Non-thermal food preservation techniques: Scope and future perspectives

Remya S. & Bindu J.
Fishing Processing Division, ICAR- CIFT, Cochin
remya03cof@gmail.com

Non-thermal preservation of food

- Conventional thermal processing results in some undesirable changes in food, such as loss of nutritional components that are temperature-sensitive, change in the texture of food due to heat, and changes in the organoleptic characteristics of food.
- Destruction of nutritional components of food and the consumer demand for convenient, high quality, and minimally processed food products are the two key drivers towards innovation of the non-destructive/non-thermal food technologies.
- Non-thermal food processing simply refers to methods where the food materials receive microbiological inactivation without the direct application of heat.
- They are relatively young technologies, which use mechanisms other than conventional heating to reduce or eliminate microorganisms. Hence it offers an alternative to conventional thermal processing.

Categorization of non-thermal preservation techniques (Pivarnik and Worobo, 2014)

- High-pressure processing
- Light (ultraviolet, pulsed light)
- Ionizing radiation (gamma irradiation, electron beam)
- Gases (ozone, chlorine dioxide, cold plasma)

- There are other techniques like Pulsed Electric Field (PEF) processing, Ultrasound processing, Dense phase carbon dioxide (DPCD) processing etc.

1. High pressure processing

- High Pressure Processing is also known as high hydrostatic pressure (HHP) or ultra-high pressure (UHL) processing.
- It is a non-thermal, cold pasteurization technique, which generally consists of subjecting food, previously sealed in flexible and water-resistant packaging, to a high level of

hydrostatic pressure (pressure transmitted by water) up to 600 MPa / 87,000 psi for a few seconds to a few minutes (1 – 20 min).

- HHP utilizes a very common medium, i.e., water, to apply the pressure on the product to be treated.
- HHP transmits isostatic pressure (100–1000 MPa) instantly to product at low temperature and might have comparable preservation effect as thermal processing through inactivating undesirable microorganisms and enzymes.
- An HPP unit consists of a pressure compartment in which food is kept and water is introduced into the chamber. Food is then pressurized using this water.

Preservative action of high hydrostatic pressure (HHP)

- HPP compromises cellular functions such as DNA replication, transcription, translation (≤100 P)
- At higher pressures, microorganisms start suffering lethal injuries due to loss of cell membrane integrity and protein functionality.
- The most sensitive to pressures are moulds, yeast and parasites.
- Inactivation of common bacteria requires higher pressure (300-600 MPa).
- The most baro-sensitive are bacterial spores that were found to survive pressures up to 1200 MPa at room temperature.
- HHP can bring about a significant decimal decrease in the population of pathogenic Gram-negative bacteria, Gram-positive bacteria, yeast, and mould and helps in food preservation for a longer duration.
- The reduction in microbial load depends on the pressure and temperature during treatment. It also largely depends on the type of food processed.

Effect of high-pressure processing on food quality

- Food, when subjected to HHP treatment, undergoes high pressure for a short duration of time.
- The pressure applied to food during treatment is in the range of 200–700 MPa.
- The quality in terms of nutritional components, sensory, and texture of HHP-processed food is excellent since the food is exposed to treatment conditions for a very short period of time. Hence, HHP-treated food shows fresh-like attributes.

Merits

- HPP technology can be applied in processing foodstuffs that are either solid or liquid.
- The technology involves less-to-no utilization of food preserving agents & relatively low amounts of energy.
- Once the operational pressure has been attained, there is usually no extra energy needed to uphold the pressure.
- Compared to the conventional heat-utilizing technologies, HPP does not require supplementary energy to cool the food product beyond the estimated treatment period.
- The transmission and pressurization fluid (water) can be reused with zero-emission of wastes and hence it can be considered as one among the eco-friendly types of non-thermal food processing techniques compared to thermal pasteurization.
- Moreover, HPP can retain the taste of food, its nutrient composition and elongate its shelf-life. Thus, the spoilage rate can be decreased, which can help raise the economic value of the food commodity.

Major applications in seafood

1. Post pack lethality intervention for RTE seafood
   - *Cold post-packaging pasteurization*: For shelf-life extension, keeping freshness, maintaining higher sensorial qualities, functional properties and improving food safety.
2. Low pressure process application
   - *Mollusc shucking*: In HPP, the muscle, which is responsible for closing the shell, will not be able to contract and the oyster will open. This exposes the meat for easy extraction, resulting in a significant yield increase.
   - *Crustacean meat extraction*: In HPP, meat of crustaceans such as lobster or king crab will contract and detach from the shell, facilitating extraction with yield of almost 100 %.

Studies at ICAR-CIFT, Kochi

- Ginson et. al. (2015) studied the effect of high-pressure treatment (250 MPa for 6 min at 25 °C) on microbiological quality of Indian white prawn (*Fenneropenaeus indicus*) during chilled storage. All microbes were reduced significantly after high pressure treatment and there was significant difference in microbial quality of control and high pressure treated samples in the entire duration of chilled storage. Delayed growth of Enterobacteriaceae and H2S producing bacteria was observed in HP treated samples.

