INTERNATIONAL
Agrophysics
www.international-agrophysics.org

Int. Agrophys., 2024, 38, 87-119
doi: 10.31545/intagr/177513

Review

Sustainable laser technology for the control of organisms and microorganisms in agri-food systems: a review

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Received November 3, 2023; accepted December 22, 2023

Abstract. We review the literature concerning the effects of laser light on organisms (insects) and microorganisms (bacteria, viruses, fungi) present in agri-food systems. The evidence obtained shows that 1) Laser light is a sustainable technology that can be applied as a pesticide with the ability to annihilate and control insects. Higher annihilation rates are observed for more pigmented products, as determined by their thermal and optical properties. 2) The most frequently used laser beams to eliminate bacteria harmful to human health operate with a steady intensity in the visible domain (blue, green, and red light). 3) Laser beams are applied to control fungi (the most studied microorganism), viruses, as well as to increase plant resistance to them. Lasers with red beams, such as those emitted by He-Ne lasers, followed by diode lasers are most frequently reported in fungal control. Furthermore, antibacterial, and germicidal effects are increased by using photosensitizers. Finally, 4) laser light potentiates the metabolites and antimicrobial activity of some plants thereby improving their activity. Agri-food systems treated with laser beams have the potential to improve the quality of life of society.

Keywords: laser technology, sustainable technology, antibacterial, antifungal, pesticidal

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1. INTRODUCTION

Laser light has potential application possibilities in agriculture and food production, this has been reported by several authors (Janicki *et al.*, 1987; Vasilevski, 2003; Hernández-Aguilar *et al.*, 2010; Nadimi *et al.*, 2021; Teng *et al.*, 2021); for a wide variety of crops at various phenological stages of agricultural production, *e.g.* pre-sowing, seedling, plant growth, post-harvest, and food processing (Hernández-Aguilar *et al.*, 2010; 2016; Shabir *et al.*, 2022). Among the main applications are those related to the quality aspects of grain, seed, product, and food; concerning different attributes of physical, physiological, sanitary, and nutritional quality (Hernández-Aguilar *et al.*, 2006; 2011; Krawiec and Dziwulska-Hunek, 2018). These applications include characterization and detection and/or quality control, for example, the speckle structure of laser light is used by means of the biospeckle technique in addition to photothermal spectroscopic and microscopic techniques

(Hernández *et al.*, 2008; Domínguez *et al.*, 2010; 2022; Aladjadjiyan, 2018; Hernández *et al.*, 2019; Nadimi *et al.*, 2022). Also, laser light was reported as a biostimulatory element; possible uses include to increase plant resistance to various biotic and abiotic stresses (Ferdosizadeh *et al.*, 2013; Awaad, 2021; Romero-Galindo *et al.*, 2022) and to reduce pesticides (Hernández-Aguilar *et al.*, 2011a; Awaad, 2021; Krawiec and Dziwulska-Hunek, 2018). Applications of laser technology have the potential to improve the quantity and quality of seeds and consequently of food production, this would favour the achievement of food security (Awaad, 2021). This is one of the complex problems that humanity is facing due to population increase and the increasing prevalence of disease.

The development of technologies designed to improve agri-food systems make a vital contribution to the achievement of a sustainable world. In recent times, as a result of the Covid-19 emergency, the sustainability index decreased for some countries and failed to evolve for others, so it is necessary to add proposals which may contribute towards fulfilling the current sustainability goals (Qaim, 2020; Hernández-Aguilar *et al.*, 2022). The emerging environmentally friendly technologies such as laser technology may be used to address several problems in agri-food systems and increase the yield, productivity and quality of crops and products. As a consequence, consumer health may be improved and adverse environmental impact may be reduced. The promotion of further research and the appropriate management of its application in real-world agri-food systems could potentially have an impact on certain sustainable development goals such as number 2 "Zero hunger and improved nutrition", number 3 "Health and well-being", number 12 "Responsible consumption and production" and number 15 "Life of terrestrial ecosystems" (Barbier and Burgess, 2017; Biermann *et al.*, 2017). Other technologies that have been proposed in the literature for seed conditioning, and those related to biophysical methods, *e.g.* magnetic field, ultrasound, microwave, plasma, UV, *etc*. (Li and Farid, 2016) could be used in conjunction with technologies based on vegetable biostimulant methods (González *et al.*, 2020).

Numerous articles have confirmed the bioeffects of the biostimulatory processes of the application of low-level laser light (Hernández-Aguilar *et al.*, 2010; 2016; Aladjadjiyan, 2012; Mandal and Maity, 2013) as seed conditioners in pre-sowing, in seedlings or plants during their development through the application of photonic energy at specific wavelengths and doses that produce a macro or micro effect (Hernández-Aguilar *et al.*, 2016). Over time, these studies have intensified, thereby demonstrating their broad potential for application in the agri-food sector, from the 1970s to the present day (Hernández-Aguilar *et al.*, 2010; 2016; Govindaraj *et al.*, 2017; Nadimi *et al.*, 2021; Awaad, 2021). However, the laser bio-effects reviewed in the literature to date have focused mainly on the application of laser light for the improvement of physiological and chemical attributes and its potential to improve seed conditions under stress. Likewise, they have also focused on bioremediation problems as a single treatment (Romero-Galindo *et al.*, 2021) or in a hybrid way, *i.e.* a combination of treatments (Dziwulska-Hunek *et al.*, 2009; Asghar *et al.*, 2016; 2017; Abdani *et al.*, 2018) which are a trend in the application of biophysical methods for bio-stimulation purposes (Hernández-Aguilar *et al.*, 2023). The use of agrochemicals must be avoided, farmers often use them to solve problems in the production system as they guarantee increased production levels and reduce losses, however, they do not provide a viable pathway toward sustainability. The decisions which farmers take are important, as they may risk their own health and the health of their families. Also, the health of their potential consumers and that of the environment are affected by the actions of farmers.

The farmer and/or agricultural producer requires quality seeds and healthy production systems. The quality attributes of agricultural seeds are essential. Healthy seeds with a high degree of health and vigour are the basis for the growth and development of plants which have the potential to ultimately produce better yields. In summary, crops with a successful growth period and yield, as well as an adequate degree of production to satisfy the demand for consumption by the population and a reduction in the incidence of both diseases and losses (Uthairatanakij *et al.*, 2007; Abawi and Widmer, 2000).

Thus, the objective of this contribution is to review the literature that shows evidence of the possibility of using laser technology as a sustainable strategy for the improvement of agri-food systems, with a particular focus on the effects of laser light on the organisms (insects) and microorganisms (bacteria, fungi, and viruses) involved. Laser technology not only has the potential to be applied in agricultural production systems as a biostimulator element and to reduce the use of fertilizers, *etc*., but also, it has the potential to reduce the use of pesticides in general.

