and manipulation of heating and pH conditions during protein concentrate processing may affect the inherent properties of the protein itself and alter the interactions of the protein with

other components of the food system. According to its acidity, the product is then heat-treated. As it is acidic food spoilage can take place, therefore a further preservation method is necessary (Hui *et al*., 2006).

The pH is a measure of the relative acidity or alkalinity of water. The pH scale runs from 0 (acid) to 14 (alkaline), with a mid-point of 7 denoting neutrality. In more scientific terms, pH is defined as the logarithm of the reciprocal of the hydrogen ion activity expressed in moles per liter. Low pH, high acidity, carbonation, and often ingredients that provide some natural antimicrobial activity.

Organic acids are major intermediary products of metabolism and are further oxidized through the Krebs cycle to provide energy for the maintenance of cell integrity. The acidity of most fresh fruits is due to these organic acids. The titratable acidity of fruits plays an important role in determining the maturity of most of these fruits. Acid content of fruits usually decreases during the ripening process as these are used in respiration or converted to sugars.

The pH of tomato has been reported to range from 3,9 to 4,9, and they can be processed as acidic foods. The lower the pH, the greater the inhibition of *Bacillus coagulans* and the less likely flat sour spoilage is to occur. The average acidity of processing tomatoes is around 0,35 %, and expressed as citric acid content. The total acid content increases during ripening to the breaker stage and then decreases. These parameters can smoothly changes during heat processing.

1.4.12. Techniques of Preservation of Tomato

In general food preservation is the process of treating and handling food to stop or slow down spoilage (loss of quality, edibility or nutritional value) and thus allow for longer storage. Preservation usually involves preventing the growth of bacteria, yeasts, fungi, and other micro-organisms (although some methods work by introducing beginner's bacteria, or fungi to the food), as well as retarding the oxidation of fats which cause rancidity. Food preservation can also include processes which inhibit visual deterioration that can occur during food preparation; such as the enzymatic

browning reaction in apples after they are cut. Foods get spoilt mainly due to the presence of micro organisms, enzymes (present in foods), insects, worms, and rats.

However, preservation implies putting micro-organisms in a hostile environment, in order to inhibit their growth so shorten their survival or cause their death. The feasible responses of micro-organisms to this hostile environment determine whether they may grow or die. Food is said to be spoilt if there is rotting (i.e. bad smell, fermentation, bubbles/gas in the food or mold, spongy growth on the food stuff). Formation of soft spots or soft brown spots on fruits and vegetables is also food spoilage.

The most important factors used in food preservation are:

- Temperature (high or low),
- Water activity (a_w),
- Acidity (pH),
- Redox potential (Eh),
- Preservatives (e. g. nitrite, sorbate, sulfite), and
- Competitive micro-organisms (e.g. lactic acid bacteria).

The spoilage and poisoning of food by microorganisms is a problem that is not yet under adequate control despite the range of preservation techniques available. In general food preservation (Gorris *et al*., 1995) can be processed by following process as:

- Evaporation,
- Oxidation (Sulphur dioxide),
- Ozonization (ozone [O3] gas or agua),
- Toxic Inhibition toxica (Carbon dioxide, acetic acid, alcohol etc.),
- Drying,
- Osmotic Inhibition (Syrup),
- Freezing,
- Ultra high pressure,
- Combined processing methods.

1.4.13. European Union Legislation of Tomato Products

In Europe, tomato products are governed by Regulation:

The Commission Regulation (EEC) No 1764/86 of 27 May 1986 laying down minimum quality requirements for products processed from tomatoes under the production aid scheme. In Article 4 and 5 references the kind of additives that tomato products are added. Only the following ingredients may be added to peeled or unpeeled tomatoes:

- water,
- Tomato juice,
- Tomato concentrate,
- Common salt (sodium chloride),
- Natural spices, aromatic herbs and their extracts, and natural aromas.

As additives in the manufacture of peeled or unpeeled tomatoes only citric acid (E 330) and calcium chloride (E509) may be used. The mould count of preserved tomatoes (the tomatoes and the covering liquid) shall not exceed 50 % positive fields and the pH level shall not exceed 4,5.

Requirements for tomato juice and tomato concentrate are reported in Article 8 to Article 10 that characterizes tomato juice. For the purposes of this title 'tomato juice' and 'tomato concentrate' mean the products defined in points 11, 12 and 18 of Article 2 of Commission Regulation (EC) No 1535/2003 (1).

The European Parliament and Council Directive No 95/2/EC of 20 February 1995, on food additives other than colors and sweeteners define tomato products as tomato pulp, puree or paste already packaged and obtained from red *Lycopersicum Esculentum* fruit.

Microbiological conditions of tomato products must follow specifications as:

- Negative presence of mesophilic bacteria;
- Negative presence of thermophilic bacteria;
- Moulds (Howard method) máx. 40 % of positive camp.

1.4.14. Tasks Prior to Validation of Control Measures

Prior to the validation of control measures by the food establishment, it is important to complete certain tasks so that validation can be accomplished effectively and efficiently. The production, manufacture and logistic tasks could be carried out either independently or in conjunction with the establishment of Global Health Professional Survey (GHPS), Hazard Analysis and Critical Control Points (HACCP), etc. In addition, as the principles of HACCP become more commonplace in the food industry, plans, the monitoring and control of their processes are regular activities.

1.4.15. Preservation of Tomato by Heat Processing

A preservation food by heat processing is important method to prevent spoilage during shelf life storage. In recent years interest is growing in the use of processed foods with low additives. Research and commercial applications have shown that natural components could preserve when tomato juice is heated (Rojas-Graü *et al*. 2009). The development of tomato products enriched with natural bio-products could contribute greatly to a new and growing market, where the consumers' concerns about their health are met (Ahmed, 2011). Industrial processing of tomato consists to inactivate enzymes in hot-break process. Then the juice is concentrated in evaporator following pasteurization, in order to destroy microorganisms by water bath or water spray retort as described in materials and methods.

II.OBJECTIVES

2.1. General Objective

The main objective of this work is to study the effect of heat processing on tomato juice or puree quality.

2.2. Specific objectives

2.2.1. Determination of the effect of pre-heating on the trituration rate. This parameter is used to see how are profit of tomato juice production and separation of peels and seeds, when it is done without and with pre-heating.

2.2.2. Study the effect of time and temperature of the hot-break process and how it effect on quality. Tomato phisical stability is supported by enzymes activity. Hot-break process is a method to inactive pectin methylesterase (PME) and polygalacturonase (PG) in order to get better consistency of product.

2.2.3. Study the effect of temperature and pressure of evaporation on the quality of tomato puree.

2.2.4. Study the effect of time and temperature of pasteurization on tomato products quality. The microbiology safety of tomato products is controlled by destroy or inactive microorganisms causing spoilage and changes physic-chemical properties. The pasteurization process was done in water spray retort and/or water bath.

2.3. Experimental Design

In order to achieve the specific objectives is done the follows studies:

2.3.1. Study 1: Effects of Preheating of Tomato Fruits on Trituration Rate

The trituration of tomato fruits is done in two ways:

- Without pre-heating of samples,
- With pre-heating of samples at 95 ºC, and 5 minutes.

The parameter of process efficiency was the ratio of juice weight by raw tomato weight.