- Kunnath et. al. (2020) reported that synergistic effect of high pressure and microbial transglutaminase (MTGase) could enhance the textural and functional properties of fish gels, when compared with the conventional cooking. A high press 250 MP given to pink perch mince samples added with and without MTGase enzyme, for a holding 12 mi 25 °C 30 mi treatments. MTGase enzyme along with pressure treatment enhanced the conformational stability and produce stronger networks through the formation of non sulfide bonds between proteins and setting reinforced these networks.
- Devatkal et. al. (2015) employed high- (300 P 5 mi) - thermal post-processing intervention to improve the shelf life and quality of cooked refrigerated chicken nuggets. High-pressure treatment and pomegranate peel extract did not influence significantly the colour and textural properties of cooked chicken nuggets. Th ($p < 0.05$) - treated nuggets. Microstructural studies revealed shrinkage in the structure and loosening of the dense network of meat emulsion due to high-pressure treatment. Pressure treatment resulted in a reduction of 2–3 $\log_{10}$ cfu/g in total plate count and *Enterobacteriaceae* count.
- Kundukulangara Pulissery et. al. (2021) compared the textural and nutritional profile of high pressure and minimally processed pineapple. Changes in the pineapple quality in terms of texture, colour, total flavonoids, total polyphenols, vitamin C and sensory properties were investigated within the domain of 100-300 MPa and 5-20 min. On the basis of microbial quality and sensory assessment, high pressure treatment at 300 MPa for 10 min was found to be suitable for preserving the quality of pineapple up to 16th day in refrigeration condition.
- Ginson et. al. (2020) investigated the piezotolerance and diversity indices of microflora of Indian white prawn (Fenneropenaeus indicus) after high pressure (HP)-treatment. *Arthrobacter spp., Listeria grayi* and *Corynebacterium spp*. were the most piezo tolerant bacteria in HP-treated samples. The apparent reduction of microflora with pressure level was clearly evident from the diversity indices. A diminished piezotolerance of Gram-negative spoilage bacteria was also observed.

Limitations of HHP processing

- During processing, the organoleptic characteristic of HHP-treated food can be changed. This can be attributed to the ability of HPP to destabilize functional proteinaceous macromolecules, such as enzymes, by ionic and hydrophobic–hydrophobic interactions.
- HPP can accelerate lipid oxidation of treated seafoods during storage. This is caused by the release of inorganic transition metal ions from their respective compounds during the HPP process.
- HPP (>200)
- HPP can induce the formation of formaldehyde, which induces protein crosslinking, thus causing an increase in the hardiness of the treated fish. This is a drawback when HPP is employed for seafood treatment.
- *Types of food and HHP processing*
  - Foods with entrapped air or with insufficient low moisture content will be crushed or compacted under high pressure.
  - HHP is not suitable for
    - Solid foods with air included (Bread and cakes & Mousse)
    - Packaged foods in completely rigid packaging (Glass packaging & Canned foods)
    - Foods with a very low water content (Spices, Dry fruits &Powders)

2. Pulsed electric field (PEF) processing

- PEF is an efficient non-thermal food processing technique using short, high voltage pulses.
- It is used for inactivation of spoilage and pathogenic microorganisms in various food products. Electric pulses are applied for destroying harmful bacteria in food.
- Microbial inactivation is achieved by dielectric breakdown of the bacterial membranes
- Food material is placed between electrodes. The field intensity is typically 20–80 kV cm^{-1}) and the exposure time is a few milliseconds or nanoseconds.
- It enhances the shelf life of the food without quality loss.
- The PEF mechanism is called ***electroporation***. Very short electric pulses of high voltage are applied to the food. Small pores are formed in the cell membrane of the food by the electric pulses without damaging the cell compounds, such as vitamins.
- Pulsed electric field is generally used for liquid food or semi-solid food that can flow easily.

PEF device

- A typical PEF device consists of a food treatment chamber, a control system, and a pulse generator.
- The food is kept in the treatment chamber in between two electrodes generally made of stainless steel.

Applications of PEF in fisheries field

- PEF improves water holding properties of fish (submitting the fish muscle to PEF made its structure more porous)
- PEF technology improves extractive effectiveness to obtain protein from mussel (Improved extraction yield of protein)
- It can be used as a pre-treatment for drying
- PEF can be used to valorize by-products from fish processing industries.
- High-intensity PEF has been identified as an improved a method to extract calcium & chondroitin sulphate from fishbone.
- PEF has been tried for extraction of collagen from fish waste.
- PEF enzymatic-assisted extraction has been used for isolation of the abalone viscera protein.
- PEF can be used as a pre-treatment for fish waste for enhancing the yield of the extraction process.