Laser technology could also reduce foodborne hazards (and therefore the risks of both current or future illnesses in the population), *i.e*., improve food safety, the risks to which may be biological (bacteria, viruses, fungi, parasites) and chemical in nature, including substances used in production systems that are harmful to the farmer, the consumer, and the environment (*e.g*., pesticide residues).

In view of chronic hunger and nutrient deficiencies which are affecting several countries at the present time (Domínguez *et al.*, 2022), and also the transmission or development of disease due to the lack of food sanitary quality; laser light may be applied to various problems of agricultural production in an achievable way and thus move the world towards community sustainability (Hernández-Aguilar *et al.*, 2022), with the additional effect of halting any destructive processes adversely affecting the soil, the environment, and the production of food for home consumption by families which provide good health as a result of avoiding the consequences of agrochemical use. In this sense, laser technology could be a viable strategy for achieving the coexistence of economic, social, and environmental interests which is one of the main challenges on the road to a sustainable world.

2. BACKGROUND

2.1. Light and its early applications on microorganisms and organisms

The fascination with light is an ancient phenomenon because it is ultimately the source of all life, the beneficial effects on the body and mind are obvious, also, light is essential for vision and warmth (Hockberger, 2002). In addition, it plays a very important role in agriculture, which is to allow to produce food that enables us to live. One of the first applications of light was proposed by Aristotle in ancient Greece. He used a dark room, and allowed light to pass through a hole, the result was that an image was created on the opposite wall. He also observed that the size of the hole influenced the sharpness of the image, Hassan ibn Hassan (965-1040) then wrote about the effect of different hole sizes; the smaller the hole, the sharper the image (Kirkland, 2007). Later, the photosensitive properties of silver were discovered, which led to another discovery, UV light. The first experiments were conducted by Angelo Sala (1614), who observed that sunlight produced a blackening of silver nitrate crystals and by Scheele who discovered that paper soaked in silver chloride solution darkened when exposed to sunlight, he observed a greater effect on the violet side than on the reddish side (Hockberger, 2002).

In 1801, Johann Wilhelm Ritter scattered light through a prism and divided it into its constituent colours. He discovered that the scattered light had a notable effect on silver salt. It was found that the salt is activated with a higher speed and colour intensity at the non-visible portion of the spectrum, beyond the violet wavelength (Barth, 1987). These rays were called “deoxidizing rays” to emphasize their chemical reactivity (also called chemical rays) and to distinguish them from the “caloric rays” discovered a year earlier by William Herschel (1738-1822) (Herschel, 1800; Hoskin, 2008). These types of radiation are known today as ultraviolet and infrared radiation respectively. The application of light in order to combat microorganisms goes back to ancient times. Sunlight was used by Egyptians to disinfect and heal wounds as early as 5000 BC (Enwemeka *et al.*, 2021) and in ancient ayurvedic medicine to purify water in 2000 BC (Patwardhan, 1990). However, the beneficial effects of sunlight were only initially formalized after the discovery of “deoxidizing” rays and the discovery of the microscope (Moghissi and Allison, 2023).

One of the most important works in the history of photobiology and its applications was completed in 1877. Downes and Blunt in 1877 published two articles, one of them in the journal Nature (Downes and Blunt, 1877), which anticipated conclusions that were later reached through investigation. The authors confirm that sunlight inhibits the development of bacteria, thereby indicating that under favourable conditions it can be applied to prevent their development and that under less favourable conditions it retards their development *i.e.* what is now known as the “bacteriostatic effect” and that these effects are achieved through insolation. In the second paper which was published in the Proceedings of the Royal Society of London, Downes (1877) described the experiments performed in detail and also the relevant findings from the months of April-October. The researchers investigated the effects of sunlight on Pasteur’s solutions (weighted and unweighted) formed with water, tartaric acid, brown sugar candy, potassic carbonate, ammonium nitrate, ammonium phosphate, the solution neutralized with ammonia, and filtered. Downes and Blunt found that sunlight inhibited the growth of microbes in test tubes containing Pasteur’s solution. Another observation that was made was that bacteria were affected more than fungi. These experiments, together with others published a year later, led them to consider the importance of the wavelength of the light, the radiation dose (which refers to the intensity and duration time of exposure) and the characteristics of the microbes in order to obtain certain effects using these light applications (Downes and Blunt, 1879). Numerous studies followed in which the scientists of the time attributed the various effects to the experimental conditions (the presence or absence of oxygen and the characteristics of the medium) or to the temperature produced by sunlight. Downes (1886) performed an experiment at low temperatures, showing that even at low temperatures bacterial growth was inhibited, and therefore the temperature was not responsible for the bactericidal effect of sunlight. On the other hand, in 1890, Janowski investigated the colour of the medium solutions, the temperature, and the different colours (wavelengths) of the applied light. He showed that both direct and diffuse sunlight produced a higher degree of bacterial growth, in yellow and black solutions relative to violet and blue solutions (Hockberger, 2000).

Early evidence obtained by Geisler (1892) emphasized that all regions of the solar spectrum (ultraviolet, visible, and infrared light) can affect bacteria (Hockberger, 2002; Reed, 2010). In addition, Ward's discoveries

carried out between 1892-1895 who reported on the relationship between the number of bacteria and the wavelength applied and the bactericidal effects (more pronounced) in the UV and blue regions. However, the mechanisms of action between the UV and the blue regions of the spectrum are different from those attributed to the visible and infrared region of the spectrum (Ward, 1893; 1894a, b).

Duclaux (as a student of Pasteur) demonstrated the different degrees of susceptibility to solar photoinhibition among the various microbes. In addition, it was shown that the tuberculosis bacillus was susceptible to sunlight; (Photon energy: 2-3.3×10^2 eV), the important regions of which are classified as UV-A (320-400 nm), UV-B (280-320 nm) and UV-C (200-280 nm). This discovery led to the use of UV to combat tuberculosis which required the development of artificial UV lamps (Enwemeka *et al.*, 2021). Previously, phototherapy was based on sunlight, in those times it was known as heliotherapy (Moghissi and Allison, 2023).

In 1897, Niels Ryberg Finsen (1860-1904) built an arc lamp (it is known today that it generated light in the violet-blue spectrum). This lamp was known as the "Finsen lamp" and it was illuminated by a reliable carbon arc torch, the light it emitted could be tuned by filters in order to select a desired wavelength. It was demonstrated that both the natural rays of the Sun and those originating from an arc lamp had antibacterial activities regarding the treatment of the disease *Lupus vulgaris* (*Cutaneous tuberculosis*), *i.e.* the disease could be cured. Finsen treated more than 800 patients with *cutaneous tuberculosis* and microbial infections, achieving a cure rate of more than 80%, for which he was awarded the Nobel Prize for Medicine in 1903 (Moller-Sorensen and Brade, 1995; Grzybowski and Pietrzak, 2012; Moghissi and Allison, 2023).

