Sudan University of Science and Technology

College of Graduate Studies

Effect of Pasteurization by Laser and Heat on the Cow's Milk Components

تأثير البسترة بالليزر والحرارة علي مكونات لبن الابقار

Thesis Submitted in Partial Fulfillment for the Requirement of M.Sc. Physics

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بسم الله الرحمن الرحيم

الاية

قال تعالي (وان لكم في الانعام لعبرة نسقيكم مما في بطونه من بين فرث ودم لبنا خالصا سائغا للشاربين)

سورة

النحل الاية (66)

الأهداء

* الي ذلك الرجل المضمخ بعبير الامنيات والمتقطر شهدا بحنان كبير ورعاية سامية الي الظل الوارف واليد السخية والنبراس المضئ والامل الواعد والقدوة الرشيده الي من علمني ان اكون كما يجب لك تقديرى واحترامي وحبي لك

** والدي العزيز متعك الله بالصحة**

* إلي تلك الحياة بشكلها الي الحديقة الوارفة وينبوع الحنان وساكبة الفرح وصانعة التضحيات الي

** امي العزيزة لك ودي وحبي**

الي اللذين يمرون في الحياه وكانما هم للقلوب فرح ويستقرون في الذاكره دونما عناء وكيفما يشاءون

الى زوجي العزيز واخواني الرائعين الي خالي الجميل محمود

وسأظل اطرق بابكم وابحث عن اشرعة جديدة تسمو بي عبر اجنحة الحياة

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الشكر والتقدير

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الشكر والتقدير ممزوج بكل الاحترام للمربي الفاضل

الدكتورعلي معروف

والاستاذة الفاضلة **بروفسور امل عمر بخيت** والي كل من مد الي يد العون

لكم الشكر الجزيل

Abstract

The main objective of this work is to investigate the cow's milk ingredients percentage after pasteurized using laser and heat treatment in order to compare them with untreated milk.

In this work, fresh cow milk sample weight (360 ml) were obtained from farms of Sudan University of Science and Technology, the sample was divided into three parts, the first part pasteurized by Nd: YAG laser with power of 50 watts for two minutes, the second part pasteurized by heating to a temperature of 72° C for 15 seconds the third part was control sample used as obtained. Moisture content, crude protein content, crude fat, ash content, total solid (TS) content, lactose content, pH of the milk samples and titratable acidity were analyzed method of for three samples.

The obtained results revealed that the ingredients percentage reduced in all heat pasteurized milk components compared to Laser pasteurized milk components.

مستخلص

الهدف الرئيسي لهذا البحث هو التحقق من مقادير مكونات حليب البقر بعد البسترة بواسطة الليزر وبالمعالجة الحرارية وذلك لمقارنتها بمقادير مكونات الحليب غير المبستر.

تم الحصول على عينة من حليب البقر وزنها(360 مل)من الحليب الطازج من مزارع جامعة السودان للعلوم والتكنولوجيا، قسمت العينة إلى ثلاثة أجزاء تمت بسترة الجزء الأول بواسطة ليزر Nd: YAGبقدرة مقدارها 50 وات لمدة، الجزء الثاني تمت بسترته بالتسخين لدرجة حرارة 72 درجة مئوية لمدة 15 ثانية وترك الجزء الثالث كما تم الحصول عليه كعينة ضابطة. محتوى الرطوبة و البروتين الخام و الدهن الخام و الرماد وإجمالي المواد المذابة و محتوى اللاكتوز والرقم الهيدروجيني والحموضة تم تحديدها وفقا لطريقة التحليل العيارية للعينات الثلاث.

أظهرت النتائج انخفاضا بسيط في مكونات الحليب المبستر الحرارة مقارنة مع مكونات الحليب المبستر بالليزر.

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Chapter One

Introduction

1.1 Introduction

Many scientific studies contradict the conventional wisdom that milk and dairy consumption help reduce osteoporotic fractures. Surprisingly, studies demonstrating that milk and dairy products actually fail to protect bones from fractures outnumber studies that prove otherwise. Even drinking milk from a young age does not protect against future fracture risk but actually increases it. Shattering the “savings account” calcium theory, Cumming and Klineberg report their study findings as follows Sara (2016).

This research studies the effect of laser on bacteria which is harmful and causes many illnesses (disease) for a human like diarrhea. One group of bacteria which includes 0157:H7 produce powerful toxin that damages the lining of small intestine, which can cause bloody diarrhea. Bacteria are found in milk to be contaminated it. Laser can use in sterilization of drinking milk from contaminated microbes (bacteria, viruses, germs, bugs) because photo-thermal interaction can occurs Sara (2016).

Most investigators agree that laser beam has very strong effect on the damage of bacteria (inactivation, inhabitation growth).Laser wavelength (UV, IR, visible) and exposure time can effect on the shape of decay Sara (2016).

We use power of ND:YAG laser (50) for tow mints constant and we found Before and after exposed the sample to power of radiation we saw the components of the milk(milkProtein-fat–lactose-ash-Total solids-acidity) , and it will be compared to the ultra –high temperature treated milk components.

Because of concerns that some potentially dangerous microorganisms may survive conventional heat pasteurization of milk and because the heat needed to sterilize milk affects marketability, the ability to efficiently cold pasteurized milk may become more desirable Sara (2016).

1.2 Research Problem:

The field of laser matter interaction may be with metal, tissue or food this research studies sterilization of milk by laser and the effect of laser on the milks components, and the sterilized milk by the ultra high temperature and the effect of it on milks components.

1.3 The objective of this thesis:

The main goal of this research is to compare the effect of laser in sterilizing milk with sterilizing the milk by heat.

Chapter Two

Literature Review

2.1 Introduction

Pulsed ultraviolet laser light used for the cold pasteurization of bovine milk, because of concerns that some potentially dangerous microorganisms may survive conventional heat pasteurization of milk and because the heat needed to sterilize milk affects marketability, the ability to efficiently cold pasteurized milk may become more desirable. In this pilot study, we investigated the use of pulsed ultraviolet (PUV) laser light to no thermally (cold) pasteurized bovine milk smith W.L, Laqunas and Cullor J.S (2002).