2.3.2. Study 2: Effects of Time-Temperature of hot-break on Tomato products quality

The hot-break of tomato juice must follows combination of temperature and time (table 2), according the ranges of time and temperature of inactivation of PME and PG enzymes.

Table 2. Time and temperature of hot-break of tomato juice.

Temperature (ºC)	Time (min.)
70	11
	16
80	16
90	16
	22

The quality parameters analyzed were:

- Lycopene concentration,
- Ascorbic acid (Vitamin C) concentration,
- Consistency,

- Brix degree,
- pH, and
- Acidity.

2.3.3. Study 3: Effects of evaporation of tomato products quality

Tomato juices samples are concentrated in two systems as shown in Table 3:

Table 3. Evaporation temperature of tomato juice

	Temperature (˚C)
Atmospheric evaporation	100,0
Vacuum evaporation	65,0

The quality parameters analyzed were:

- Lycopene concentration,
- Ascorbic acid (Vitamin C) concentration,
- Consistency,
- Brix degree,
- pH, and
- Acidity.

2.3.4. Study 4: Effects of pasteurization on the tomato products quality

The pasteurization process is done in two systems as shown in Table 4:

Table 4. Pasteurization condition of tomato products.

	Temperature (˚C)	**Time (min)**	**Pressure**
Water bath	100,0	14,0	Atmospheric
Water spray retort	110,0	10,0	1500 mbars

The quality parameters analyzed were:

- Lycopene concentration,

- Ascorbic acid (Vitamin C) concentration,
- Consistency,
- Brix degree,
- pH, and
- Acidity.

III. MATERIALS AND METHODS

3.1. Materials and Equipments

Tomato fruits (*Roma* variety) were purchased in a local grocery store. Pilot Plants, Equipments, Apparatus, Laboratory Accessories and Reagents used at practical work were belonging to Department of Technology Alimentary in University Public of Navarra- Spain.

To achieve above objectives preliminary experiments were conducted to define the correct methodology to work.

The materials used tomato processing was:

- Plastic dishes,
- Beakers,
- Kitchen pots,
- Spatulas,
- Sieves with (850 μm of hole size),
- Glass bottles,
- Cans.

Equipments:

- Trituation Machine model (Robot Coupe C80),
- Balance model (COBOS 2K12 and C-300-SX),
- Temperature Control Systems,
- Heat plate model (IKAMAG-RH),
- Vacuum Evaporator Systems model OPPAC,
- Data Logger Systems,
- Water Spray Retort model (MicroMar 21581),
- Agitator Systems.

3.2. Methods

3.2.1. Processing of Tomato

Processing of tomato was conducted as it is described in Figure 8.

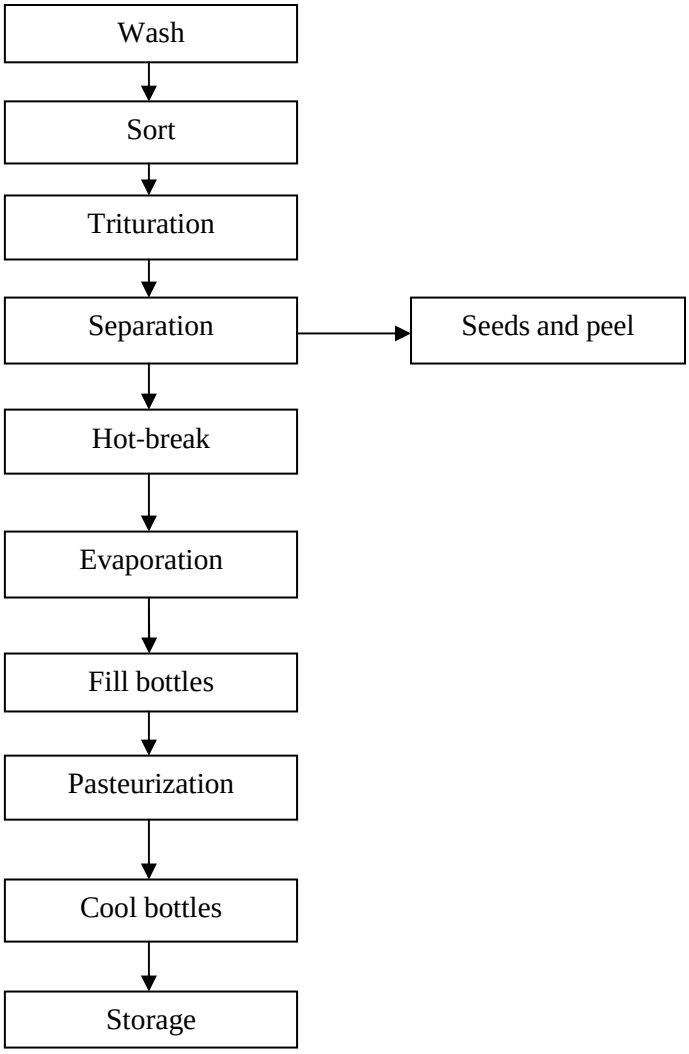


Figure 8. Processing of tomato products diagram.

All steps were performed in subdued lighting at room temperature. Satisfactoriness' results were got by trying more than one time experimental procedures of analysis in laboratories of the Department of Technology Alimentary – UPNA. Technical support was done by pointed specialists with know-how of food process industries.

3.2.1.1. Washing

Tomato fruits was processed in 1,5 kg each sample. The samples was washed with water contain sodium hypochlorite solution (65 ppm of active chlorine), then cleaned with potable water in order to remove residual chlorine Fig. 9a).

3.2.1.2. Sorting

The parameter used for tomato sorting is color and physic state of fruits.

3.2.1.3. Trituration

Trituration process was done with triturating machine. It consists to introducing tomato a fruit into machine and then tomato juice is collected separated to seeds and peels (Fig.9b). Triturating condition is done according study 1.

a) b)

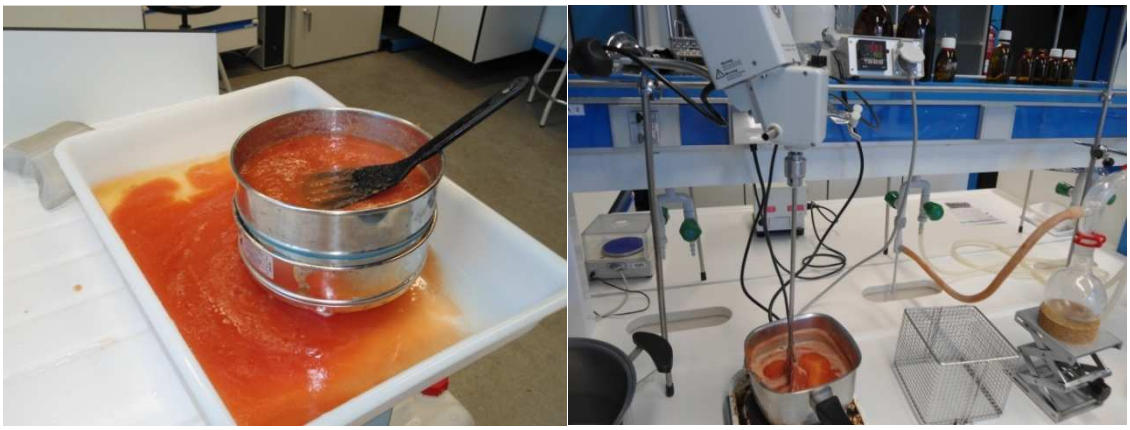
Figure 9. Tomatoes washing a) and b) Triturating process.