Various diseases are related to certain microorganisms, so the effects of sunlight radiation had an impact on the diseases of the time (typhoid, cholera, diarrhoea, dysentery, anthrax, *etc.*) (Hobday and Dancer, 2013; Enwemeka *et al.*, 2021) and in addition, various types of lamps continued to be developed, including mercury lamps in 1901. Likewise, quartz was recognized as the ideal lamp envelope for this type of lamp (Wright and Cairns, 1998). Discoveries and applications such as these, at a time when antibiotics had not yet been discovered, were revolutionary (Moghissi and Allison, 2023).

The applications of light diversified over time, it was first used to disinfect water in 1909 in France and then in the United States (Darré *et al.*, 2022). Subsequently it was applied to environmental issues, it was applied to surfaces in different areas such as in the food industry for the disinfection of fruits and vegetables. In addition, it was also used on work surfaces, conveyor belts, and on packaging etc. From among the various regions of ultraviolet light (UV- vacuum (100-200 nm), UV-C (200-280 nm), UV-B (280-315 nm), and UV-A (315-400 nm)) (Meulemans, 1986), the UV-C (200-280 nm) region is the one that has been most widely used, this is due to its greater effects against viruses, bacteria, fungi and other natural threats (Bera *et al.*, 2022; Darré *et al.*, 2022). Furthermore, it has the highest photon energy given by Planck´s Equation $E = hv = hc/\lambda$, (where h is Planck's constant (6.626 x 10^{-34} joule-second), c is the velocity of light, λ is the wavelength and v is the frequency (c/λ)) (Abu-Elsaoud *et al.*, 2022).

The lamps used most frequently were mercury lamps emitting light at a wavelength of 254 nm and pulsed Xenon lamps, which are capable of emitting short pulses (in the order of milliseconds) of broad-spectrum radiation including wavelengths in the UV and near-infrared range. Pulsed light technology involves the application of intense light in the form of short, high-intensity pulses aimed at a target of interest, with a high degree of penetration, maximum power emission capability and at a maximum power distribution during the short pulses. The pulsed UV light effectively inactivates micro-organisms and organisms such as bacteria, yeasts, fungi, viruses, and also insects (Gala *et al.*, 2021). However, it should be remembered that there are dangers associated with UV radiation, its misuse is not compatible with human health, this depends on the lamps and wavelengths used, for example, UV-C should not be viewed with the naked eye, adequate ventilation is vital as this wavelength sometimes generates ozone. From the point of view of ameliorating the possible harmful effects of the application, of light, advances in technology in the development of new artificial light sources are important, as well as demonstrating beneficial effects in the visible regions.

Technological discoveries and increasing needs have allowed for the development of the practical applications of light in the real world. In the 1960s, laser light emitting devices were developed, it was yet another revolutionary form of light with a wide range of applications in different areas of life, society, and in a variety of disciplines. From its development to the present day it has presented a wide range of advantages in relation to other artificial light sources. In biology, its use began with the first research by Bessis *et al.* (1962) who used a Ruby laser (red band). Other applications have been reported in the fields of phototherapy, photochemotherapy and dynamic phototherapy most frequently with the use of the He-Ne laser (Hernández *et al.*, 2010; Moghissi and Allison, 2023). In just the first few years of its discovery, both in the areas of medicine and biology, several articles covering the topic had already been published according to Eichler and Lenz (1977), from 1963-1974, various authors had published a total of 900 papers, covering 23 laser applications, among which the following areas stand out: Microprobe applications, Basic research, Neurosurgery, Biochemistry, Oncology, Blood applications, Ophthalmology (Bones and Clinical), Cytology, Research, Dentistry, Otolaryngology, Dermatology, Embryology, Review papers, Holography, Scattering, Instrumentation, Surgery, Laser safety, Urology, Wounds, plants, entomology and applications involving insects and bacteria.

Therefore, it may be stated that laser applications to benefit agri-food systems have been found from the early days of its discovery and that several applications involving insects, bacteria and plants were developed. The laser was introduced as a biostimulant in agriculture in 1969 and 1970 (Hernández *et al.*, 2010) after the discovery of the "biostimulation" phenomenon by Mester *et al.* (1967) according to Enwemeka (1988) and Hernández *et al.* (2010). The first reported investigations were conducted by Wilde *et al.* (1969) and Paleg and Aspinall (1970) as noted by (Hernández *et al.*, 2010). This was followed by the studies of Usmanov *et al.* (1970), Musayev (1971) and Floyd *et al.* (1970) who reported the mutagenic effect of laser irradiation in the seeds of *Arabidopsis thaliana* L. and tomatoes and also the photoinduced reactions at low temperatures in green leaves and chloroplasts. The mutagenic effects point to the need to define the window of values where beneficial effects can be found with the application of laser light. Therefore it is necessary to determine the adequate irradiation parameters without exceeding certain irradiation levels; this is because some mutagenic process could occur at long exposure times and high intensities, depending on the object of the study (Ritambhara and Kumar, 2013). The basis of the stimulation mechanism at any physiological stage of the plant is the synergy between the polarized monochromatic laser beams and the photoreceptors (Bielozierskich and Zolotariewa, 1981; Koper, 1996, Hernández *et al.* 2010; 2016).

However, laser light not only has the potential to be applied as a biostimulatory element in agriculture, and to present the possibility of reducing the use of chemical fertilizers or as a growth inhibitor when applied to weeds (Hernández *et al.*, 2010; 2016), its application may increase tolerance to environmental stress and raise water use efficiency and treatment (Vasilevsky, 2003; Awaad, 2021). In addition, applications in the food industry such as the pretreatment of food raw materials have been reported, these processes include packaging, cutting, peeling, milling, marking, drying, cooking, ablation, fermentation and the shelf-life extension of liquid food, *etc*. (Teng *et al.*, 2021; Chiabrando *et al.*, 2019). In general, laser technology may be applied in agri-food systems, it has different functions such as: 1) in food processing, 2) as an agricultural biostimulant element, 3) to enhance quality control through the diagnosis and characterization of study objects and 4) the control of organisms and microorganisms as an element of pest control, in order to limit the adverse effects of insects, bacteria, viruses, and fungi and/or enhance the beneficial effects of active substances in order to improve the metabolites of plants and microorganisms applied to seeds or plants. In this paper, we focus on the latter application (4). In addition to their application in food processing and in the conditioning of seeds, seedlings and plants, laser light sources could be applied to improve sanitary quality and help to reduce the use of agrochemicals during the development of crops in the face of different problems of the agri-food system all the way up to the level of home consumption, in circumstances where agrochemicals or chemicals that harm the environment and human beings are currently being used. This physical technology has advantages over the chemical inactivation of microbicidal procedures and is considered to be environmentally friendly as it does not leave potentially toxic residues in the environment and has the potential to extend the shelf life of perishable foods (Govindaraj *et al.,* 2017; Gala *et al.*, 2021; Chavan *et al.*, 2023; Gonca *et al.*, 2023).