Dairy bulk tank milk was treated with UV light (248 nm) emitted from a pulsed excimer laser. The samples were then analyzed for surviving bacteria by spiral plate counting and sub culturing in Trypticase soy broth. Other bulk tank milk samples were inoculated with one of eight relevant milk bacterial species before being exposed to laser light. There was no growth observed for any of the plated or subcultured samples exposed to 25 J/cm2. One bacterial isolate was then used to inoculate milk to further investigate bactericidal laser light doses. Growth was observed for samples treated with an average of (0.3 to 6.6) J/cm^2 but not for those treated with 12.6 J/cm2. The results indicate that in principle, the bacterial content of milk can be adequately controlled by exposure to PUV laser light.

Their study was to monitor the effects of radiation emitted by a low energy laser on the growth of microorganisms in vitro from milk of cows with elevated SCC, microorganism diversification, and SCC after the lead treatment in vivo by the laser. Laser irradiated microorganism cultures exhibited a weaker incidence of environmental microorganisms, especially fungi and Streptococci sp. No laser light effect was noted on S. aureus culture development. Our data shows that after laser treatment the variety of micro-organism species immediately decreases 64.28% and this indicator remains unchanged after 21 days. 21 days after completion of the therapy course the SCC decreased 20.11%. 70 days after treatment the SCC increase compared to the 21 day period increased by 20.3%, which can be associated with factors unrelated to the method of therapy. It is advisable to treat increases in SCC with low intensity laser rays conditional to environmental mastitis causative agents. Moreover, since due to the effect of laser radiation certain irradiated micro-organism cultures become more susceptible to antibiotics, it is advisable to coordinate laser therapy with antibiotic therapy. Zilaitis et al (2008)

Considering that the quality of raw milk is a prerequisite condition to obtain a good quality probiotics yoghurt, our studies aimed the measurement of milk factors which can affect the multiplication of probiotics lactic acid bacteria (LABs) *Lactobacillus acidophilus* (LA-5. The probiotics strains Bifid bacterium BB-12 and *Lactobacillus acidophilus* LA-5-we used for trials- are tested probiotics by Christian Hansen company. We studied comparatively raw and pasteurized milk, their chemical composition and the correlations between the spontaneous microbial floras (NTG) found in milk samples and the impact of this flora on the multiplication of LABs. We investigated as well the effect milk proteins, added prebiotics (lactose, molasses) on pH and LAB development, the influence of NTG (number of total germs), NCS (somatic cells number) in raw milk before and after pasteurization, on lactic fermentations and LA-5(*Lactobacillus acidophilus*) Generally multiplication of LA-5 strains was reversely correlated with NTG values. There is a direct correlation between presence of prebiotics and probiotics bacteria activity. Eva Csutak(2009).

studied the possibility of using lasers in the sterilization of water and milk . Two types of diode laser has been used, the first one 2-watt 810 nm wavelength and the second 5 watt wavelength 1064 nm. Irradiation of samples was taken place from the physiological saline containing the *Escherichia coli* bacteria in three sizes (0.3,0.2, 0.1) cm^3 in test tubes of size (0.5) cm^3. Samples were irradiated at different periods of time for each laser. The results showed that after each irradiation we have obtained a highest kill of 100% of the bacteria by laser diode (2 W) through a period of 20 minutes. In the diode laser (5 W) a recording of the highest kill rate of 100% has been obtained in 1.5 min period).The conducted results of disinfecting water with the killing rate 100% have been applied in the sterilization of milk contaminated with bacteria under the same conditions in which 100% kill rate have been achieved for both lasers Naama et al (2011).

determined micro minerals in milk from farm and pasteurized cow, goat and camel; using inductively coupled Plasma-Optical Emission Spectrometry, This study covers raw fresh milk of cow, goat and camel (farm and pasture-reared), in addition to two brands of commercial milk samples, liquid milk of powder origin and drinking yoghurt samples. Camel milk showed a relatively lower pH range (6.15 - 6.46) compared cow, goat and commercial milk. The pH of drinking yoghurt was found (4.35 - 4.47).Microwave digestion, was selected followed by mineral analysis using Inductively Coupled Plasma-Optical Emission Spectrometry. Micro minerals; Cd, Cr, Cu, Fe, Mn and Pb, ranged from not detected to 23.4$\pm$0.52mg/L for Fe while Sr (0.32$\pm$0.005 – 2.51$\pm$0.043 mg/L) and Zn (1.58$\pm$0.01 – 8.91$\pm$0.14 mg/L) in all milk samples Elhardallou and El-naggar (2014).

2.2 Laser:

The word (laser) is an acronym derived from Light Amplification by Stimulated Emission of Radiation. The light emitted by laser is different from that produced by more conventional light sources. Laser is a device that generates or amplifies coherent radiation at frequencies in the infrared, visible or ultraviolet and other regions of the electromagnetic spectrum J. Michael Hollas – 2004

Lasers are distinguished from other light sources by their coherence. Spatial coherence is typically expressed through the output being an arrow beam which is diffraction-limited, often a so-called "pencil beam." Laser beams can be focused to very tiny spots, achieving a very high irradiance, or they can be launched into beams of very low divergence in order to concentrate their power at large distance.

Temporal (or longitudinal) coherence implies a polarized wave at a single frequency whose phase is correlated over a relatively large distance (the coherence length) along the beam. A beam produced by thermal or other incoherent light source has an instantaneous amplitude and phase which vary randomly with respect to time and position, and thus a very short coherence length (Florian Ion Petrescu – 2011).

Most so-called "single wavelength “lasers actually produce radiation in several modes having slightly different frequencies (wavelengths), often not in a single polarization, and although temporal coherence implies monochromaticity, there are even lasers that emit broad spectrum of light, or emit different wavelengths of light simultaneously. There are some lasers which are not single spatial mode and consequently their light beams diverge more than required by the diffraction limit. However all such devices are classified as "lasers" based on their method of producing that light: stimulated emission, lasers are employed in applications where light of the required spatial or temporal coherence could not be produced using simpler technologies (Gaurav Gupta - 2015).