Advantages of PEF processing

- PEF processing maintains the physical, organoleptic and functional characteristics of the final product, i.e., causing minimal changes in the flavour, vitamins, and other nutrients.
- It controls the presence of microorganisms in foods in a fast and homogeneous way
- Reduced process time
- Low energy consumption
- Continuity of the process
- Efficient and eco-friendly method
- Extended shelf life of the food product

Limitations of PEF processing

- The high initial capital investment is the main barrier that limits the application of PEF in the fish processing industry at this moment.
- PEF is a continuous processing method that may not be suitable for solid food products that cannot be pumped. Therefore, the conveyor is important to include in the design of the machine.
- In addition, inefficiency of this technique against the reduction of naturally occurring enzymes in the fish is another shortcoming of this emerging technology.
- The electrical conductivity of the product is a crucial parameter that limits the application of PEF to materials with moderate conductivity. PEF processing is limited to food products with low electrical conductivity and no air bubbles.

3. Irradiation/Radiation processing

- Refers to the process by which an object is exposed to radiation (A deliberate exposure to radiation)
- There are two forms of radiation: Ionizing radiation (IR) and non-ionizing radiation (NIR)
- IR includes high-energy electron beam, X- γ -rays.
- IR leads to the production of charged particles or ions in material it comes in contacts with.
- Irradiation is a process of applying low levels of ionizing radiation to food material to sterilize or extend its shelf life.
- Radiation inactivates food spoilage organisms, including bacteria, moulds, and yeasts.
- It is effective in lengthening the shelf-life of fresh fruits and vegetables by controlling the normal biological changes associated with ripening, maturation, sprouting, and finally aging.
- Radiation also destroys disease-causing organisms, including parasitic worms and insect pests, that damage food in storage.
- Irradiation is harmful or noxious to humans. However, the dose for seafood pre-treatment is low, therefore making it safe for consumption.
- Food irradiated under approved conditions does not become radioactive.

Two approaches to irradiation

1. Use of radioactive isotopes, such as Caesium or Cobalt: Isotopes produce penetrating gamma rays and require expensive facilities with heavy shielding, because the radiation is always on and could pose a hazard to workers.
2. Electrically generated radiation, such as X-rays or electron beams: Electrically generated radiation has less penetration strength and so is only useful for surface sterilization or on thin products. However, it is safer and less expensive to use, because it is turned on and off as needed and does not require shielding.

Agri-food applications of irradiation

Radicidation and Radurization: Refer to these applications of less than 10 kGy doses.

- Radurization: Application of an ionization dose sufficient to preserve the quality of food by ensuring a substantial reduction in the number of spoilage bacteria.
- Radicidation: Application to the food of a dose of ionization sufficient to reduce the specific number of viable pathogenic bacteria to a level such that they are not detectable by any known method. This term also applies to the destruction of specific parasites.

Radappertization: Application of high dose (10 to 60 kGy) of ionization to food in order to reduce the number and/or activity of living microorganisms so that none (except viruses) is detectable by any recognized method. Such radio-sanitized products can then be stored for up to 2 years at room temperature in sealed plastic packaging.

Table 1: Dose requirement in various applications of food irradiation

D **L**	**D**	**A**
	<1	I , I , ,
	1–10	x , , q

H	>10	H z , , z q

Labelling requirements

- For ionizing radiation, information concerning its application to the food and/or one of the ingredients of the food is mandatory.
- “ z ” , “ z ”, “ ” x “ ”, x H , the use of radura symbol is voluntary.

Merits

- It is among the non-thermal preservation methods with minimum effect on the quality, taste, appearance, and texture of foods.
- Ionizing radiation acceptability elevates as the consumer desire for minimally processed and yet safe food increases.
- The effectiveness of ionizing radiation is not only against destroying microbial entities and inhibiting pathogenic/spoilage bacteria; but also inhibiting insects, mites, and pests.
- The application of ionizing radiation has been shown as an alternative technique to detoxify aflatoxin present in foodstuffs.
- The processing time of ionizing radiation is reasonably less, considered as eco-friendly as well as leaving no chemicals/residue.

Demerits

- Excess accumulation as a result of constant exposure to irradiation is a major threat for the processors or workers.
- Some amino acids can be cleaved by high-dose irradiation, thereby changing the flavour and aroma of foods.
- Lipid oxidation is enhanced in the irradiated product since irradiation can accelerate auto-oxidation of lipids, producing hydroperoxides and off-flavours in food, especially seafoods rich in polyunsaturated fatty acids.