The pioneering works that demonstrated that laser light could be a promising light source used to control the spread of harmful organisms and microorganisms were conducted in the 1960s shortly after the development of this light source, these studies were undertaken even before its biostimulant capacity was considered. In this way, the first studies with agri-food systems were specifically centred around its potential as a pesticide. It is important to emphasize that more than 50 years ago in a publication of Wilde *et al.* (1969), possible applications of laser light were visualized with reference to the reduction of the use of pesticides in the development of agricultural crops.

3. EFFECT OF LASER LIGHT ON ORGANISMS: INSECTS

The application of laser light as a pesticide to control or exterminate insects that damage agrifood systems has been reported by several authors. The lasers used were both pulsed and continuous wave variants, although the latter was used to a lesser extent. Since its development, pulsed light has been applied using a Ruby laser (red), and later in the form of ultraviolet light (UV-C), blue light (405, 444, 445, 450 nm), green laser light (527, 532 nm), red light (640 nm) and in the far infrared (FIR) of CO_2 (10, 600 nm), near and mid-infrared (NIR and MIR)- (976, 1064, 1070 and 1470 nm). However, continuous light has also been employed using low intensity laser diodes, e.g. at 650 nm and 5mW, as well as at 532 nm and 200 mW. The main characteristic of a pulsed light source as compared to continuous light is that it operates over short periods of time and utilizes a higher power of the laser light source. The most probable mechanism of action with regard to the interaction of laser light with the living cells of insects is a photochemical interaction; absorbing chromophores or photoacceptors that have a high degree of absorbance at the wavelength of the laser light applied are worthy of consideration and these in turn produce a heating of the affected area, thus causing tissue changes which cause insect mortality or harmful structural changes in the insect (Chernova and Vorsobina, 2002; Meng *et al.*, 2009).

The first applications of laser light concerning insect control and/or extermination were made by Wilde (1965; 1967), one of the pioneers of seed biostimulation (Wilde *et al.*, 1969), he discovered that laser light could cause some changes to insects and developed novel agricultural systems. Wilde (1967) investigated the effects of pulsed ruby laser light on various arthropods, among which were mites (*Panonychus ulmi* (Koch) and *Tetranychus urticae* (Koch)), greenhouse whitefly (*Trialeurodes vaporariorum* (Westwood)); the green aphid found on peach trees (*Mysus* *persicae* (Sulzer) and the psylla of pear tree (*Psylla pyricola Foerster*). The host plants were pear seedlings (*Pyrus communis*), alfalfa (*Medicago sativa*) and rye grass (*Bromus* sp.). The authors reported damage to both arthropods and the host plants. The darker the arthropod pigmentation, the greater the damage, this effect was influenced by the structural shape and the microdistance between the arthropods (at different stages of development) and size of host plant area. Other studies conducted on insects by Wilde (1965), were concerned with *Dermestidae larvae, Trogoderma versicolor* (Creutzer) and adult American cockroaches, *Periplaneta americana* (Linné). These specimens were bombarded by ruby laser pulses of two intensities: 0.06 joules with a 10-7 s duration and 0.57 joules lasting just 0.2 milliseconds. The laser impact zones were 1.5 mm in diameter. The first intensity caused mortality in 3-48 h. The total mortality in the five treated specimens occurred in 8-14 days as compared to 45-68 days in the controls.

Several authors have evaluated the effects of various laser light wavelengths: green (532 nm), far infrared (10, 600 nm), near and mid infrared (976, 1064 and 1470 nm) on mosquitoes Anopheles stephensi hembra and flying insects that present risks to agricultural production and are reported to carry malaria. The researchers applied different pulse spot sizes, pulse intensities and pulse durations. Pulses with larger laser spots required more energy to produce a sufficient degree of lethality to kill or disable insects with optical energy. The authors found that green (532 nm) and far infrared (10, 600 nm) wavelengths were more effective than near and mid-infrared wavelengths. The pulse duration and power deployed were 2-45 ms, 0.5-8.5 W and 2-25 ms, 1.1-10.2 W for the green and 10, 600 nm lasers, respectively (Keller *et al.*, 2016). These results allowed for an automated system to be designed to combat the flying insect mosquitoes with the necessary parameters and suitable laser type.

The effects of pulsed laser light have been reported concerning mosquitoes which pose a potential risk to horticultural crops and cause losses in citrus crops since they transmit *Candidatus Liberibacter asiaticus*; the pathogen that causes citrus greening disease and malaria vectors such as *Anopheles stephensi*. Mullen *et al.* (2016) showed the susceptibility to laser radiation of insects by using a 445 nm laser with a power output of 670 mW. *Diaphorina citri* were placed in a transparent acrylic box and anesthetized with CO_2. The authors applied the laser light on one side of the thorax and in the same plane relative to the laser. The laser pulse times ranged from 3-28 ms with a power density of 440 W cm^{-2}. A 90% mortality rate of the mosquitoes was found at 15 mJ, although the onset occurred at a low energy level (2 mJ). Thus, the authors could integrate their real-time flying insect detection system and a control and eradication of flying insect system by means of lethal pulses of laser light.

Gaetani *et al.* (2021) have pointed out that laser light is one of the viable options of the available physical methods for pest control, it can provide a promising alternative to chemical pesticides. Some authors have investigated the lethal dose required to kill 90% of the populations of two aphid species (*Acyrthosiphon pisum* and *Rhopalosiphum padi*) which are important pests in crops, as they damage them and can also spread diseases that generate economic losses.

Three types of lasers with pulsed light emissions (CO_2, 532 nm and 1 070 nm manufactured by IPG) were used. The aphids were reared on plants of the *Vicia faba*, L. cv. *Aquadulce* and *Triticum aestivum Linnaeus*. The authors demonstrated that the irradiation of day-old nymphal insects reduces the lethal dose without affecting plant growth and health. The laser is lethal in most cases, but it can also cause stunted insect growth and the reduced fecundity of the survivors. The laser that produced the best result was the CO_2 infrared laser.

The evidence found in several reports indicates the importance of estimating the appropriate laser parameters. The power used, the irradiation area, laser aperture time and the position of the irradiation on the object of study are all important considerations. This has been proven by several studies. The *P. rapae antifeedant larvae* are cruciferous pests (which have an impact on cruciferous plants as they consume all of their leaves and can even cause soft rot) were the subject of another study conducted by Li *et al.* (2021). The authors found that within 24 h there was a 100% mortality rate. A semiconductor laser at 450 nm with 10 W of power was applied. The optimal parameters were a laser power of 7.5 W, irradiation area: 6.189 mm^2, laser opening time: 1.177 s and the position of incidence of the laser light was directed towards the abdomen to achieve control of these pests. The diameters of the irradiation area were set at 0.1-3 mm and laser irradiation was performed without fixing the larvae, four different laser opening times (0.5, 1, 2 and 3 s) were tested in the trials.