2.3 Properties of laser:

Laser radiation shows as extremely high degree of monochromaticity ,coherence , directionality and brightness as compared to other no coherent light sources (MZ Khalid - 2016).

2.3.1Monochromaticity:

The monochromaticity of laser radiation is a unique property of laser light, results from the circumstance that light oscillation sets in at one resonance frequency of the optical cavity, and owing to the balance between gain and loss in CW operation the line width ΔvL of the oscillating mode is ultimately limited by quantum noise(Frank Träger – 2012).

2.3.2 Coherence:

The coherence of the laser radiation refers to the time period Δt in which the phase undergoes random changes, and the coherence length is a measure of the propagation distance over which the beam stays coherence (Wikipedia).

2.3.3 Directionality:

The directionality of the laser beam is due to the fact that the gain medium is placed inside an open optical resonat. (Frank Träger - 2012)

2.3.4 Brightness:

The brightness of laser radiation is closely related to the directionality and stems from the capability of a laser oscillator to emit a high optical power in a small solid angle of space (Frank Träger – 2012).

2.4 Elements of laser:

A laser generally requires three components for its operation:

(a) an active medium in the form of a laser rod, with energy levels that can be selectively populated;

(b) a pumping process to produce population inversion between some of these energy levels;

(c) a resonant cavity containing the active medium which serves to store the emitted radiation and provides feedback to maintain the coherence of the radiation. The main problem in designing a laser is to involve produces a sufficiently high population of atoms in the excited state. For this, many ingenious ways fully all have been evolved. The most common method of center excitation is by sending an intense beam of light from a flash lamp or a continuous source of light through the material in the form of a cylindrical rod or a container tube with a suitable gas. Only those materials which can be pumped to achieve population inversion are used to give laser radiation. The existence of states, whose mean life times are relatively long so as to help pile up considerable energy in the excited levels, is necessary. Long life time of a level and the sharpness of the spectrum lines usually go together, and so, the materials that can be best used to give laser radiation are crystals with sharp lines, and gases at low pressure. An important aspect of the laser operation involves the design of a resonator cavity to maximize the process of stimulated emission. Two carefully aligned mirrors, one having more than 99 percent reflectivity and the other having less reflectivity, are placed at either end of the cavity containing the laser rod and the flash lamp. The stimulated radiation multiplies by bouncing back and forth many times between the two mirrors and passing through the laser medium. And, when it exceeds a certain limit, the laser light comes out citation in the form of a narrow pencil beam through the semi-transparent mirror (LH Gami - 2014).

2.5 Laser construction:

A laser system is constructed from three main parts:

2.5.1 Pumping source:

It can provide energy to the laser system for example electrical discharge, flash lamp, light from another laser, chemical reactions and even explosive devices. The type of pumping source uses principally depends on the gain medium, and this also determines how the energy is transmitted to the medium (Dariush Honardoust – 2016).

2.5.2 Laser gain medium:

Also calls lasing medium results from stimulated emission of electronic or molecular transition from higher to lower energy state populated by a pump source (RH Omer - 2016)

2.5.3 The optical resonator or optical cavity:

In its simplest form is two parallel mirrors placed around the gain medium which provide feedback of the light. Cavity designed to internally reflect infrared, visible, ultra-violet. It contains gases, liquids and solids, Cavity materials can determine the wavelength of the output Figure (2.1) (K Ahmad Shazwi - 2008).

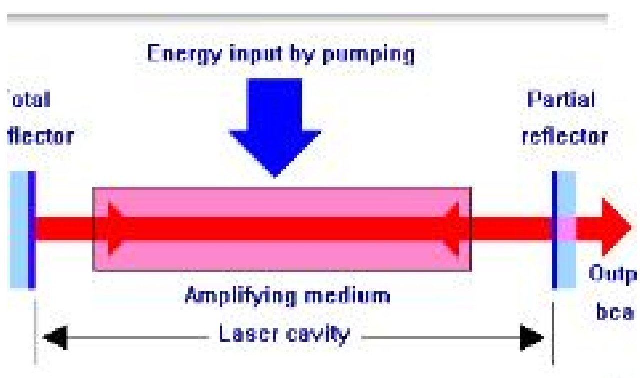

Figure (2.1) Elements of laser

2.6 Laser types:

The various laser types developed so far display a wide range of physical and operating parameters. Indeed, if lasers are characterized according to the physical state of the active material, we call them solid-state, liquid, or gas lasers. A rather special case is where the active material consists of free electrons at relativistic velocities passing through a spatially periodic magnetic field free electron lasers. If lasers are characterized by the wavelength of emitted radiation, one refers to infrared lasers, visible lasers, and ultraviolet (UV) and x-ray lasers (Orazio Svelto – 2010).

2.6.1 Gas lasers:

A gas laser contains atoms or molecules. Stimulated transitions occur in atoms between electronic states and in molecules between rotational, vibration, or electronic states. We describe various gas discharge lasers: helium–neon laser; metal vapor laser; argon ion laser; exciter laser; nitrogen laser; CO2 laser; optically pumped gas lasers (SI Almahal - 2016).

2.6.2 Solid state lasers:

We discuss solid state lasers that make use of electronic states of impurity ions in dielectric crystals or in glasses — other types of solid state lasers, namely semiconductor lasers that are based on electrons in energy bands of semiconductors. It consists the ruby laser, the titanium–sapphire laser, neodymium-doped YAG laser, of other neodymium lasers, and of other YAG lasers(SI Almahal - 2016).

?????Nd: YAG laser:

Nd:YAG lasers are optically pumped using a flashtube or laser diodes. These are one of the most common types of laser, and are used for many different applications. Nd: YAG lasers typically emit light with a wavelength of 1064 nm, in the infrared However; there are also transitions near 940, 1120, 1320, and 1440 nm. Nd: YAG lasers operate in both pulsed and continuous mode. Pulsed Nd: YAG lasers are typically operated in the so-called Q-switching mode Figure (2.2): An optical switch is inserted in the laser cavity waiting for a maximum population inversion in the neodymium ions before it opens. Then the light wave can run through the cavity, depopulating the excited laser medium at maximum population inversion. In this Q-switched mode, output powers of 250 megawatts and pulse durations of 10 to 25 nanoseconds have been achieved. The high-intensity pulses may be efficiently frequency doubled to generate laser light at 532 nm, or higher harmonics at 355, 266 and 213 nm Figure (2.3) (wikipedia).