Studies at ICAR-CIFT, Kochi

- Annamalai et. al. (2020) assessed the effect of electron beam irradiation ((0, 2.5, 5.0, 7.5 and 10 kGy) on the biochemical, microbiological and sensory quality of vacuum packed headless *Litopenaeus vannamei* during chilled storage (2 °C). There is a significant ($p<0.05$) reduction in *Brochothrix thermosphacta* and *lactobacillus* count in the irradiated sample. Based on the microbial and sensory analysis control had a shelf life up to 12^{th} day. However, electron beam irradiated sample had extended shelf life of 15-23 days with respect to dose level.

4. Ultraviolet (UV) Radiation

- A very economical non-thermal technology
- Non-heat technique for decontamination for improving both the shelf-life and safety of foodstuff.
- It is basically used to reduce the microbial load on the surface of food materials that are indirectly exposed to radiation, because of its low depth of penetration.
- UV radiation is a form of energy considered to be non-ionizing radiation having in general germicidal properties at wavelengths in the range of 200–280 nm (usually termed UV-C).
- UV irradiation has demonstrated to be effective not only in reducing microbial load but also inactivating enzymes activity in plant products.

Effect UV-C on microbes

- When food is exposed to UV-C, with 200–280 nm, these short wavelengths are absorbed by the microbial cell nucleic acids.
- These absorbed photons cause the breakage of the bond and interlinking between thymine and pyrimidine of different strands and the formation of dimers of pyrimidine.
- These dimers (Photo products) prevent DNA transcription and translation, thus leading to the malfunctioning of the genetic material, which causes microbial cell death.
- In principle, the UV radiation operates by destroying the genetic constituent of the pathogen to prevent division, multiplication and subsequently hinder its propagation.
- Usually, different kinds of food products require different doses of UV radiation (termed as UV-inactivation dose measured in mJ/cm^2) to inactivate different kinds of pathogens.

UV-inactivation dose (mJ/cm²)

- Bacteria: 1–10
- Yeast: 2–8
- Fungus: 20–200
- Protozoa: 100–150
- Algae: 300–400

Factors affecting the efficiency of UV-C radiation

- The source and dose of the UV radiation
- The duration by which the product is exposed
- The nature of the foodstuff
- The alignment of the apparatus
- The nature of the microbe

Applications in the fisheries sector

- For food products, UV-C light technology application has been mostly confined to liquids and free-flowing foods.
- UV light is used in the fish industry to decrease the microbial load and increase the shelf life of fish, reduce the microbiological load in fish meal, disinfect working surfaces, and to sterilize the water in aquaculture and wastewater facilities.
- However, to achieve a more effective reduction in bacterial load, the studies indicate that UV light should not be used as a stand-alone strategy, but integrated with other technologies.

Merits

- The lethality effects of UV radiation against microbes are higher compared to the conventional chemical agents, for example, hydrogen peroxide and chlorine.
- Moreover, UV radiation is easy to utilize (user friendly) and cost-efficient.
- It has minimal effects on the quality of foods as it enhances sensory features such as taste for certain foods.
- It prevents recontamination as it can be applied in already packed food products.
- It is environmentally friendly.
- It can be used not only for liquid foods, but also for solid ones.

- Its processing time described as shorter and it also exhibits outstanding permeation capabilities to foodstuffs.

Limitations

- Accelerated senescence and surface discoloration in seafood can occur and deteriorate the treated seafood.
- UV radiation can induce oxidation of lipids in treated seafoods since hydrogen peroxide, superoxide radicals, and lipid radicals are indirectly formed by UV light.
- Peroxide created during extended UV light treatment can diminish the pigments and the fat-soluble vitamins.
- Cross-linking and fragmentation of protein, carbohydrate cross-linking, and peroxidation of unsaturated fatty acids in ultraviolet-treated seafood can be induced by superoxide radicals.
- Protein, aromatic amino acids, and enzymes are denatured by UV radiation, which could affect the composition of seafood.
- Consumers seem to be worried about the fact that UV radiation might be leading to radioactive materials in foods, which may subject them to serious health issues.
- Additionally, huge investment requirement is also a limiting factor for achieving the complete feasibility of the UV radiation process.

5. Pulsed Light (PL) Preservation

- Pulsed light (PL) is an alternative technique to continuous ultraviolet treatment for solid and liquid foods.
- PL consists of successive repetition of high-power pulses of light/short time high-peak pulses of broad-spectrum white light.
- Comparatively, PL has a thousand times strength greater than the normal UV light which is quite continuous.
- Pulsed xenon UV uses the full spectrum of ultraviolet light to disperse germ-killing energy.
- The light spectrum includes wavelengths from 180 to 1100 nm with a considerable amount of light in the short-wave UV spectrum.
- Similar to other non-thermal food processing technologies, PL also has potential in the inactivation or elimination of microbes in food.

- Specific examples of foods processed by PL include fish, vegetables, fruits, and meat.
- PL can be used alongside other novel technologies as a hurdle in the inactivation of microbes on the surfaces of foods.