Most recently, Zaidem *et al.* (2023) reported a reduction in the population of the whitefly Bemisia tabaci, another common pest in agricultural systems, bean crops (*Phaseolus vulgaris* L.) were the object of the study and the appropriate laser irradiation parameters were applied. The authors applied continuous wave lasers with different wavelengths (444, 527 and 640 nm) and optical intensities directly on the insects to eliminate them. They point out that the laser wavelength which produced the best result was 444 nm. By using a homemade system they reported 100% mortality of insects irradiated at a wavelength of 454 nm on 3rd instar nymphs of *Bemisia tabaci*, with the following laser irradiation parameters in terms of time and intensity of 1s, 10 W cm^{-2}; 3 s, 4 W cm^{-2}, and 4 s, 4 W cm^{-2}. In addition, the laser irradiation trial did not affect plant yield or development, it was reported that using a laser photonics technique had the potential for whitefly control.

Insects which consume food products have also been studied in order to demonstrate the suitability of laser radiation as an alternative to chemical pesticides for the control of two types of insects originating from stored flour: *Tribolium castaneum* and *Trogoderma granarium*. In this study, laser light at 405 nm and 110 W was used, it was focused on a container containing insect larvae or adults, using an experimental design with four exposure times plus the control (0, 10, 20, 30 and 40 min). The treated insects were transferred to food media and after 24 h the biological objects of each pest were counted. The study showed a decrease in the survival rate of adults and larvae of the pests which were evaluated with increasing exposure times. The researchers found that the various exposure times caused significantly different mortalities in adults of the two types of pests tested when compared to the control samples. They reported that for *T. castaneum* and T. *granarium* an adult mortality rate of 100 and 83.3% occurred at exposure times of 30 and 40 min, respectively (Ubaid, 2016).

Low-intensity diode lasers, which are less expensive commercially, have also been reportedly used for insect extermination. Mahdi *et al.* (2021) evaluated the effects of diode lasers at 650 nm and 5 mW on white ants (*Psammotermes hypostoma*). Various exposure times (60, 70, 80, 90 and 100 s) were applied at 3 and 5 cm, and the effects were evaluated at 12, 24, 48 and 72 h. The authors concluded that the thermal effects of the laser resulted in an increase in the destruction of bodily structures causing the deformation of the insect as the exposure time increased, thus increasing the mortality rate, the most affected samples were those closest to the radiation source. In this sense, the interaction of laser light with tissues may be described as an absorption effect, leading to the evaporation and shrinkage of parts of the insect as a result of fluid and nutrient loss due to thermal changes which occur upon exposure to laser radiation.

The researchers Amaar and El-Refai, (2015) used a Khapra beetle, *Trogoderma granarium* (*Everts*) (*Coleoptera Dermestidae*); one of the main insects which consume stored cereals, a diode laser of 532 nm, a power output of 250 mW, and exposure times of 30, 45, 60 and 75 s. The reported results are that laser light reduced egg hatchability and pupation, as well as adult emergence from exposed 0-1-day old eggs or 0-1-day old pupae. However, although laser irradiation significantly reduced the germination rate, it had no effect on chlorophyll c.

Other studies with laser diodes and ants have been reported, *e.g.*, Rashid *et al.* (2021) experimented with ants (*Camponotus consobrinus*). In this research, the authors used a 532 nm laser diode with 200 mW of power placed at (10, 12 and 14 cm) under the exposure times of 10, 15 and 20 s. In addition, they described the thermal component produced by the laser light. They stated that thermal diffusion produces an increase in the mortality rate of ants due to the damage inflicted on their internal structures which occurs when they are heated by a laser. It was found that an increase in the exposure time and a decrease in the separation distance between the laser and the object of study produces the highest mortality rate and deformation of the ants.

Rashid *et al.* (2018) studied the cowpea beetle insect, which affects legume seeds such as chickpea and others, causing weight loss during storage. In this study they used the pulsed Nd:YAG laser, with a wavelength of 1 064 nm and exposure times of 10, 20 and 30 s with 5 pulses/s at a distance of 10 cm between the laser radiation source and the specimen. Evaluations were carried out at 12, 24 and 48 h. The laser provoked a thermal effect causing damage to the insect structures and therefore increasing the percentage of insects which died.

Combinations of light-based systems to solve insect based problems in crops have been proposed in the literature wherein insects are attracted by ultraviolet light and annihilated by lasers. Nair *et al.* (2023) constructed a trap for fungus gnats (*Diptera: Sciaridae*) which are common in seta crops using UV-A light from LEDS (365 nm) with a frequency of 40 Hz. The insects were annihilated by pulsed blue laser light (445 nm) with an activation every 10 s and a duration time of 200 ms.The authors found that the assembled system effectively contributed to reducing or controlling fungus gnat numbers in a fungus (*Pleurotus ostreatus*) plantation.

4. EFFECT OF LASERS ON MICROORGANISMS

4.1. Virus and bacteria

The application of laser light as a sterilizing element was discovered in the 1960s. Ruby lasers were the first lasers to be used and demonstrated their biocidal capacity against *Spirogyra* and *Amoeba* with the experiments of Saks and Roth (1963). These and other experiments produced results which cumulatively provided proof that laser light has a sterilizing effect on bacteria (McGuff and Bell, 1966; Deschauz *et al.*, 1969; Korn and Chel'ny, 1970).

Macmillan *et al.* (1966) used a 632.8 nm continuous wave, low power gas laser of 21-30 mW with exposure times of 5-60 min to irradiate the cells of seven species (*Sarcina lutea* (two strains), *Escherichia coli*, *Chromobacterium violaceum, Arthrobacter atrocyanus, Pseudomonas aeruginosa, Saccharomyces cerevisiae* and *Rhodotorula glutinis*), which were rapidly killed when aerobically irradiated in aqueous toluidine blue solutions. According to Blum (1941), photodynamic action, or death caused by light and oxygen, is the sensitization of a biological system to light, it is caused by a substance that absorbs light and participates in photochemical reactions that require oxygen.

This approach opens up a wide spectrum of possible applications of laser light on various microorganisms that are harmful to agricultural crops, and in general to agri-food systems, including the media used in them. As a consequence, these treatments can potentially be applied, both in agricultural systems, as was also pointed out by Maktabi *et al.* (2011), and in the food industry, using pulsed or continuous laser light with applications of interest being used to preserve the shelf life of food (Mahendran *et al.*, 2019).

Also, the use of pulsed laser light has been reported in liquid foods. Smith *et al.* (2002) investigated the effects of pulsed UV-C (248 nm) excimer laser light. In this research, it was reported that the authors expanded the laser beam through a flat convex lens and used the laser light of an intensity of 0.23-0.25 J cm^{-2} to treat bovine milk that was inoculated using bacterial cells (*Escherichia coli, Listeria monocytogenes, Salmonella choleraesuis, Yersinia enterocolitica, Staphylococcus aureus, Aeromonas hydrophilia*, and *Serratia marcescens* pigmented red). Growth was observed at intensities of 0.3 and 6.6 J cm^{-2}, but no growth occurred at 12 J cm^{-2}. The studies showed that laser light at the UV-C wavelength is capable of rapidly and irreversibly altering the genetic mechanisms of bacterial targets (cell wall or ribosomes), mainly through non-thermal photolytic processes.