3.2.1.4. Separation

Tomatoes juices is then pressure forced through finishers of sizes screens 850 μm in a process called pulping (Fig. 10a).

3.2.1.5. Hot-break

The juice are rapidly heated to produce either hot-break processing (Fig. 10b), involves heating tomato to different temperature and times combination as mentioned in study 2.



a) b)

Figure 10. Tomato pulping a) and b) Hot-break process.

3.2.1.6. Evaporation

The mechanism of concentration tomato juice is done in evaporation process. It consists in evaporate water into samples to achieve 15 % Brix of soluble solids concentration. For this step was used two systems, one was putting tomato juice into kitchen pot then it is heated to boil temperature of sample (Evaporation in atmospheric pressure) and other was introducing tomato sample into closed systems were vacuum pressure and heat is applied, then the sample could boil at low temperature (65 ºC) (Vacuum evaporation systems), as shown in Fig. 11a.

a) b)

Figure 11. The Evaporation systems of tomato juice a) and b) Retort systems.

3.2.1.7. Pasteurization

After evaporation, the puree is canned or bottled and pasteurized in water bath (approximately 100,0 ^{o}C for 14 minutes, in atmospheric pressure) or in water spray retort (approximately 110,0 ^{o}C for 10 minutes, in 1500 mbars), flash cooled and stored environment temperature (Fig. 11b). The length of time of pasteurization was reference to range were temperature was constant.

3.2.2. Qualities Parameters Analysis

Physic-chemical analysis methodologies to quantify analytical parameter (color, Brix degree, viscosity, pH, acidity, concentration of lycopene and vitamin C) of tomato products, was done according Reference Methods of Analysis as AOAC 2000 and UNE.

3.2.2.1. Color measurement

The surface color of the tomatoes was measured at random spots near the equatorial region of each fruit. The color of tomato juice or puree was measured by glass cell method.

The aims of this analysis were to verify the stage of tomato fruit mature and color changes of tomato juice and puree in processing steps.

3.2.2.1.1. Laboratory Equipments and Materials

- Colorimetric Spectrophotometer model CR 2500d,
- Glass cell,
- Beaker,
- Distillate water,
- Absorbent paper.

3.2.2.1.2. Methods of Analysis

Color of tomato fruit, tomato juice and tomato pulp samples were measured using a colorimetric spectrophotometer (model CR 2500d) calibrated with reference color tiles. The color values were expressed as L* (lightness or brightness/darkness), a* (redness/greenness) and b* (yellowness/blueness). The L* parameter which indicates the magnitude brightness of color change and the parameter (a*/b*) for maturation of tomato fruits are determined. Color measurements were taken in triplicate. For tomato juice and tomato pulp was used glass cell cylindrical cuvette.

3.2.2.1.3. Samples of tomato

The tomato sample was get at each phase of process and direct measured with spectrophotometer (Fig. 13).

Figure 13. The color measurement of tomato fruits.

3.2.2.1.4. Samples Evaluation

Other methods of colour measure were taken from five separate points on surface of the samples using colour spectrophotometer (example, CR2500d, Minolta, Japan). Before readings were made, the colorimeter was calibrated against a standard tiles. The results are expressed in L* and a*/b* parameters.

3.2.2.1.5. Cleaning Protocol

The glass cell, colorimeter glass and beakers were cleaned by distillate water and current water and dried by absorbent paper.

3.2.2.2. Lycopene determination

3.2.2.2.1. Laboratory Equipments and Materials

- Spectrophotometer model Cintra 20, UV-Visible GBC.
- Volumetric flasks
- Beaker
- Micropipettes
- Balance
- Pipettes
- Aluminum paper
- Filters;
- Funnel;
- Magnetic stirrer
- Quartz cuvettes

3.2.2.2.2. Characteristics of Chemical Reagents

All chemical products used in analysis were quality reagents for analytical determination as:

- Hexane,
- Acetone and
- Ethanol.

For dilution distillate water was used and for clean used pump water.

3.2.2.2.3. Methods of Analysis

The reference method of analysis applied in lycopene determination is Standardization of a Rapid Spectrophotometric Method for Lycopene Analysis proposed by the California League of Food Processors and recommended by Department of Food Science and Technology of University of California-Davis. The methodology consisted of the extraction of lycopene compound present in the tomato

puree with solution of Hexane:Ethanol:Acetone(HEA). Then, the lycopene extract was separated in organic phase adding distillated water.

3.2.2.2.4. Preparation of Reagents

Lycopene in tomato sample is extracted by mixer of Hexane:Ethanol:Acetone: (HEA) in proportion of 2:1:1.

First is prepared 8,0 ml of reagent solution with 4,0 ml of hexane, 2,0 ml of acetone and 2,0 ml of ethanol then is used extraction.

3.2.2.2.5. Samples of Tomato and Lycopene Extraction

All steps were performed in subdued lighting at room temperature. Approximately 100 µL (equal to 100 mg) of each varietal sample were measured. Tomatoes sample were obtained in processing step in each phase of treatment and different conditions trituration to canning. Then it is weighted and mixed with the HEA solvent and incubated during 10 minutes, after that 1.1 ml of distillate water is added to separate phases and the lycopene absorbance of the upper layer determinate. Samples were mixed by using a vortex when the solvent was added and again after the addition of the water.

The literature is full of statements that, once extracted into organic solvents, lycopene becomes highly susceptible to isomerization and/or degradation by light and oxidants. To minimize it samples was prepared in low light environment and at covered flasks.

The steps involved in the procedure were as follows:

1. Weight 100,0 mg of tomato pulp in beaker.
2. Add 8,0 ml of 4:2:2 (v/v) Hexane : Ethanol : Acetone using a pipette. This solvent should be mixed fresh daily to volumetric flask.
3. Cap and vortex the volumetric flask then incubate, out of bright light, with occasional vortexing.
4. After 10 minutes add 1.1 ml distillated water to each sample and briefly shake.

5. Let samples stand few minutes to allow phases to separate and all air bubbles to disappear.

6. Remove a sample of the hexane layer and read absorbance (λ = 503 nm) in the spectrophotometer. To zero the spectrophotometer prepares one or two samples with 100 µl water instead of tomato pulp. It is important that the cuvette be rinsed with the hexane layer from this zero sample prior to reading the lycopene samples

3.2.2.2.6. Sample Evaluation

Quantification of lycopene is by spectrophotometer model (Cintra 20, UV-Visible GBC) which determine the concentration of lycopene in the extract. The determination of the upper layer volume was based on the following. According to Beer's Law, the optical absorbance (at λ= 503 nm) of a lycopene solution is proportional to the lycopene concentration which it is the amount of lycopene per volume (Borges-Miquel, 2007).

In this case, the Lambert–Beer law can be described as:

$$A_{503} = \varepsilon(M^{-1}\cdot cm^{-1})\cdot b(cm)\cdot[\text{Lycopene concentration (M)}] \quad (1)$$

Where: A_{503}- Absorbance at λ=503 nm,

ε- Coeffient of extinction,

b- cell length.