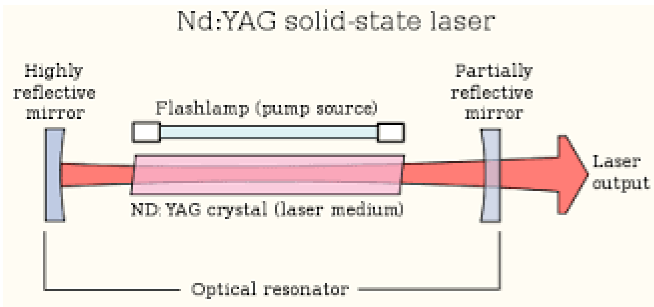

Figure (2.2) Elements of Nd:YAGlaser

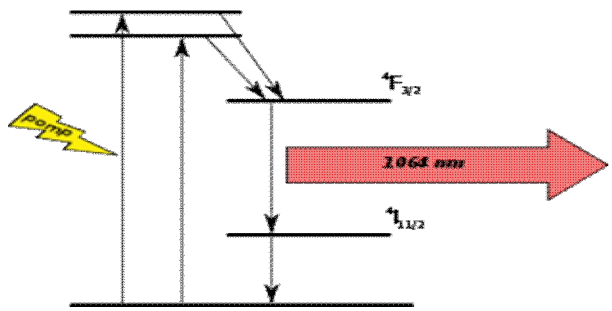

Figure (2.3) energy state of Nd: YAG laser

2.6.3 Semiconductor lasers:

Semiconductor lasers represent one of the most important class of lasers in use today, not only because of the large variety of direct applications in which they are involved, but also because they have found a widespread use as pumps for solid-state lasers.

These lasers will therefore be considered at some length . For the active medium, semiconductor lasers require a direct-gap material, so normal elemental semiconductors (e.g., Si or Ge) cannot be used.The majority of semiconductor laser materials are based on a combination of elements in the third group of the

Periodic Table (such as AI, Ga, In) and the fifth group (such as N, P, As, Sb) hence referred to as 1II-V compounds [4] (RH Omer - 2016).

2.6.4 Liquid Dye lasers

Liquid Dye Lasers use a solution of complex dye material as the active medium. The dyes are large organic molecules, with molecular weights of several hundred. The dye material is dissolved in an organic solvent, like methyl alcohol. Thus the active medium is a liquid. Dye lasers are the only types of liquid lasers which have reached a well developed status (U Brackmann - 2000).

One of the most important features that dye lasers offer is tenability, that is, the color of the output beam can be varied by adjusting the inter cavity tuning element and also by changing the type of the that is used. The monochromatic output of available dye lasers can be turned over a broad range, from the ultraviolet, to the near infrared. Liquid dye lasers that can be tuned to any visible wavelength, and to portions of the infrared and ultraviolet, are commercially available in both pulsed and continuous models. Dye lasers are chosen for applications, like spectroscopy, in which tenability is important (Halina Abramczyk – 2005).

2.7 Laser applications:

Lasers are employed over a wide range of applications from scientific research, biomedicine, and environmental sciences to industrial material process, microelectronics and entertainment.

Some applications are: Industrial application like cutting , welding ,drilling by using CO2 laser, ruby laser, argon ion laser , pulse N d:YAG laser. Medical application like phototherapy of eye , tissue surgery , using (CO2 laser, N d: YAG laser ,argon ion laser, and dye laser).Military applications include range finders and beam weapons , by (CO2 laser, N d:YAG laser, chemical laser, semiconductor laser). Other applications include Communication , information processing , super market scanners , printers , reading device for compact disc player , holography and spectroscopy(RH Omer - 2016).

2.7.1Scientific Research:

Lasers have opened new fields of investigation in science and technology. It has given physics a versatile tool for the study of interaction of light and matter. The powerful beam of laser has become an important tool for spectroscopic analysis. A laser system, known as microprobe, is used for exciting emission from solid samples for spectrographic analysis. In 1928, Prof. CV Raman discovered a new phenomenon, known as Raman Effect. By which molecular structures of different substances can be investigated by passing monochromatic light through them. He found that when light passes through a transparent substance, it is scattered and emerges with a change of frequency caused due to the vibration of molecules in the substance. This produces additional lines (known as Raman lines) in the scattered light spectrum. The discovery of laser is a great boon for recording the Raman spectra. The use of lasers has enabled recording of Raman lines within seconds, which otherwise would require long exposure times of or few hours using ordinary light sources. The analysis of Raman lines gives the fundamental properties of the substances. Similarly, lasers can also be used for analyzing liquids. A laser beam, when passed through a liquid, gives several colures (wavelengths) and the process is called fluorescence. The study of the fluorescence spectra thus obtained gives the properties of the liquids. Lasers offer attractive possibilities in terms of the exploration of molecular structure and determination of nature of chemical reactions. A laser beam can initiate and hasten a chemical reaction. Since different reactions require different wavelengths of light, a 'tunable' laser (i.e., a source whose wavelength can be altered as in radio tuning) is of immense help to a chemist. Tunable lasers, particularly dye lasers, now cover the entire visible spectrum and have revolutionized optical spectroscopy. In photochemistry, lasers with short duration pulses are highly useful for inducing and monitoring ultrafast chemical reactions more efficiently than by any conventional method. Laser also finds application in biological research. Using laser techniques, biological studies have been carried out in enzymes, proteins, cellular components and isolated cells, microorganisms, tissue culture, isolated physiological systems individual organs, etc. Using a ruby laser coupled with a microscope, single cells have been irradiated with laser beams focused on to a spot of the order of one micron to destroy individual chromosomes, thus making available a highly delicate instrument for genetic studies. It is also possible to produce laser beams as narrow as the diameter of a protein molecule and use it to alter genetic properties of living organisms (B Alberts - 2002).