In addition, Kasahara *et al.* (2015) studied the effects of pulsed UV laser light on goat milk inoculated with *Escherichia coli* bacteria. As a light source, they employed a monochromatic exciplex laser unit (248 nm), that had radiation parameters with a pulse frequency of 20 Hz; a laser beam area (irradiation footprint) = 3 cm^2 average; and a half maximum average energy = 410 mJ $pulse^{-1}$. The irradiation doses applied were 1 300; 2 500; 3 800; 5 000; 6 300; 7 500; 8 800 and 10 000 mJ cm^{-2}. The findings reported by the authors were the inactivation of *E. coli* as a function of the increasing dose applied. They reported a 6-Log reduction of the initial microbial population at a laser radiation dose of 10 000 mJ cm^{-2}. Although they reported that the flavour of the milk did not change, there were changes in the aroma from a dose of 5 000 mJ cm^{-2}.

Other authors have also explored different wavelength levels in the infrared region and other types of bacteria, *e.g. Pseudomonaus aeruginosa*. Shelygina *et al.* (2021) applied femtosecond lasers in the mid-infrared range (5.8 μm) in order to evaluate the possibility of the inactivation of microorganisms using resonant vibrational excitation. The ultrashort pulsed laser beam had a duration of 130 fs and a pulse energy of 2 μJ. The intensity of the pulses were applied without filters and with filters of 5 and 0.3 GW cm^{-2}, respectively. The bacterial layers showed differences in the spectra obtained through FT-IR spectroscopy when compared to the control samples, this may indicate the inactivation of pathogenic microorganisms. As a result of the irradiation of bacterial cell cultures of *P. aeruginosa*, a blue shift was demonstrated in the region of the absorption bands characteristic of proteins and nucleotides of the bacterial cell. This indicates the cleavage of the hydrogen bonds responsible for the formation of the secondary and tertiary structures of the proteins related to the inactivation of microorganisms.

Kohmura *et al.* (2020) produced evidence of the transient photothermal inactivation of *E. coli* stained with visible dyes (safranin dye, rhodamine B) using a 532 nm nanosecond YAG laser applying two types of continuous and pulsed light. Of the dyes used, the one with the highest absorbance at 532 nm was safranin dye (0.38) as compared to the absorbance of rhodamine (0.2) and bacteria without dye (0.08). The use of dyes with an absorption spectrum peaking at 532 nm, produced an effective absorption of laser light and the results reported by the authors indicate the inactivation of *E. coli*. with the pulsed laser light producing the best results, likewise the bacteria dyed with safranin were inactivated at lower doses of laser light. The authors emphasized the correlation between the inactivation of the bacterium *E. coli* in relation to the absorbance, *e.g.* the rhodamine dye had a reduction at 180 kJ cm^{-2}, but when the higher absorbance dye (safranin) was used it required a dose of one-fourth that to produce the inactivation of *E. coli* (45 kJ cm^{-2}). Pulsed laser light reaches a temperature of 2 000°K, as compared to continuous light which only increases temperatures by 50°K which then stays constant during the exposure time.

The mechanism of the photothermal action of the laser interacting with the bacteria has been demonstrated by Tatsuno *et al.* (2021) who explained the inactivation process of the *E. coli*, through optical absorption and scattering effects. Because the incoming radiation is scattered by the bacteria the actual dose received by them is reduced. Thus, results show that the inactivation originates from the thermal destruction of the *E. coli* cells. The laser used was a YAG laser, different radiation wavelengths were selected (420, 500, 600 and 650 nm) with a pulse duration of 10 ns, a frequency of 10 Hz and a pulse energy of 10 mJ. The laser light was guided by a borosilicate glass microtube, and the samples were dyed with crystal violet. A convex lens with a focal length of 100 mm was used in the instrumentation for the radiation of the suspension. The power of the laser light source was 100 mW, and the power density was 50 W cm^{-2}. However, the obtained inactivation results did not correlate with the absorbance values of the dyed and non-dyed solutions. Upon further investigation, the researchers found that, even with the same irradiation length and energy density, continuous wave laser treatments did not change the colour of the colloid suspensions with gold nanoparticles. However, pulsed laser beam treatments significantly changed the suspension colour for all nanoparticle diameter sizes.

Gonca *et al.* (2023) investigated the effects of red, green, and blue laser diodes on pathogenic microorganisms such as *E. coli, Staphylococcus aureus* and *Candida albicans*. As a result of the study, the researchers found that the blue laser was more effective than the red and green lasers. They also reported an inhibition percentage for a 15 min treatment at a laser power density of 0.36 W cm^{-2} in *S. aureus* (65.9%), *E. coli* (34.52%) and *C. albicans* (43.63%), respectively. The blue laser inhibited microbial growth at 30 min; inhibitions of microbial growth ranged from 85.39, 41.18 and 54.55% for *S. aureus, E. coli* and *C. albicans*. The highest biofilm inhibition was 94.61% when *S. aureus* cells were exposed to blue laser irradiation for 60 min. The microbial growth kinetics of the three microorganisms were tested using a laser power density of 0.54 W cm^{-2} for 28 h, and no microbial growth was observed in *S. aureus* cells. Thus, laser-based methods seem promising for both the selective inactivation of various viruses and bacteria on different items containing them.

As we have seen, the effects of laser radiation depend upon various factors, and have associated thermal and optical components when interacting with organisms and microorganisms. It may be stated with considerable confidence that various researchers have found favourable results, under certain conditions of the medium being studied, oxygen availability, the characteristics of the biological object under study (optical and thermal characteristics), the type of light wave (continuous, pulsed) used, its intensity, dose, wavelength, and the artificial dye added or not added to the medium or to the organism and microorganism, among other considerations.

Dobrowolski (1996) conducted research with various microorganisms, bacteria, yeasts and fungi from different agri-food systems, agricultural soils, and tomato strains. He used a He-Ne laser (632.8 nm), Argon laser (514 nm) and diode laser (670 nm). The authors reported that irradiation with a diode laser (with an exposure time of 120 s) on the bacterial culture of *Arthrobacter globiformis* and *Bacillus macerans* (inoculated on a sandstone surface) destroyed the vitality of the bacteria to a significant extent. However, the laser treatment of infected tomato seeds did not destroy the phytopathogenic bacteria *Corynebacterium michiganense Jensen*.