The extinction coefficients for lycopene in hexane were determined first by Zechmeister (1943) and a value of 172,000 M^{-1} cm^{-1} for 503 nm is generally accepted. Using this extinction coefficient, the concentration of lycopene in a hexane extract can be determined from the absorbance of the solution at 503 nm. To convert this concentration into the weight of lycopene present in the original tomato sample, four additional numbers need to be factored in:

1. The ratio of the upper layer volume to the volume of mixed extracting solvents added, 0.55 for hexane:ethanol:acetone (2:1:1), as discussed above.
2. The molecular weight of lycopene, 537 (g/mole).

3. The weight of tomato juice analyzed (or its volumetric equivalent as described above), W (mg).
4. The volume of mixed solvents added, V.

Duplicate samples (a and b) of lycopene were analyzed in each sample. Lycopene concentration C was expressed as the average value in mg/ kg fresh weight, according the following equation:

$$\mathbf{C\ (mg/kg) = 137.4 * A_{503}} \quad \mathbf{(2)}$$

Where: A_i – sample absorbance.

3.2.2.2.7. Cleaning Protocol

All materials and apparatus were cleaned with current water and distillated water then dried with absorbent paper.

3.2.2.3. Ascorbic Acid (Vitamin C) determination

In ascorbic acid analysis important reaction occurs when reagents are mixed. Ascorbic acid is oxidized to dehydroascorbic acid by the action of bromine solution. L-dehydroascorbic acid reacts with 2,4-dinitrophenylhydrazine and produces an osazone which on treatment with 85 % H_2SO_4 forms red colored solution.

3.2.2.3.1. Laboratory Equipments and Materials

- Spectrophotometer model Cintra 20, UV-Visible GBC.
- Cuvettes
- Volumetric flasks
- Beakers
- Pipettes
- Incubator
- Graduate Provette

- Burette
- Glass watch
- Filter
- Erlenmeyer
- Funnel
- Balance
- Stirrer
- Extractor

3.2.2.3.2. Characteristics of Chemical Reagents

For ascorbic acid analysis was used reagent as:

- 2,4-dinitrophenylhydrazine,
- Thiourea,
- Sulphuric acid 85 % v/v,
- Metaphosphoric acid,
- Acetic acid,
- Ethanol,
- L-ascorbic acid,
- Bromine solution.

3.2.2.3.3. Methods of Analysis

Reference method for ascorbic acid analysis was according Joseph, 1960 and Rahman, 2006, “A Simple UV-spectrophotometric Method for the Determination of Vitamin C by the 2,4-Dinitrophenylhydrazine in Fruits and vegetables”.

3.2.2.3.4. Samples Evaluation

1. Samples must be under 50 and never less than 20 part of a solution containing 5 % metaphosphoric acid and 1 % of Thiourea solutions.
2. Put 4 ml of filtrated sample at spectrophotemetric cuvettes.

3. One is kept for a blank.
4. Put 1 ml of 2 % 2,4-DNPH solution to each of others in approximately 9 N sulfuric acid.
5. Held at 37,0 ºC for 3 hours.
6. Cool together with blank, in ice water.
7. To each three tubes, while in the ice water bath, put 5,0 ml of 85 % H_2SO_4 from burette a drop at a time, during not less than 1 minute.
8. Finally put 1 ml of the 2 % 2,4-DNPH solution at blank tube.
9. Shake the tube thoroughly under the ice water.
10. Remove to a rack, wipe dry after 30 minutes.
11. Read in a spectrophotometric with λ = 521 nm.

Spectrophotometric readings should be taken 30 to 45 minutes after removal from the ice bath. In some tomato extracts a slight yellowish coloration appears when the 85 % H_2SO_4 is added (Fig. 14a).

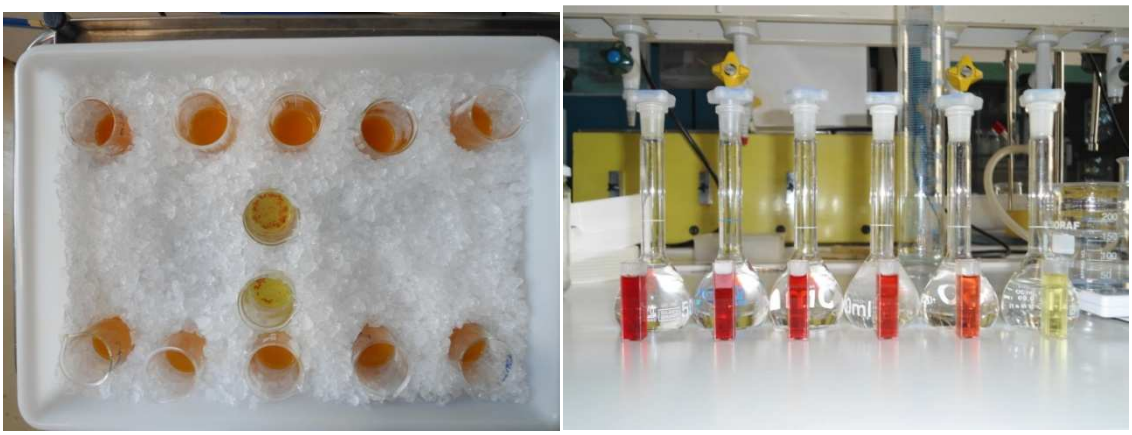


Figure 14a. Tomato extracts for ascorbic acid determination.

3.2.2.3.5. Calibration curve with standard solution

Absorption spectra were recorded at the wavelength λ = 521 nm in 1 cm polystyrene cuvettes using a GBC UV/Visible spectrophotometer Cintra 20 (GBC Scientific Equipment Pty Ltd, Dandenong, Australia) with data processing Spectral Software. The calibration curve was obtained by standard ascorbic acid colored complex samples in triplicate. The absorbances of different concentration were taken to construct a

calibration curve. The calibration curve (Fig. 14b) was constructed by plotting the concentration C (ppm) versus the corresponding absorbance (Abs). The results shown regression equation (R^2 = 0,9345):

Abs = 0,0086*C + 0,0358 (4)