2.8 Milk:

Milk is an indispensable food item and is considered as nature's perfect food for human beings as well as other animals. Mammals secretes milk for the nourishment of their young ones and milk of animals like cattle, buffalo, goat, sheep, camel, yak, llama, etc are being used as food for human beings (NZFSA, 2003).

2.8.1 Importance of Milk and Milk Products in Diet

Fluid milk is not only nature's food for a new born infant, but also a source for a whole range of dairy products consumed by mankind. Fluid milk is about 87% water and 13 % solids. The fat portion of the milk contains fat-soluble vitamins. The solids other than fat include proteins, carbohydrate, water-soluble vitamins and minerals. Milk products contain high quality proteins. The whey proteins constitute about 18% of the protein content of the milk. Casein, a protein found only in milk, contains all of the essential amino acids and accounts for 82 % of the total proteins in milk. Milk also contains calcium, phosphorus, magnesium, and potassium. The calcium found in milk is readily absorbed by the body; Vitamin D plays a role in calcium absorption and utilization. Milk is also a significant source of riboflavin (vitamin B_2), which helps promote healthy skin and eyes (Dairy Facts, 2003). Dairy products such as yogurts, cheeses and ice creams contain nutrients such as proteins, vitamins and minerals. Consumption of dairy products been associated with decreased risk of osteoporosis, hypertension, colon cancer, obesity and insulin resistance syndrome (IRS) (Weaver, 2003).

2.8.2 Nutritive value of milk

Milk is an important nutrition source for people around the world. Recently, much consideration has been given to milk quality, especially milk protein (Shi and Zubarev, 2010). Cow milk is comprised of approximately 3.3% protein, 4% fat, 87.1% water, 4.6% lactose, many essential vitamins (vitamin E and A), and is major source of calcium (Dissanayake, 2011). Milk is a major source of dietary energy, protein and fat, contributing on average 134 kcal of energy/capita per day, 8 g of protein/capita per day and 7.3 g of fat/capita per day (FAOSTAT, 2012).

2.8.3 Protein

Traditionally milk proteins have been divided in three crude groups, whey proteins, milk fat globular membrane (MFGM) proteins, and casein (Cunsolo *et al.*, 2011).Currently, milk protein and its products are highly researched because of the significant impact on health and high biological value (Santos and Lies, 2015). Milk proteins play an important role for growth factors, enzymes, immune system function, hormones, and antibodies (Korhonen and Gill, 2000). In addition, nutritional scientists found that using a higher amount of milk protein may buildup muscles, prevent bone breakdown, improve muscle movement, raise satiety, control blood sugar, and decrease the risk of some cancers (Karen *et al .*, 2013).

2.8.3.1 Casein

Casein (CN) is the principle protein and represents 80% of the cow milk protein. Casein is known as micellar. There are 5 kinds of casein micelles in milk that are different in moleculear composition but are similar in structure (alpha S_1 (αs1-CN), alpha S_2 (αs2-CN), beta (β-CN), kappa (κ-CN) and (γ- CN) casein). They typically have a molecular weight between 14 and 25 kDa (Heino, 2010).

2.8.3.2 Whey protein

Whey proteins are a specific group of proteins that have various biological, physiological and practical properties and are known as a rapid digested protein (Korhonen and Gill, 2000). Whey proteins have significant biological effect on the human immune and growth systems (Heino, 2010). Additionally, they have various effects in the human body such as being useful in human brain function and muscle protein synthesis (Santos *et al.,* 2012). Whey proteins contribute to 18-20 % of total milk proteins. The most important proteins in whey are bovine serum albumin (BSA) 10%, α-lactalbumin (α-LA) 20%, β-Lactoglobulin (β-LG) 50%, Casein, lactoferin (LF), immunoglobulins (Ig) 10% and glycomacropeptide (GMP) (Santos and Lies, 2015).

2.8.4 Fat

Milk fat is one of the most complex natural fats. It has a unique flavor and nutritional properties. The fat content in cow's milk is in the range of 3 to 6% (Lopez *et al.,* 2011). Milk lipids have the main responsibility for the textural properties of fat-based dairy products. The microstructure and textural properties of milk fat-based products are influenced by the crystallization of milk fat. Milk fat crystallization is affected by chemical and physical parameters, such as the fatty acid (FA) composition, presence of milk fat globules, temperature conditions and shear forces. The quality of the raw material and the parameters used during product manufacturing are fundamental for the crystallization process, thus for the final quality of the product (Fredrick *et al.,* 2011).

Milk fat globules are composed of a triacylglycerol (TAG) core surrounded by a complex membrane, the milk fat globule membrane (MFGM). MFGM accounts for 2-6% of the total mass of the fat globule. Approximately 90% of the MFGM is composed of polar lipids and proteins, whereas the remaining part includes glycoproteins, cholesterol, enzymes and other bioactive components (Dewettinck *et al.,* 2008). The core of the milk fat globule is mainly composed of TAGs which account for 98.3% (w/w) of the total composition; however, the remaining 1.7% includes several other components (Lopez *et al.,* 2011).

2.8.5 Lactose

Lactose is a disaccharide; Lactose is one of important energy sources and is sometimes referred to simply as milk sugar because it is the primary carbohydrate found in dairy products. It is the primary source of carbohydrates during mammal development. It constitutes 40% of energy consumed during the nursing period (Coultate, 2002). Lactose in milk has comparatively lower glycemic index compared to glucose or sucrose thereby making it suitable for diabetic people (Adolfson *et al.,* 2004). Chemically lactose is composed of one molecule each of glucose and galactose. Fig 1

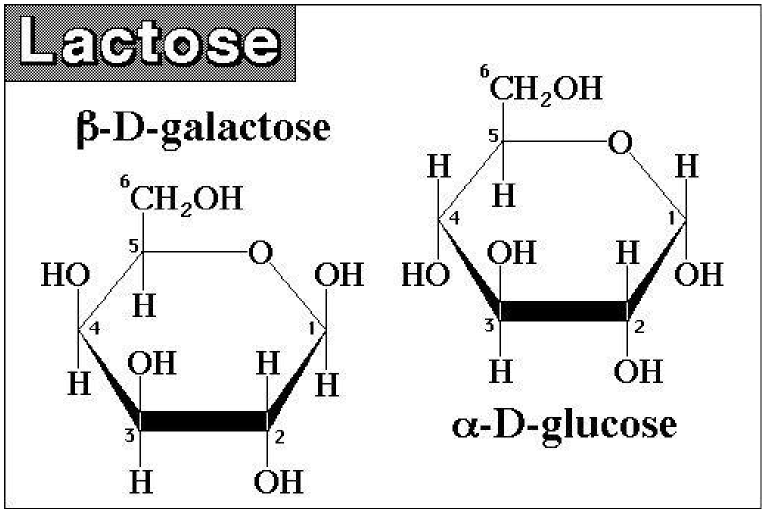

Figure 1: Molecular structure of lactose (Coultate, 2002).