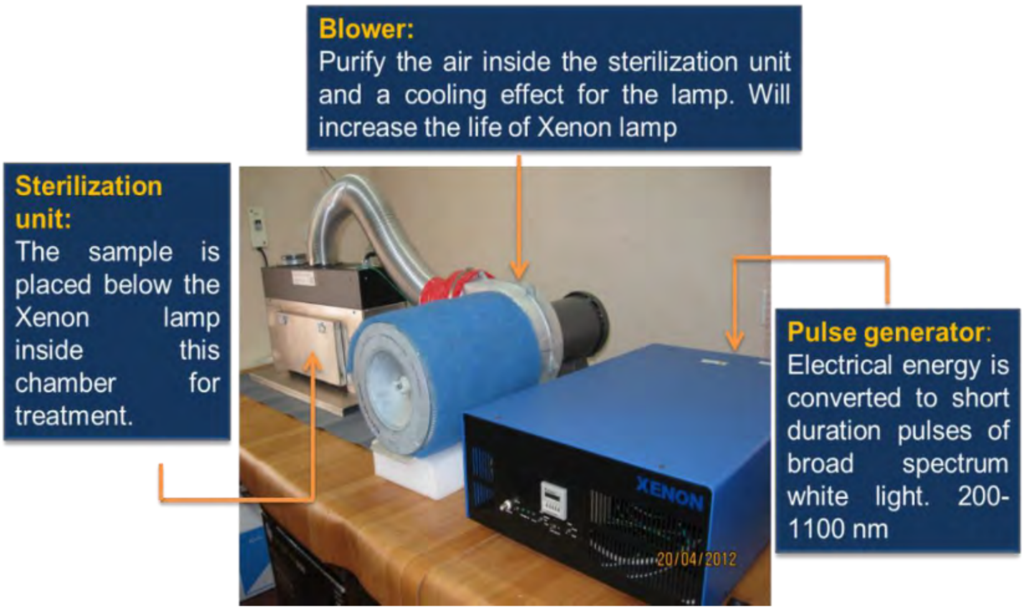

Figure 1: Pulsed Light Equipment of CIFT

Studies at ICAR-CIFT, Kochi

- Ananthanarayanan et. al. (2019) studied effect of pulsed light (PL) treatment on the shelf-life extension of yellowfin tuna (*Thunnus albacares*) steaks stored at 2±1 °C. Tuna steaks of 1 cm thickness weighing 80 g packed in 300 gauge cast polypropylene pouches were subjected to PL treatment using Xenon pulse light machine RC-847. Shelf-life studies were carried out in terms of reduction of aerobic flora as inferred from the total plate count (TPC) and the psychrophilic count. An overall extension of 13 days of shelf life was achieved for PL treated samples.

Merits

- PL serves as a rapid disinfection food processing technology.
- In addition, it exhibits much less damage to the nutritional content of foodstuffs that it has been applied to.
- PL is also shown to ensure microbial inactivation while at the same time retaining the

- The technology boasts a huge advantage compared to UV radiation by exhibiting an outstandingly short time energy transmission.
- Furthermore, besides the fact that PL exhibits a substantial reduction of bacteria in an exceptionally short time; it has huge adaptability, and is eco-friendly.
- Consequently, after PL application, the threat due to food-inherent disease-causing microorganisms is decreased; the shelf-life of foods increased as well as a promised enhanced economic return especially during the transportation period.
- In addition to that, PL has demonstrated promising results in the prevention of contamination of packaged products; the treatment is known to be applied even when the food is within the packages.

Demerits

- Similar to UV technology, the pulsed light comes with huge capital/costs in order to achieve successful investment.
- Pulsed light has been shown as not suitable for application in foods that are opaque and irregularly shaped, as they can be potential habitats for bacteria proliferation.
- , x P “ ” products and in the end affect the effectiveness of the bacterial destruction.

6. Ultrasound (US) processing

- US is a compressional wave with a frequency of over 20 kHz.
- US is sound wave bearing certain frequency that is more than the normal human hearing frequency, which is more than 20 kHz.
- The frequency of US used in the food industry for microbial inactivation ranges from 20 kHz to 10 MHz.
- The bactericidal action of US is mainly due to the cavitation process, in which microbubbles are produced and collapsed within a liquid medium.
- During the cavitation process, the temperature can increase to as high as 5500 °C and the pressure can increase up to 100 MPa, resulting in localized microbial sterilization.
- The bactericidal mechanisms of ultrasound include breakage of cell walls, disruption and thinning of cell membranes and free radical activity due to the collapse of cavitation bubbles.

Method of application of ultrasound

- *Ultrasonic horn*: Horn is dipped in the liquid solution or juice and is treated with certain treatment frequency.
- *Ultrasonic bath*: Food material or packaged food is kept and the sound waves are generated in a bath that creates ultrasound effect and brings about desired changes in food.