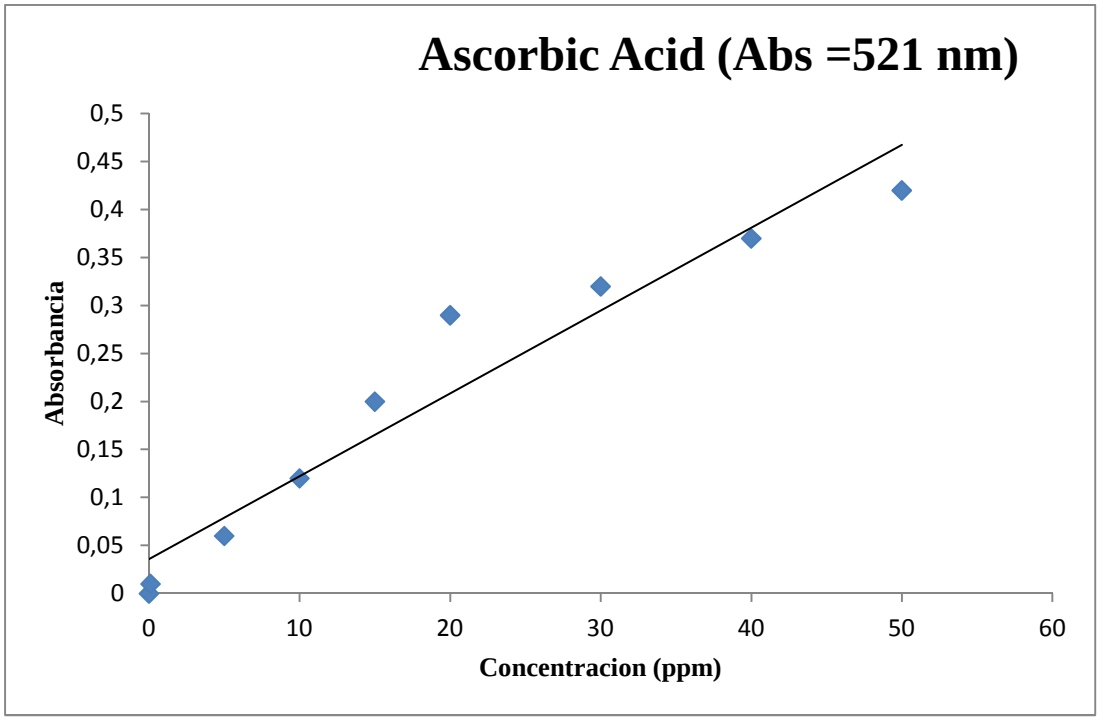


Figure 14b. Calibration graphic of standard ascorbic acid solution.

Concentration range of standard solution from 0,25 to 15,0 mg/ml.

1. Weight 25,0 mg of ascorbic acid and pipette 25,0 ml of 5 % metaphosphoric acid.
2. Mixer it then shakes well.
3. Add 1 or 2 drops of bromine water, shaken until yellow.
4. Dilute with 5 % metaphosphoric acid.
5. Add 1 % Thiourea.

3.2.2.3.6. Ascorbic Acid Reactions

1. Ascorbic acid is oxidized to dehydroascorbic acid by the action of bromine solution.
2. L-dehydroascorbic acid reacts with 2,4-dinitrophenylhydrazine and produces an osazone which on treatment with 85 % H_2SO_4 forms red color solutions.

3.2.2.3.7. Results Equation Expression

$$\textbf{\% of AA loss} = \frac{C_i - C_f}{C_i} \; x \; 100 \qquad (3)$$

Where: C_i – initial concentration of ascorbic acid,

C_f – final concentration of ascorbic acid.

3.2.2.3.8. Cleaning Protocol

All materials and apparatus were cleaned with current water and distillated water then dried with absorbent paper.

3.2.2.4. Consistency measurement

3.2.2.4.1. Laboratory Equipments and Materials

- Bostwick consistometer,
- Beaker,
- Thermometer,
- Heat plate.

3.2.2.4.2. Methods of Analysis

The most commonly used methods for the evaluation of textural properties are those that apply large deforming forces (e.g., via puncture or compression) and are therefore destructive. Because of the empirical nature of these tests, however, they do not provide us with an understanding of food microstructure or force-deformation and failure mechanisms at the cellular level (Jackman and Stanley, 1992). Recently, there has been

a resurgence of interest in nondestructive tests that rely on well-defined fundamental principles and thereby may provide a better understanding of tomato tissue microstructure and the forces that lead to tissue failure. Both destructive and nondestructive tests are Bostwick consistency measurement (McCarthy *et al*., 2008).

The viscosity of the tomato juice and puree was measured at 20,0 ºC, during 30 s in a controlled consistometer at planed surface. The proceeds (Fig. 15) were:

1. The compartment was loaded with 75,0 ml of tomato sample and
2. The gate was opened and distance is measured.

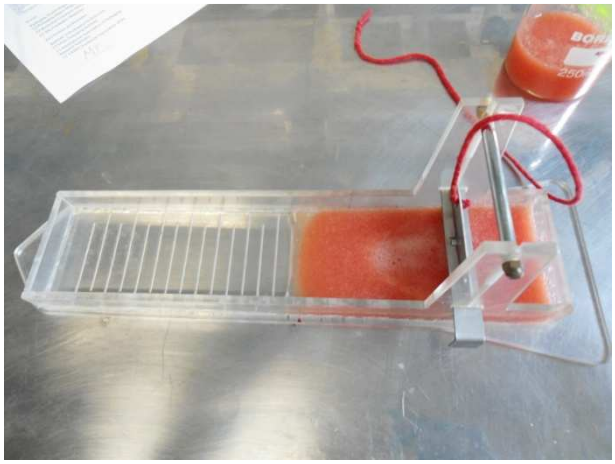


Figure 15. Bostwick consistometer filled with tomato sample.

All rheological measurements were carried out at least in duplicate. The maximum relative standard error (RSE). Same samples were evaluated after 24 h and the results are expressed in (cm/30 s).

3.2.2.4.3. Cleaning Protocol

All materials and apparatus were cleaned with current water and distillated water then dried with absorbent paper.

3.2.2.5. Brix Degree (Total soluble Solids)

3.2.2.5.1. Laboratory Equipments and Materials

- Refractometer model ATAGO 7000 α,
- Funnel,
- Erlenmeyer and
- Paper filter.

3.2.2.5.2. Methods of Analysis

Total Soluble Solids for tomato products is determined according AOAC Official Method 970.59/2000. The tomato sample was filter and then it is directly measured in refractometer. Adjust refractometer for 0 % Brix with H_2O at 20,0 ^oC.

3.2.2.5.3. Samples Evaluation

Steps for Brix degree measurement:

1. Test sample was filter into beaker until filtrate is clear,
2. Quickly funnel was removed and transferred large drop of filtrate directly from funnel to refractometer prism, and
3. Read refractometer, preferably at 20,0 ^oC.

The results are directly read in apparatus screen (Figure 16) and expressed in % Brix (the quantity of sucrose in tomato sample).

Figure 16. Refractometer with test sample.

3.2.2.5.3. Cleaning Protocol

The prism glass is washed with distillate water and dried with absorber paper. The materials are washed with pump water.

3.2.2.6. pH

3.2.2.6.1. Laboratory Equipment and Materials

- pH meter model CRISON Basic 20,
- Beaker,

3.2.2.6.2. Methods of Analysis

The reference method for pH determination was pH of Acidified Foods AOAC Official Method 981.12/2000

3.2.2.6.3. Fundaments of Analysis

The pH is measurement of Hydrogen ion activity and indicates acidity. It may be measured by determining electric potential between glass and reference electrodes, using commercial apparatus standardized against NIST primary standard pH buffers solutions. Standard buffer solutions were pH = 4,0 and 7,0 for calibration.

3.2.2.6.4. Samples of Tomato

Sample obtained from tomato juice or puree already processed and let temperature equilibrate to 25,0 ^{o}C, and then determine pH by immersing electrodes in sample (Fig. 17).

Figure 17. pHmeter with test tomato juice sample.