2.8.6 Minerals and Vitamins

Minerals are essential elements swallowed through the diet. They are classified into three groups: macro minerals, micro minerals and trace elements. Macro minerals are sodium (Na), calcium (Ca), potassium (K), phosphorus (P), magnesium (Mg) and chloride (Cl); the micro minerals are iron (Fe), zinc (Zn), copper (Cu), fluorine (F), iodine (I), selenium (Se), manganese (Mn), chrome (Cr) and cobalt (Co); trace elements group include several minerals not essential for the organism such as bromine (Br), nickel (Ni), lithium (Li), silicon (Si) and tin (Sn). Minerals represent 4% of total human body mass and they are found in tissues, liquids, cells and organs. Minerals have structural, biochemical, catalytic and nutritional functions, thus they are fundamental for human health . Cow milk and dairy products are valuable sources of essential minerals (on average 10 to 20% of daily dietary intake) (Zamberlin *et al.*, 2012). Minerals represent a small portion of milk (about 8-9 g/L) and they occur in different chemical forms: inorganic ions and salts or as parts of proteins, nucleic acids, fats and carbohydrates (Summer *et al.*, 2009). The demand of dairy products and, among them, of fortified products, is increasing in the global market. The consumers require healthy food and thus they pay serious attention to the nutritional composition. Some studies argued that minerals have better functional effects when combined with other compounds such as vitamins, proteins and fatty acids (Soyeurt *et al.,* 2009).

2.8.7 Vitamins

Milk contains the water soluble vitamins thiamin (vitamin B_1), riboflavin (vitamin B_2), niacin (vitamin B_3), pantothenic acid (vitamin B_5), vitamin B_6 (pyridoxine), vitamin B_{12} (cobalamin), vitamin C, and folate. Milk is a good source of thiamin, riboflavin and vitamin B_{12}. Milk contains small amounts of niacin, pantothenic acid, vitamin B_6, vitamin C, and folate and is not considered a major source of these vitamins in the diet. Milk contains the fat soluble vitamins A, D, E, and K. The content level of fat soluble vitamins in dairy products depends on the fat content of the product. Reduced fat (2% fat), low fat (1% fat), and skim milk must be fortified with vitamin A to be nutritionally equivalent to whole milk. Vitamin A is necessary for maintenance of skeletal muscle and epithelial tissue as well as for normal immune function, vision, growth, and spermatogenesis (Tamime, 2009). Fortification of all milk with vitamin D is voluntary. Milk contains small amounts of vitamins E and K. Vitamin E is a lipid soluble cellular antioxidant having important roles in maintenance of cellular membranes, immunity, and reproduction (Adolfson *et al.,* 2004).

2.8.8 Enzyme

Indigenous milk enzymes are found in, or associated with various, casein micelles, milk fat globule membrane, milk serum or somatic cells and may originate from blood, the milk fat globule membrane (MFGM) or the cell cytoplasm. Important indigenous milk enzymes, e.g. plasmin, lipoprotein, lipase, alkaline, phosphatase and lactoperoxidase (Tamime, 2009).

2.9 **Pasteurization**:

Is process of heating a liquid by the purpose of getting rid of bacteria and germ**s**, and this is process that happens to the milk and the dairy of milk . in this process the milk is heated to a very high temperature to kill bacteria that exist in milk .in addition to this it also destroy many causes of disease that live in the milk ,so that it will be impossible for them to cause any more diseases .with taking in consideration that the productions should be stored in a safe place ,and should be used before it’s time is over .

There are of course advantages of pasteurization for instance killing germs which is the best advantage and disadvantages like killing some of the elements that live in milk.

Chapter Three
Materials and Methods

3.1 Materials:

3.1.1 Milk

Fresh milk samples weight (360ml) were obtained from Sudan University of Science and Technology farms in Bahri city and kept in ice container then transferred directly to the animal’s products department-National Food Research Center (NFRC).Chemicals and reagents used were obtained from the store of the NFRC, Ministry of Higher Education, and Sudan. All the chemicals and reagents were of analytical grades.

3.1.2 Devices:

3.2.1 Nd: YAG laser:

Nd: YAG lasers are optically pumped using a flashtube or laser diodes. These are one of the most common types of laser, and are used for many different applications.

Nd: YAG laser Figure (3.1) typically emit light with a wavelength of 1064 nm, and maximum output power 100 Watt, adjusted 1 Watt in step, the Nd: YAG laser mode lDORNIER med Tech Medilas 5100 fiber tom Glass I.

Figure.3.1. Nd: YAG laser

3.2.2 Magnetic stirrer:

The magnetic stirrer device Figure (3.3) is used to make homogenous solution by mixing the milk compound with Nd:YAG laser radiation .A rotation field of magnetic force is employed to induce variable speed a stirring action within either closed or opened vessels .The stirring is accomplished with the aid of small permanent magnets sealed in Pyrex glass. This device was manufactured by Scott science and healthcare limited it speed 60 to 1500 pm.themagneticstirrer is hot plate stirrer :Model L M S -1003 scott science U k.

Figure (3.3) Magnetic stirrer

3.2 Methods

Fresh cow milk samples were obtained from farms of Sudan University of Science and Technology, one of the samples pasteurized by Nd: YAG laser with power of 60 watts for two minutes and that based on previous studies, the second sample pasteurized by heating to a temperature of 72 ° C for 15 seconds the third sample was control sample as obtained.