Applications in the seafood industry

Freezing

- Improves freezing by better preservation of the microstructure; Requires less time and small crystal size; Improved diffusion & Rapid decrease in temperature.

Thawing

- Reduction in thawing time; Preserve colour; Inhibits lipid oxidation; Improved product quality & Reduced product dehydration.

Brining/Pickling

- Low water activity and longer shelf life; Require less sodium chloride & Uniform distribution of salt in less time.

Drying

- Intensification of mass transfer; Shorter processing time; Enhanced organoleptic properties & Increased drying rate due to less resistance.

Merits

- Eco-friendly, green technology
- Ultrasound has successfully proven its potential in the food sector in various critical areas like;
  - -Food preservation
  - -Extraction
  - -Intensified synthesis
  - -Improvement of the physical and chemical properties of food.
- Production of a better-quality product at lower temperatures, with an improved rate of heat and mass transfer.

Demerits

- The very limited technical information about ultrasonication.

- Ultrasound, when applied at high intensities generates heat due to an escalation in temperature, which has detrimental effects on the organoleptic and nutritional characteristics of the food product.
- Lack of consumer awareness about ultrasonic-processed food.

7. Cold Plasma (CP) Technology

- Plasma: Fourth state of matter after solid, liquid, and gas.
- When the energy of gases crosses a certain value, it results in the ionization of gas molecules. Ionization of gas molecules gives rise to plasma.
- Two types

  -Thermal plasma

  -*Cold plasma* (non-thermal)
- Cold plasma is a non-thermal treatment that works in the temperature range 25–65 °C.
- Cold plasma has high antimicrobial activity and efficient enzyme inactivation capacity.
- The composition of the plasma reactive species largely depends on the composition of gas which is ionized.
- The gases commonly used for the generation of plasma include argon, helium, oxygen, nitrogen and air.

Cold plasma generation

- The gases are subjected to any of the types of energy like thermal, electrical, magnetic field, etc., to generate plasma containing positive ions, negative ions, and reactive species like ozone and singlet oxygen.
- Methods

  -Radio frequency plasma

  -Dielectric barrier discharges

  -Gliding arc discharge

  -Microwave

  -Corona discharges
- Cold plasma is an ionized gas generated through gas ionization under corona discharge, dielectric barrier discharge, microwaves or radiofrequency waves.

Advantages & Applications

- Reduction of the microbial load in food or on the surface of food. All kinds of microbes are said to be inactivated by cold plasma technology, including viruses, fungi, and bacteria.

- Enhance the physical and chemical properties of food constituents like lipids and proteins.
- Sterilization of food processing equipments.
- Inactivation of food spoilage enzymes.
- Treatment of food packaging material. Cold plasma can serve for in-package sterilization.
- Treatment of wastewater.
- Cold plasma is produced at near ambient temperature and does not depend on high temperature for microbial inactivation.
- Since the temperature used is ambient, there are no chances of thermal damage to heat-sensitive food material.
- It has continually been referred to as an eco-friendly technique since, besides having minimal changes on the food matrix, its application does not result to the generation of toxic residuals/wastes.

Limitations

- Treatment with CP can induce lipid oxidation in fatty foods and other food products susceptible to oxidation. This may lead to the creation of short-chain fatty acids, aldehydes, hydroxyl acids, and keto acids, thus causing off-flavour and off-odour during storage.
- Undesirable textural properties, acidity, and discoloration of treated food can occur.
- Also, surface topography can be influenced by plasma treatment.
- The high cost of installation is also a major drawback.

8. Ozone treatment

- Ozone is a colorless gas with a typical odor.
- It contains three molecules of oxygen and is chemically written as O_3. It is formed when molecular oxygen (O_2) combines with singlet O.
- Ozone is a very reactive gas, and it is very much unstable and cannot be stored and needs to be produced on the spot when needed.
- Ozone is extensively employed as an effective antibacterial against many bacteria in food. Due to its high oxidizing potential and the ability to attack cellular components, ozone has broad-spectrum of disinfection.

- Ozone treatment is a chemical method of food decontamination that involves exposing contaminated foodstuffs (fruits, vegetables, beverages, spices, herbs, meat, fish, and so on) to ozone in aqueous and/or gaseous phases.

Effect of ozone on microbes

- Ozone alters the permeability of cells by damaging the microbial cell membranes.
- Ozone is also known to damage the structure of proteins, leading to the malfunctioning of microbial enzymes, which affects the metabolic activity and finally results in microbial cell death.
- Chemical composition, pH, additives, temperature, initial bacteria population, and ozone contact time with food and food surface type are factors determining the efficiency of ozone treatment on microbial reduction in seafoods

Merits

- GRAS (Generally Recognized as Safe) chemical with US FDA approval, as well as an antimicrobial additive for direct contact with foods.
- Ozone cleans and disinfects better than chlorine because o ’ inactivation rate owed to concentration limitations posed by regulations.
- It can be used in gas form or it can be mixed with water to form ozonated water.
- Ozone has very great biocidal activity at reduced contact times.
- The lower energy consumption is worth mentioning as a strong merit.