3.2.2.6.5. Samples Evaluation

1. The instrument was switch on and let electronic components warm up and stabilizes before proceeding.
2. The electrodes were equilibrating in buffers solution, and samples at same temperature (25,0 ^{o}C) before pH measurements.
3. Set temperature compensator control of instrument at observed temperature.
4. The electrode tips was Immerse in buffer solution and read pH = 7,0, letting meter stabilize 1 min, and repeat it with buffer solution pH = 4,0.
5. Measure the sample after calibration proceeds.

The results were obtained by direct reading values in pH meter screen.

3.2.2.6.6. Cleaning Protocol

The electrode is watched with distilled water and dried with absorber paper. The materials are washed with pump water.

3.2.2.7. Acidity determination

3.2.2.7.1. Laboratory Equipment and Materials

- pH meter model CRISON Basic 20,
- Beaker,
- Burette,
- Agitator.

3.2.2.7.2. Methods of Analysis

The acidity (Titratable) of tomato products was done according "Norma Española UNE 34-211-81: Determinacion de la acidez valorable para productos derivados de frutas y verduras".

3.2.2.7.3. Characteristics of Chemical Reagents

- Standard buffer solutions pH = 4,0 and 7,0,
- Sodium hydroxide (NaOH) 0,1 N.

3.2.2.7.4. Fundaments of Analysis

Titratable acidity can be expressed conventionally in g acid per 100 g or per 100 mL product, as appropriate, by using the factor appropriate to the acid as 0,070 for citric acid.

3.2.2.7.5. Samples Evaluation

1. Before use, check apparatus with standard buffer solutions pH 7,0 and 4,0.
2. Rinse glass electrode in H_2O several times until reading is pH = 6,0.
3. Immerse electrodes in test sample contained in beaker. (Test sample should titrate 10-50 mL 0,1M NaOH and be contained in initial volume of 100-200 mL.) Stir moderately.
4. Add alkali quite rapidly until near pH= 6,0. Then add alkali slowly to pH=7,0. After pH=7,0 is reached, finish titration by adding 0,1M alkali 4 drops at time, and record total volume and pH reading after each addition.

5. Continue titration 4 drops beyond pH 8,1, and interpolate data for titration corresponding to pH 8,1. pH values used for interpolation should lie in range 8,1 ± 0,2.

3.2.2.7.6. Results Equation Expression

$$\% \text{ Citric Ac.} = (V_{NaOH} * C_{NaOH} * 0{,}070 * 100) / V_{sample} \quad (4)$$

Where: V_i – tritratable volume of solution and sample;

C_i – concentration of solution.

3.2.2.7.7. Cleaning Protocol

The electrode is cleaned with distilled water and dried with absorber paper and materials are washed with pump water.

IV. RESULTS and DISCUSSION

4.1. Raw Material Characterization

In this study the raw tomato properties was characterized by physic-chemical parameter as color, pH acidity, Brix degree, viscosity, vitamin C and lycopene concentration (Table 5). For destructive analysis the sample was double measured and for non destructive analysis the sample was triplicate measured. In general, the sample weight 1,5 kg of tomato fruit each and 9 samples in total.

Table 5. Average values of Physic-Chemical Characterization of raw tomatoes

	Color		pH	Acidity	Brix(%)	Viscosity	Vitamin C	Lycopene
	L	a/b		(%)		(cm/30 s)	(mg/100 g)	(mg/100 g)
Average(SD)	32,44 (0,42)	0,86 (0,06)	4,1 (0,1)	0,33	5,75 (0,57)	8,2 (0,5)	33,9 (3,6)	14,7 (1,4)
CV (%)	1,26	7,07	1,12	3,03	10,0	6,1	10,5	9,5

Table 5 summarizes the characteristics of raw tomatoes used to experimental work. It was found that tomato maturity was good to processing with parameter a*/b* (0,86) and a low pH (4,1), acidity (0,33). However, the ratio a*/b* and acidity is associated to variability of cultivar, harvest and storage condition. The content of vitamin C was 33,9 mg/100 g and lycopene was 14,7 mg/100 g and it is above of lycopene concentration in commercial canned tomato puree.

Lugasi *et al*., 2003, find significantly difference of lycopene content in tomato samples in function of harvest season. They concluded that in winter the lycopene concentration is low level, around 1,0 mg/100 g and summer season had high level of lycopene ranged from 7,6 to 13,6 mg/100 g.

4.2. Study 1: Effects of Preheating of Tomato Fruits on Trituration Rate

The trituration rate is affected by efficiency of juice, seeds and peels separation and maturity of tomato fruits. Tables 6 illustrate the average values of trituration process of tomato fruits with preheating (95 ºC, 5 min.) in water bath and without preheating. In this experimental work was used a sieve size 850 μm for juice separation (Fig. 10).

Table 6. Trituration results of raw tomatoes with and without pre-heating (95 ºC, 5 min.).

		Initial weight (kg)	Final weight (kg)
Without preheating	**Average (SD)**	1,53 (0,09)	0,82 (0,08)
	CV (%)	0,06	9,67
	Trituration Rate (%)	53,8	
With preheating	**Average (SD)**	1,38 (0,17)	0,96 (0,12)
	CV (%)	12,77	12,81
	Trituration Rate (%)	69,24	

CV-Coefficient of variation (%); SD- Standard Deviation.

According the two conditions of trituration methods provided different quantities of tomato juice. We observed that good separation was obtained when tomato fruits are preheated before triturating with 69,24 % of trituration rate (see table 4).

4.3. Study 2: Effects of Time-Temperature of hot-break on Tomato products quality

Heat treatment caused significant changes in tomato juice, mainly the physical stability of tomato products in self time storage and changes of some compound as ascorbic acid and lycopene. Heat treatment did not affect pH and acidity during tomato processing.

In Table 7 shown the results obtained in study 2, with the temperature of the hot-break 70,0 ºC, during 11 and 16 minutes respectively, 80,0 ºC, 16 minutes and 90,0 ºC, 16 and 22 minutes respectively.

Results on the behavior of vitamin C were comparable to existing data. The vitamin C content of tomatoes depended on the species, season and the cultivation conditions. For our investigations, *Roma* tomatoes were used with a vitamin C content of 25,3 – 37,7 mg/100h fresh weight which come from Spain land and produced in winter season. Many studies showed the decline in vitamin C during the production and cooking (Bohm *et al*., 2003). With increasing heating time and processing steps of tomato puree, a continuous loss of the water soluble vitamin C was observed in our investigations.

Table 7. Hot-break of tomato juices and puree.

Samples Temperature (ºC)	**Time (min)**	**pH**	**Acidity (%)**	**Brix(%)**	**Viscosity (cm/30 s)**	**Vitamin C Loss (%)**	**Licopeno (mg/100 g)**	**Physisc stability**
Control	-	4,1 (0,2)	0,33	4,60 (0,09)	8,4 (0,2)	-	14,5 (2,2)	-
70	**11**	4,1 (0,1)	0,32	4,56 (0,11)	8,3 (0,1)	5,82	9,8 (4,1)	PS
	16	4,1 (0,3)	0,32	4,55 (0,05)	8,5 (0,2)	15,85	13,3 (0,1)	
80	**16**	4,0 (0,1)	0,33	11,19 (0,17)	5,4 (0,1)	19,1	14,9 (0,6)	NPS
90	**16**	4,1 (0,1)	0,31	11,06 (0,04)	4,5 (0,1)	21,46	14,0 (0,3)	
	22	4,0 (0,3)	0,32	11,30 (0,07)	4,2 (0,1)	25,30	17,4 (0,4)	NPS

PS-Phase separation; NPS- Non Phase Separation.