Almuhayawi *et al.* (2021) applied He-Ne laser light (632.8 nm) to agricultural seeds, mustard, cauliflower, and turnip seeds were irradiated with the operating parameters of 5 mW of power and 500 mJ in intensity and the duration time of laser exposure was 5 min. In the instrumentation used for seed treatment, the laser light source was placed 12 cm from the seed on the embryo side. The results reported by the authors were that *Brassica sprouts* from the seeds treated with laser light showed a higher degree of antibacterial activity against foodborne pathogens (*Staphylococcus aureus, Escherichia coli, Listeria monocytogenes*, and *Salmonella* spp.). The effect was different for each seed with regard to its antibacterial activity against various bacteria, for example, mustard improved against bacteria *E. coli*, *Bacillus subtilis* and *Pseudomonas aeruginosa*; and cauliflower and turnip increased their antimicrobial properties against *E. coli, Bacillus subtilis* and *Pseudomonas aeruginosa*. The antimicrobial effects and secondary metabolite activities of the sprouts from the treated seeds were related between them. It is worth noting that the authors emphasized that turnip sprouts presented more evident responses to the positive effect of laser light as compared to the results obtained using cauliflower and black mustard sprouts. Also, turnip sprouts were shown to have a higher level of antioxidant and antiproliferative properties.

In relation to antiviral applications in agricultural systems, Hamrick and Cleary *et al.* (1968) reported the application of laser light on tobacco mosaic virus, which mainly infects tobacco crops and *Solanaceae*. In this experiment, laser-induced acoustic transients were applied by exposing monodisperse solutions of tobacco mosaic virus in an optically dense solution of blue dye to the ruby laser wavelength (694.3 nm). The laser light applied was pulsed at 50 ns and 1 J. The authors found that the laser power density required to break down the virus particles depended upon the conditions of the irradiated medium. In this way, alterations were produced in the biological system by breaking the molecular bonds. In 1983, the research group of Gregoraszczuk *et al*. (1983) observed differences in resistance to viral infection after the He-Ne laser irradiation of tomato biological material. They also reported that the amount of selenium in tomato fruit increased tenfold as a consequence of laser treatment of the seed (Dobrowolski *et al.*, 1997).

4.2. Fungi

Laser technology has also been confirmed by several studies as a sustainable strategy in agri-food systems for fungal control applications. Dobrowolski *et al.* (1997) demonstrated the beneficial effect of irradiation using diode (670 nm) and Ar (514 nm) lasers for fungal control by conducting *in vitro* experiments. The irradiation parameters applied were a power of 42 mW and exposure times of 120 and 180 s on fungal cultures *Aspergillus ochraceus, Chaetomium funicolum, Penicyllium lividum* and *Trichoderma polysporum.* It was shown that laser light produces deformations in fungi, so it may be a viable method for reducing the adverse effects of various mould species.

It has been established that photosensitizing the organisms or microorganisms is relevant to determining the appropriate irradiation dose and laser wavelength to use, as reviewed above. In the case of fungi, He-Ne lasers, laser diodes (650 and 660 nm) and 532 nm Nd-YAG lasers have been applied as fungicidal elements. The effects of laser light have been found to be enhanced by using photosensitizers (dyes in the seeds). There are several seeds in which the effect of laser light on their sanitary quality has been evaluated, among which soybean (*Glycine max* L.) has been one of the most studied. One of the most complete studies found in the scientific literature is by authors Ouf and Abdel-Hady (1999) who reported the effects of using the He-Ne laser (continuous light) with 7.3 mW of power for 1, 3, 6 and 10 min to irradiate soybean seed that was dyed and not dyed and placed in a rotating container to receive the laser radiation. The results that were reported at 3 min indicated a reduction in the number of seed-borne fungi and this reduction became more pronounced as the irradiation time increased. The pretreatment of the seeds with methylene blue, methyl red and carmine enhanced the fungicidal effect on fungi *Rhizoctonia solani, Alternaria tenuissima, Cercospora kikuclui* and *Colletotrichum truncatun,* which were eliminated when the seeds were irradiated for 10 min.

Moustafa *et al.* (2004) applied a semiconductor laser diode (630-660 nm) with an output power of 1 mW to rice seed with five irradiation times including the control (0, 15, 30, 45 and 60 s). The predominant fungi of the rice seed were *Pyricularia oryzae* and *Helminthosporium,* which were both affected in all seeds that received laser irradiation. Fungus of the genus *Helminthosporium* was eliminated after 45 s of radiation. Similarly, three isolates of *Fusarium* sp. that were obtained from the endosperm of non-irradiated seeds (control) were eliminated after 45 s. In plants from treated seeds, the authors observed different percentages of plant disease, a higher percentage was found in plants from untreated seed (86.6%) and this decreased in plants from seed treated for 45 s, the percentage of diseased plants decreased to 19.9%. Leaf severity was also reduced in the leaves of plants from seed treated with laser light. In the panicles of the seed treated plants no disease was found in contrast to the control plants. Laser light reduced the incidence of plant disease, treated plants were found to have a greater degree of resistance and less contamination by disease-causing fungi.

Other authors investigated one of the most important oilseed plants found in Poland, one which is exposed to potential risk from the fungal pathogen *Phoma lingam*. For this purpose, the effects of laser light in producing resistance to plant disease were investigated. A He-Ne laser (632 nm) with a 24 mW power output was applied. Three types of seeds were used (yellow seed strains, and two pollination varieties named “Bolko” and “Idol”) and four exposure times to laser light plus the control (0, 30, 60, 60, 90 and 120 min) were applied to *B. napus*. The researchers found that the type of seed variety with the highest number of plants resistant to the pathogen was the pollination variety (“Bolko”) at an exposure time of 30 min. However, in general terms the best results were found between the times of 30-90 min in the yellow seed varieties and “Bolko” and also in the “Idol” varieties at exposure times of 30 and 60 min (Starzycki *et al.*, 2005).

Other authors have reported the efficacy of laser light from diodes for quality improvement due to its effect on common fungi in wheat grain. The Nenadić *et al.* (2008) experiments were carried out by inoculating wheat seeds with *Penicillium, Fusarium* and *Alternaria* fungi at two infestation levels (high and low). The laser treatment consisted of irradiating samples using laser diodes with a low intensity of power (5mW) and different radiation times (3 and 6 s) under two laser regimes (2R, 3 s and 2R, 6s). The authors concluded that laser light was effective in reducing the presence of fungi. The optimal treatment was found to be 2 regimens and 6 s of laser light exposure. The percentage of fusarium fungus presence decreased from 25% (control samples) to 4% (samples treated with 2R, 6s). It is worth noting that in their experiment the authors compared the efficacy of their laser with a chemical fungicide (Vitavax), and found that the chemical treatment was more effective in that it resulted in 0% fungi relative to, for example, the 4% of fusarium left after treatment using laser light. However, although it is more effective, it is also more harmful, so the use of laser light may be a feasible way of reducing the use of chemicals.