Limitations

- Although ozone treatment can prolong the shelf-life of seafood by reducing the microbial load, pre-treatment with ozone can induce oxidation in seafood. This may cause it to smell or taste less palatable to consumers.
- Due to the enhanced protein oxidation induced by ozone, the functionality of protein in seafood can be decreased, leading to poor-quality products.
- Ozone is one of the strongest oxidizing agents widely used for disinfection of wastewater and removal of organic substances and offensive odour. There is usually a high risk of post-

contamination, since ozone can only lower the microbial load before and during treatment but has less effect on prevention of contamination after treatment.

Other methods

Acidic Electrolyte Water

- Electrolyte water (EW) is made from water without the addition of any hazardous chemicals except sodium chloride.
- EW is known as either a sanitizer (EW containing HOCl, an acidic electrolyte water) or a cleaner (EW containing NaOH, an alkaline electrolyte water.
- The simplicity of EW production and application is the foremost reason for its popularity.
- In numerous fields such as medical sterilization, agriculture, food sanitation and livestock management, EW is gaining attention because of its antimicrobial properties.

Dense phase carbon dioxide (DPCD)

- DPCD processing utilizes the liquefied carbon dioxide and performs at mild temperature and relatively low pressure, about one tenth of the pressures for HHP.
- It is applied to cold pasteurize and extend the shelf life of product without heating.
- Carbon dioxide is a nontoxic, non-flammable and low-cost gas; in the supercritical state, the fluid CO_2 rapidly penetrate porous materials due to its low viscosity ($3–7 \times 10^{-5}$ Pas) and surface tension. This penetration is accompanied with pH decrease, bicarbonate ion generation and cell disruption, which contribute to the microbial and enzyme inactivation.

High voltage electrical discharge (HVED) processing

- Different from PEF in electrode geometry, shape of pulses and mode of actions, HVED generally consists a needle electrode and a grounded one (normally flat geometry) or wire plane.
- Though the advantages of PEF and HVED are promising, the release of metals from the corrosion or migration of electrode materials should be concerned and investigated in the future applications.

Non-thermal food preservation methods: Future Prospects

- Non-thermal treatments are among the most focused research areas in the food sector due to consumer demands for safe and nutritious food free from microbes.

- Despite the active studies on the innovation and improvement of the discussed non-conventional technologies, conventional processing is still dominant in food/fish processing.
- There are still a lot of barriers before the scaling up of the non-conventional technologies in food industry, such as the huge equipment and installation cost, and complex operation process.
- It is very important for the food industries to fully understand the respective action mechanisms as well as merits and demerits of non-thermal food technologies before and even during their implementation.
- Streamlining the process mechanisms of each technique and consumer education about the strengths and prospects of non-thermal technologies could help to raise awareness, prior to considerations on how to amend their designs, if their cost-effectiveness and scale-up capacity for industrial-level applications are to be improved.
- Deep evaluation of the processing line *via* hazard analysis and critical control points (HACCP) methodology to enhance and sustain the improved food hygiene, quality and safety processes.
- Cost comparisons of the selected non-thermal food processing technologies to choose the suitable technology that meets the food production requirements based on the capacities and operational needs.
- When a target food industry that operates at either small-, medium- or large-scale desires to implement a specific non-thermal food processing technology, the prerequisites already prescribed by the manufacturers should be adhered to, despite the variations in facilities/equipment, operational/production scales, intended food product(s), factors of production, as well as consumer targets.
- Developing a hurdle-like non-thermal technology that combines a number of processing methods, designing the intended equipment particularly for large scale application as well as formulating the rules and regulations governing the intended foodstuff safety when using these technologies should be among the future priorities for the food industry and its stakeholders.
- Overall, the clear advantage of these technologies, especially in right combination, makes them a promising approach for inactivation of microorganisms while maintaining sensory attributes and nutritive value.

Conclusion

The demand from consumer for safe and nutritious food products has promoted the rapid development of non-conventional processing technologies. With non-thermal treatments, consumers get high quality, healthy, and safe food products. But there are two sides of the coin: with advantages come some disadvantages as well. If food is exposed for a longer period or treated at a higher intensity, these non-thermal technologies may lead to some undesirable changes in food, such as oxidation of lipids and loss of colour and flavour. But these technologies have many advantages compared to thermal processing. After overcoming the limitations properly in a planned manner, non-thermal technologies will have a broader scope for development and commercialization in food processing industries.