According the experimental results, the lycopene concentration increase with increasing of process temperature and ascorbic acid concentration decrease in same order.

To compare the extent of lycopene changes observed in this present study with the other literature studies, the retention of lycopene was determinate after 24 hours of storage, from lycopene concentration parameter change reported in tables below.

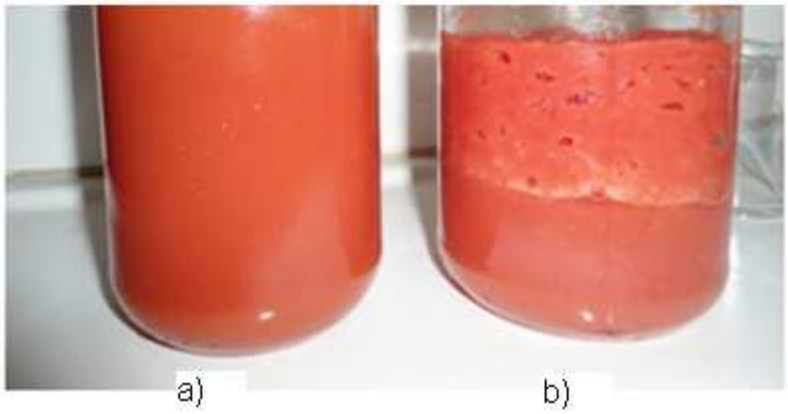


Figure 18. Tomato puree after heat-treatment. The sample a) in left was submitted at 80,0 ºC and b) in right at 70,0 ºC of hot-break temperature.

These results agree with those reported by Barringer *et al*. 2001 and Lavelli *et.al*. (2010), who described lycopene concentration increase of 9-28 % after thermal processing of tomatoes into paste, rising with the extension of the heating.

After heating about 16 and 22 min at 90,0 ºC, less than 21,46 and 25,30 % of the original ascorbic acid could be loss in tomato products. Heat treatment caused a significant increase in samples treated at 90,0 ºC of hot-break temperature. The breakdown of the cell structure after 24 hours could be seen at physical stability of tomato product, which allowed a phase separation from the juice matrix at sample treated at 70,0 ºC of hot-break.

These results according Jacob *et al*., 2010, show that domestic cooking might cause greater losses of ascorbic acid than industrial production of tomato paste (28 % Brix), in which vitamin C loss was determined to be about 55 %. The reason might be that the industrial process was carried out under vacuum, whereas domestic cooking occurs under normal atmosphere. Daood *et al*., 2000 and Barringer, *et al*., 2001, concluded in his study that during hot-break extraction, tomato lost about 38 % of the original ascorbic acid, while further processing to produce tomato paste by vacuum evaporation caused the product to lose more than 16 % of its ascorbic acid content. That's happened too when duration of process is long time low temperature and short time high temperature. The degradation of ascorbic acid in aqueous solutions depends on a number of parameters as oxygen, partial pressure, pH, temperature, light and the presence of heavy metal ions of great importance.

4.4. Study 3: Effects of evaporation of tomato products quality

Comparing the results obtained at control samples, we fixed the samples processed at 80 ºC, 16 minute, to evaluate evaporation condition on tomato products. The evaporation temperature was 65 ºC and vacuum system. The tomato juice with 5,75 % Brix was concentrated to15,0 % Brix, the measured Bostwick was 3,5 ± 0,2 cm, in 30 s (Table 8). The value is into range predicted by McCarthy *et al*., 2009, Bostwick viscosity range of 3 to 7 cm for tomato puree with 15 % Brix at 20 ºC and 30 s.

Table 8. Evaporation results of tomato juice to get tomato puree with 15,0 % Brix.

Evaporation	pH	Acidity (%)	Brix(%)	Viscosity (cm/30 s)	Vitamin C Loss (%)	Licopeno (mg/100 g)
Atmospheric	4,1 (0,2)	0,34	15,0 (0,7)	3,4 (0,4)	27,96	20,6 (0,5)
Vacuum	4,0 (0,1)	0,33	14,9 (0,5)	3,5 (0,2)	20,28	17,4 (0,4)

Comparing the results of atmospheric evaporation and vacuum evaporation test system, differences can be seen. The increase of concentration of tomato juice after evaporation process.

The consistency of tomato purees vary depending on a number of factors including the ripening stage of the tomato fruits and processing conditions. For consumer preference, tomato purees with a high consistency or viscosity (i.e., with low Bostwick values for the same Brix concentration) are very valuable.

4.5. Study 4: Effects of pasteurization on the tomato products quality

4.5.1. Lethality Quantification of Pasteurization of Tomato products

Pasteurization and sterilization process results are represented in Tables 9. To illustrate microbial lethality time-temperature of tomato sample was measured. The tomato puree pasteurization and sterilization was done in water bath and water spray retort, respectively. The lethality quantification was based in reference microorganism's existent in acidity food (as thermoacidophiles bacteria). The calculation using the lethality curve is specifying by $F_{(93;8,6)}$ and F_0 value for the required process.

Thus the *F* value is equal to integral area of temperature diagram, it can simplify as:

$$F = \Delta t \; x \; \sum_0^n 10^{\frac{T_i - T_{ref}}{z}} \qquad (5)$$

Where: T_i –Sample temperature evaluation each minute in centre of can or bottle;

T_{ref} –Reference temperature (93,0 ºC for water bath and 110,0 ºC for water spray retort), and

z – For water bath retort z = 8,8 ºC and for water spray retort z = 10,0 ºC.

In this way information regarding specific microorganisms or numbers of log-cycles reduction can be replaced by the *F* value (lethality) as a process specification (Heldman, 2007).

Table 9. Pasteurization of tomato puree in water bath (100,0 ºC, 14 min) and water spray retort (10,0 ºC, 10 min).

Sample	F0 (Water Spray Retort)		F (93/8,8) (Water bath)	
	Average(SD)	CV (%)	Average(SD)	CV (%)
Tomato Puree	1,32 (0,17)	12,88	32,8 (2,7)	8,35

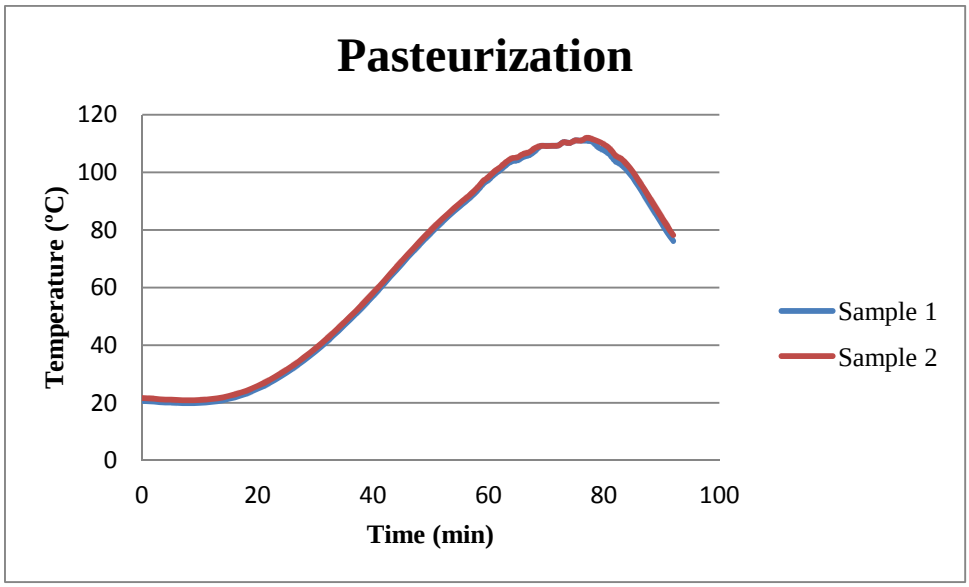


Figure 19a. Temperature and time evaluation at critical point of product in water spray retort (110 ºC and 10 minutes).