3.2.1. Moisture content

The moisture content was determined according to the standard method of the Association of Official Analytical Chemists (AOAC, 2003).and it was as follow:

Principle:

The moisture content in a weighed sample is removed by heating the sample in an oven (under atmospheric pressure) at 105 ± 1Cº.Then, the difference in weight before and after drying is calculated as a percentage from the initial sample weight.

Procedure:

A sample of 5 ml will be weighed into a pre-dried and tarred dish. Then, the sample will be placed into an oven (Kat-NR.2851, Elektrohelios, Sweden) Figure.3.4 and left to dry at 105±1Cº until a constant weight is reached . After drying, the covered sample will be transferred to desiccators and cooled to room temperature before reweighing. Triplicate sample will be used.

Figure.3.4 Oven (Kat-NR.2851, Elektrohelios)

$$\text{Moisture content [\%]} = \frac{[m2-m3]}{[m2-m1]} \times 100$$

Where:

m1 = mass of dish + cover

m2 = mass of dish + cover + sample before drying

m3 = mass of dish + cover + sample after drying

3.2.2 Crude protein

The crude protein content will be determined in all samples by micro-Kjeldahl method using a copper sulphate or sodium sulphate catalyst according to the Official Method of the AOAC (2003).

Principle:

The principle of method consists of sample oxidation and conversion of nitrogen to ammonia, which reacts with the excess amount of sulphuric acid forming ammonium sulphate. The solution is made alkaline and the ammonia is distilled into a standard solution of boric acid (2%) to form the ammonia-boric acid complex, which is titrated against a standard solution of HCL (0.1N). Accordingly, the crude protein content is calculated by multiplying the total N % by 6.38 as a conversion factor for protein.

Procedure:

0.5 ml of sample will be accurately weighed and transferred together with 2-3 glass pellets, kjeldahl catalyst (No 33064, BDH, England) Figure (3.5) and 20ml concentrated sulphuric acid (No 18474420, Mark AG, Germany) into kjeldahl digestion flask. After that, the flask will be placed into a kjeldahl digestion unit (Tecator, Sweden) for about 3hours, until a colorless digest was obtained. Following, the flask was left to cool to room temperature. The distillation of ammonia was carried out in 30 ml boric acid (2 %) by using 40 ml distilled water and 60 ml sodium hydroxide solution (33 %). Finally, the distillate was titrated with standard solution of 0.1N HCL in the presence of 2-3 drops of indicator (Bromocreasol green and methyl red) until a brown reddish color was observed.

Figure (3.5) kjeldahl digestion unit

Calculation:

$$\text{Nitrogen (\%)} = T \times 0.1 \times 0.014 \times 100/ \text{Weight of sample}$$

$$\text{Protein (\%)} = \text{Nitrogen \%} \times 6.38$$

$$\text{Protein conversion factor} = 6.38\%$$

3.2.3 Fat content

The crude fat in the product will be determined according to the standard analytical method of A.O.A.C, (2003).

Ten ml sulfuric acid (density 1.815 gm/ml at 20ºC) will be poured into a clean Gerber tube, followed by the addition 10 ml of sample then 1 ml of amyl alcohol will be added to the tube followed by addition of distilled water. The tubes will be then thoroughly mixed till no white particles were see, centrifuged at 1100 revolution per minute (rpm) and transferred to a water bath at 65ºC for 3 minutes. The columns of the fat will be then recorded immediately

3. 2.4 Ash content

The standard analytical method of A.O.A.C, (2003) will be used for determination of ash content in the samples.

Principle:

The inorganic materials which are varying in concentration and composition are customary determined as a residue after being ignited at a specified heat degree.

Procedure:

Two ml of the sample will be weighed into a pre- heated, cooled weighed and tarred porcelain crucible. Before ashing, the sample was pre-washed on an electrical pre-asher and placed into a muffle furnace (Carbolite, Sheffeild, England) at 525 to 600 C °until a constant weight was obtained. The weight of the residue after ashing was defined as ash content and expressed as a percentage based on the dry matter content in the ground sample.

Calculation:

$$\textbf{Ash content} = W_1 / W_0 \times 100$$

Where

W_1 = Weight of ash

W_0 = Weight of sample

3.2.5 Total solids (TS)

Total solid (TS) content was determined according to AOAC (1995).A clean aluminum dishes were dried at 105ºC for 3hrs.Five grams of the sample were weighed in dry clean flat bottomed aluminum dish and heated on a steam bath for 15 minutes. The dishes were placed into a forced draft oven at 100ºC for 3 hrs. Then cooled in a desiccators and weighed quick .Weighing was repeated until the difference between the two reading was <0.1mg.The total solids (T.S.) content were calculate as follows:

$$\text{T.S.}(\%) = \mathbf{w}1/\mathbf{w}2 \times 100$$

Where:

W_1 =Weight of sample after drying

W_2 =Weight of sample before drying

3.2.6 Lactose content

The lactose content determined by An throne Method (Richard, 1959).One ml milk will be pipette in a 500 milliliters volumetric flask and diluted to 500 milliliters was transferred in a boiling test tube (in Triplicate) the samples were placed in ice bath, and shacked while adding 10 ml of ice cod an throne reagent, the tubes contents were mixed and then placed in a boiling water bath for 6min , then transferred back to the ice bath for 30 min ,the optical density of the colored solution was then read at 625min. A blank consisting of distilled water 0.5 milliners and anthron reagent andstandard containing 100mg/ml of lactose and anthron reagent were included in each batch of analysis. The percentage of lactose was then calculated using the following formula:

Lactose content = <u>O.D of sample- O.D of blank</u> X 4.75 =g/1000ml

S.D of stander-O.D of blank

OD: Optical density.

S: Sample.

SD: stander.

B: blank.

3.2.7pH-value

The pH of the milk samples was measured by using a recalibrated pH meter model (HI 8521 microprocessor bench pH/ MV/ C° meter) Figure (3.6). This will be calibrated with two standard buffers (PH6.8 and 4.0).