References

- Ananthanarayanan TR, Nithin CT, Toms C Joseph, Bindu J, Srivivasa Gopal TK. Effect of pulsed light on shelf life of chill stored yellowfin tuna (*Thunnus albacares*) steaks. Indian J. Fish. 2019 66(4): 125-134. DOI: 10.21077/ijf.2019.66.4.82781-16
- Annamalai J, Sivam V, Unnikrishnan P, Kuppa Sivasankara S, Kaushlesh Pansingh R, Shaik Abdul K, Lakshmi NM, Chandragiri Nagarajarao R. Effect of electron beam irradiation on the biochemical, microbiological and sensory quality of Litopenaeus vannamei during chilled storage. J Food Sci Technol. 2020 Jun;57(6):2150-2158. doi: 10.1007/s13197-020-04250-7. Epub 2020 Jan 23. PMID: 32431341; PMCID: PMC7230099.
- Bhargava N, Mor RS, Kumar K, Sharanagat VS. Advances in application of ultrasound in food processing: A review. Ultrason Sonochem. 2021 Jan;70:105293. doi: 10.1016/j.ultsonch.2020.105293. Epub 2020 Jul 30. PMID: 32750658; PMCID: PMC7786530.
- Chacha JS, Zhang L, Ofoedu CE, Suleiman RA, Dotto JM, Roobab U, Agunbiade AO, Duguma HT, Mkojera BT, Hossaini SM, Rasaq WA, Shorstkii I, Okpala COR, Korzeniowska M, Guiné RPF. Revisiting Non-Thermal Food Processing and Preservation Methods-Action Mechanisms, Pros and Cons: A Technological Update (2016-2021). Foods. 2021 Jun 20;10(6):1430. doi: 10.3390/foods10061430. PMID: 34203089; PMCID: PMC8234293.

- Devatkal S, Anurag R, Jaganath B, Rao S. Microstructure, microbial profile and quality characteristics of high-pressure-treated chicken nuggets. Food Sci Technol Int. 2015 Oct;21(7):481-91. doi: 10.1177/1082013214546957. Epub 2014 Aug 20. PMID: 25143059.
- Egon Andoni, Enkeleda Ozuni, Bizena Bijo, Fatmira Shehu, Raffaella Branciari, Dino Miraglia & David Ranucci (2021) Efficacy of Non-thermal Processing Methods to Prevent Fish Spoilage, Journal of Aquatic Food Product Technology, 30:2, 228-245, DOI: 10.1080/10498850.2020.1866131
- J. Ginson, Satyen Kumar Panda, C. K. Kamalakanth & J. Bindu (2020) Changes of microflora in high pressure treated Indian white prawn (Fenneropenaeus indicus), High Pressure Research, 40:2, 283-297, DOI: 10.1080/08957959.2019.1708909
- Ginson J, Panda SK, Bindu J, Kamalakanth CK, Srinivasa Gopal TK. Effect of high-pressure treatment on microbiological quality of Indian white prawn (*Fenneropenaeus indicus*) during chilled storage. Food Microbiol. 2015 Apr; 46:596-603. doi: 10.1016/j.fm.2014.10.008. Epub 2014 Nov 1. PMID: 25475333.
- Jadhav HB, Annapure US, Deshmukh RR. Non-thermal Technologies for Food Processing. Front Nutr. 2021 Jun 8; 8:657090. doi: 10.3389/fnut.2021.657090. PMID: 34169087; PMCID: PMC8217760.
- Kundukulangara Pulissery S, Kallahalli Boregowda S, Suseela S, Jaganath B. A comparative study on the textural and nutritional profile of high pressure and minimally processed pineapple. J Food Sci Technol. 2021 Oct;58(10):3734-3742. doi: 10.1007/s13197-020-04831-6. Epub 2020 Oct 13. PMID: 34471297; PMCID: PMC8357897.
- Kunnath S, Jaganath B, Panda SK, Balange AK, Gudipati V. Effect of high pressure and setting condition on physico-chemical, structural and functional characteristics of transglutaminase mediated fish gels. Food Sci Technol Int. 2020 Dec 10:1082013220978103. doi: 10.1177/1082013220978103. Epub ahead of print. PMID: 33302726.
- Li F, Chen G, Zhang B, Fu X. Current applications and new opportunities for the thermal and non-thermal processing technologies to generate berry product or extracts with high nutraceutical contents. Food Res Int. 2017 Oct;100(Pt 2):19-30. doi: 10.1016/j.foodres.2017.08.035. Epub 2017 Aug 24. PMID: 28888440.

- Munir MT, Federighi M. Control of Foodborne Biological Hazards by Ionizing Radiations. Foods. 2020 Jul 3;9(7):878. doi: 10.3390/foods9070878. PMID: 32635407; PMCID: PMC7404640.
- Olatunde OO, Benjakul S. Nonthermal Processes for Shelf-Life Extension of Seafoods: A Revisit. Compr Rev Food Sci Food Saf. 2018 Jul;17(4):892-904. doi: 10.1111/1541-4337.12354. Epub 2018 May 10. PMID: 33350115.