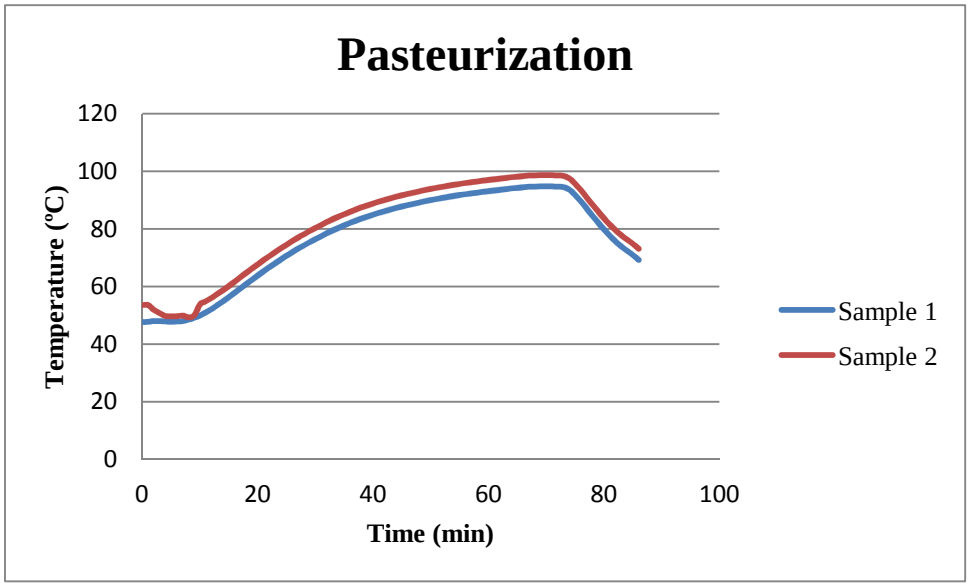


Figure 19b. Temperature and time evaluation at critical point of product in water bath (approximately 100 ºC and 14 minutes).

To evaluate time and temperature pasteurization (Fig.19 a) and b)) were used data logger systems. Microbiological safety of tomato products is closely associated with the quality of raw materials and hygienic practices on farms and in the food processing plants (Verran *et al.* 2008).

The results of the present study showed that both thermal treatment were safety because the values obtained is F0 = 1,32 for sterilization and $F_{(93;8,8)}$ = 32,8 for pasteurization. The reference values of this kind of products is F_0 = 0,75 for sterilization and $F_{(93;8,8)}$ = 20 for pasteurization, if pH < 4.2 of tomato products.

Depending on the hygiene standards applied at the processing facility, there is a low probability of the recontamination of the tomato product by microorganisms.

Table 10. Results of tomato puree after pasteurization.

Pasteurization	pH	Acidity (%)	Brix(%)	Viscosity (cm/30 s)	Vitamin C Loss (%)	Licopeno (mg/100 g)
Water bath	4,0 (0,2)	0,34	15,0 (0,7)	3,3 (0,5)	21,66	17,9 (0,4)
Retort	3,9 (0,3)	0,33	14,9 (0,5)	3,5 (0,3)	22,28	18,4 (0,6)

In Table 10 we can see the influence of residence time in pasteurization systems of tomato puree on quality. Resuming these results, lycopene content increase by increasing of temperature and ascorbic acid decrease significantly.

V. CONCLUSION

In conclusion, although this work is experimental model in laboratory, it allowed applying in industrial process to critically analyze the role of the involved quantities, and to obtain novel results of broad scientific interest. The control of food process quality in modern industrial plants is essential issues for alimentary safety and providing nutritional foodstuffs for consumers.

The state and composition of raw materials determine the quality of final products as tomato puree. In agro alimentary industry quality management is a focus point and it must be based in principles of HACCP (Hazard Analysis and Critical Control Points) and good practice of production and manufacture.

The raw tomato used to analytical experimental work has got good quality. It present acidity food characteristics with pH = 4,1 ± 0.1 and acidity = 0,33 ± 0,01 %. The Brix degree, lycopene and ascorbic acid concentration values are normal range as mentioned with other researchers as 5,75 ± 0,57 % Brix, 14,7 ± 1,4 mg/100 g and 33,9 ± 3,6 mg/100 g, respectively. Trituration rate with pre-heating of tomato fruit at 95,0 ^{o}C, 5 minutes was 69,24 %.

The main objective of food preservation is to potential long shelf life storage. For tomato products heat treatment could apply in order to inactive enzymes and destroy microorganisms. Thermal inactivation of pectin methylesterase (PME) and polygalacturonase (PG) in tomato juice is important for physical stability of tomato puree. To avoid high loss of ascorbic acid and others volatile components in tomato juice, it was postulated that the condition of temperature and time of hot-break were 80 ^{o}C and 16 minutes. These conditions would be enough to reduce PME and PG activity in tomato juice to the desired level of non separation of liquid-solid phases.

Even tomato puree must have 15 % Brix of soluble sugar concentration for our objective and experimental conditions; tomato juice was submitted in vacuum evaporation at 65,0 ^{o}C to concentrate. Heat processing has considerable effect on ascorbic acid concentration loss and lycopene concentration increase in tomato puree, 20,28 % and 17,4 ± 0,4 mg/100 g, respectively. The consistency was 3,5 ± 0,2 cm/30 s, according Bostwick consistency method.

Microbial destruction was done by pasteurization and sterilization process. Although the level of microbial contamination generally did not quantified in the production process, the contamination remained relatively low level even after sanitation. Thus, this study reveals that more efficient sanitation programs should be adopted. For example, tomato fruits should be washed in the early stages of process (at least three times during the food-processing operation) and with disinfectants enriched with suitable sodium hypochlorite solution, 65 ppm of active chlorine. All equipments and accessories used in tomato puree processing were well cleaned and disinfected. The microbial safety is guaranteed by heat treatment $F_{(93;8,8)} = 32,8 \pm 2,7$, in water bath and $F_0 = 1,32 \pm 0,17$, in water spray retort. Reference value for tomato puree is $F_{(93;8,8)} > 20$ and $F_0 = 0,75$, minimal value.

Concluding the results, the experiments led to new results in field of the tomato preservation by heat processing. For the judgment of the alteration of lycopene and ascorbic acid concentration, it might be very important to determine optimal temperature, time and pressure of systems in order to get low loss of tomatoes nutrients.

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