Figure (3.6) pH meter

3.2.8 Titratable acidity

This test will be carried out according to method described by A.O.A.C (2003).Ten grams of samples were weighed in to a small beaker, the sample was mixed well, 2-3 drops of phenolphthalein were added, and the sample was titrated against 0.1N NaOH till a faint pink color. The titration figure divided by ten to get the percentage of lactic acid (1 millitres of 0.1N NaOH sodium hydroxide = 0.009gm of lactic acid).

Chapter Four

Results and Discussion

4.1 Results:

The effect of the irradiation of Nd: YAG laser and heat treatment (72°c/15 s) on the milk components shown in table 4.1.

The statistical analysis (Table 1) showed that no significant ($P<0.05$) variations were found in the chemical composition of the milk samples in all treatments. However, slight decrease in the, moisture content of milk samples pasteurized at 72 ˚C/15 s was observed while the higher moisture was in the raw milk sample. The raw milk samples showed increase in fat content. Almost the protein contents of the milk samples treated with lazer and the raw one is the sample while slight decrease in the protein content was observed in the pasteurized milk sample with 72 ˚C/15 s.

The ash contents of the milk samples showed the same trend of the protein. The lactose was not found to be significantly different in all the milk samples. Therefore the high total solids were found in the raw milk samples while the lower one was in the heat treated samples.

The pH and the acidity of the milk samples were not significantly different in all treatments

Table (4.1) and figure (4.1) Effect of pasteurization by using laser and heat treatment on the cow milk components.

Parameters %	Pasteurize milk by heat at72°c for 15sec	Pasteurize milk By Laser for2min	Control Sample	Level of significant
Moisture	88.33±0.44	88.69±0.055	88.58±0.13	NS
Fat	4.03±0.20	4.10±0.20	4.16±0.15	NS
Protein	3.13±0.15	3.20±0.10	3.23±0.15	NS
Ash	0.83±0.03	0.84±0.026	0.84±0.025	NS
Lactose	3.06±0.11	3.16±0.115	3.16±0.115	NS
Total solids	11.06±0.11	11.30±0.55	11.41±0.136	NS
pH	6.16±0.02	6.20±0.005	6.33±0.047	NS
Acidity(lactic acid)	0.19±0.005	0.20±0.000	0.19±0.000	NS

NS means Not Significantly ($P<0.05$) different

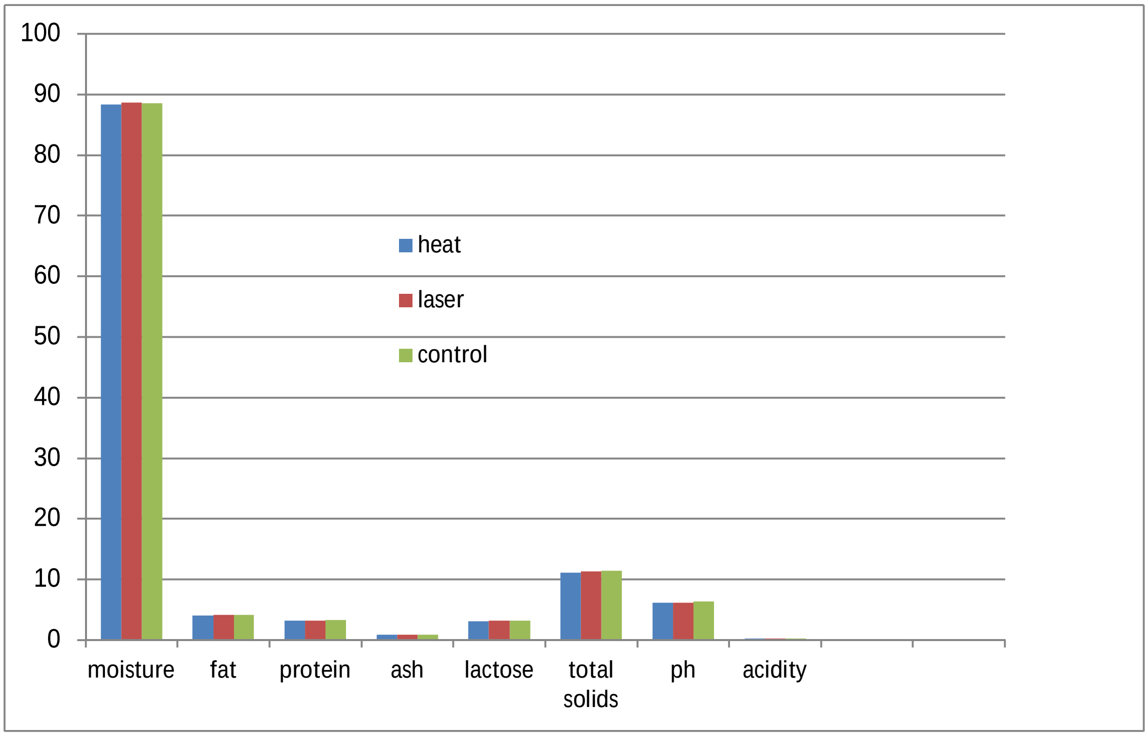

Figure 4.1Comparative of characteristics of tow pasteurized samples (heat & laser) according to control sample.

4.2 Discussion

The summary of the three experimental milk samples listed in table one , there were no statistical differences in basic compositions among the pH and acidity (lactic acid) for the milk pasteurized by laser and heat.

while the moisture increase up to 88.6 (May be because a tarred dish is not dry) and dropped to 88.4 for the milk pasteurized by heat .

But the protein , fat ,ash and lactose remain the same in the milk pasteurized by laser and dropped down in the milk pasteurized by heat .

Analyzing the data shows there is no significant difference between the milk components that pasteurized by laser and the one pasteurized by heat.

4.3 Conclusion:

In conclusion, to save the value of the most milk components, it was found the milk that pasteurized by laser better in protein and fat than the milk pasteurized by heat .The irradiation process leads to the pasteurized the milk of milk. This technique keeps the protein and fat of the milk. In the future further studies could possibly be done.

4. Recommendations:

- Based on the results of this research, it s recommended considering the use on ND: YAG laser on the dairy milk pasteurization.

Additional research is needed to study the effect of pasteurizing milk by another laser and the affect on milk vitamins

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