Our research, which began in 2001, has concerned the effects of laser light on physiological and sanitary quality, this was accomplished by evaluating the effects of light radiation on seeds of common agricultural crops (Hernández-Aguilar *et al.*, 2005; 2006; 2008, 2009; 2011a). In addition, the seed was characterized using spectroscopic techniques (Hernández-Aguilar *et al.*, 2011b; 2015a; b; 2019) in order to determine the photoacoustic absorption and optical absorption coefficient of the seed under natural conditions and the seed was also dyed prior to laser treatment. Also, the temperature changes due to laser radiation have been studied.

In a study reported by Hernández-Aguilar *et al.* (2011a), a laser diode (655 nm) with a power output of 27.4 mW, five exposure times including the control (0, 60, 180, 300 and 600 s) and two levels of irradiation intensity (16.3 and 4.6 mW cm^{-2}) were applied. The seeds were photosensitized before treatment by laser irradiation with methyl red dye diluted in water and soaking the seeds were soaked in the diluted dye for 25 min. In this research, the effects of light on the mycoflora naturally associated with the seed were evaluated (*Fusarium* spp., *Aspergillus* spp., *Penicillium* spp. *Alternaria* spp. *Cladosporium* spp., *Rhizopus* spp., *Trichoderma* spp. and *Helminthosporium* spp.). The findings were that the percentage of seed infested with fungi was a function of the intensity of the laser light and the time of radiation applied. For all of the applied exposure times there was a statistically significant decrease in fungi as compared to the control samples (without radiation), it was found that the optimal exposure time was 300 s, for both of the applied radiation intensities. The infested seed (total fungi) and the seed infested by *Fusarium* sp. fungi decreased from 73 to 27 and from 65 to 23%, respectively.

In our investigations with photoacoustic spectroscopy which was used to characterize the coloured and uncoloured seeds we discovered the manner in which the absorption spectra and optical responses are modified by colouring the seed, *e.g.* with methyl red. In the studies reported by Hernández-Aguilar *et al.* (2005; 2011b) two conditions of corn (*Zea mays* L.) seeds, namely dyed and undyed with methyl red were established. The process of colouring the seed consisted of soaking them in water with the dye (1 g of dye/L of H_2O) for 25 min. The results were reported in terms of the optical penetration length ($l_\beta = 1/\beta$), the inverse of the optical absorption coefficient is a function of wavelength (λ). The seed without dye in the λ range evaluated (320-700 nm), had an optically opaque behaviour. However, the coloured seed (with methyl red) behaved in an optically opaque (320-620 nm) and optically transparent (621-700 nm) way as a function of λ. Thus, the l_β and β values vary according to λ. By colouring the biological object of study, l_β and β were modified. As a consequence, the effect which occurs when light interacts with the specimen is modified. The value of β was increased (11 cm^{-1}) relative to the corresponding value of the seed in its natural form (uncoloured) (2.3 cm^{-1}). This allows for a modification of the effects produced by the light this is brought about through defining the parameters of the applied radiation with the best effects leading to the desired results, depending on the characteristics of the object of study. In the case of the evaluation of the effects of low intensity red laser light on sanitary quality; it was observed, the percentage of infected seed decreases when laser light is applied prior to dyeing when compared to seed that was not dyed.

Another phenomenon that has been associated with the response of sanitary quality to light are the thermal changes produced in biological objects, which lead to changes in the moisture content. It has been established that the development of fungi in biological objects depends on several factors, humidity is one of them. Thus, laser light may produce thermal changes that modify such objects, and this may result in the deterioration of the storage conditions of seeds and/or grain. In studies carried out by Hernandez-Aguilar *et al.* (2015a; b) using corn it was found that laser radiation presented a temperature evolution when the light was incident and an involution when the laser light was turned off; depending on the colour of the seed and whether or not it was dyed. The maximum values reached depended on the colour of the seed. Both crystalline and floury seeds were used, which were dyed black. Laser light with a wavelength of 650 nm and a power of 27.4 mW was applied for 1 min to the maize seeds. The temperature changes reached were a function of the optical characteristics of the seed. Seeds that had a natural colour (without artificial colouring), underwent a temperature increase that was lower than that of the dyed seed. Between the two types of seed used in the study, crystalline and non-crystalline, it is possible to say that the highest temperature reached for 1 min was achieved by the non-crystalline seed (32°C), with an elevation of almost 10°C with respect to the initial temperature (before laser treatment). It is worth mentioning that the non-crystalline seed had a higher optical absorption coefficient than the crystalline seed and a lower optical penetration length. It has been established that the optical penetration length depends on the wavelength (Hernández-Aguilar *et al.*, 2015a). In this sense, laser irradiation may be viewed as an alternative treatment for the control of disease in corn seeds (*Zea mays* L.) or in other types of seeds and food, just so long as the appropriate operating conditions for the application of the laser light are chosen with regard to the appropriate characteristics of the target organism and microorganism. However, there is another detail to consider when dealing with biological systems: the container in which the object of study is placed and its characteristics.

Hernández-Aguilar *et al.* (2015b) studied the thermal changes occurring in seeds placed in different containers: seeds suspended in the air and those placed on aluminium material and on cardboard. The evidence found indicated that the seed suspended in air in the evolutionary stage (laser on) reached a higher temperature level (9°C above the initial temperature) when compared to the other seed conditions. In relation to the temperature decay stage, the seed placed on cardboard decayed more slowly. Therefore, the characteristics of the medium of where the biological object is placed are relevant since they modify the temperature changes (ΔT) in response to the laser light being turned on and off. This finding coincides with those of other authors who have associated insect extermination processes with certain optical and thermal components involving the interaction between laser light and biological objects.

Other research which has been conducted in the research group evaluated the effects of laser light on barley seeds (*Hordeum vulgare* L.). In this research, the seeds were in their natural form (without colouring) and dyed (with methylene blue). The laser used was a red laser diode (655 nm) which was applied during five exposure times (0, 60, 120, 240 and 480 s). As was the case with the corn seeds, in this research, at all exposure times there was a reduction in the number of seeds with naturally associated mycobiota (*Alternaria* sp., *Helminthosporium* sp., *Fusarium* sp., *Cladosporium* sp., *A. flavus*, *E. nidulans*, *A. terreus*, *Penicillium* sp., *A. niger, Rhizopus* sp.) in both dyed and undyed seed conditions. A greater reduction of mycobiota-infected seed was found in the coloured seed, for all exposure times. Although in the dyed seed the exposure time required to obtain the best result was reduced to 180 s, while in the non-coloured seed the best result was found at 480 s (Pérez-Reyes *et al.*, 2015).

Other research has been carried out on “pinto” beans in order to evaluate the effects of different wavelengths on the sanitary quality of the seed. In this way, by using diode lasers emitting three wavelengths 408, 532 and 650 nm, and with exposure times of 0, 3, 6 and 9 min and omitting to dye the seed, it was found that there were no significant statistical differences related to the mycobiota associated with the seeds, but there were significant differences related to the bacteria. The laser wavelength with which the best results were obtained was the red laser (650 nm) at 6 min of exposure, followed by the green laser (532 nm) at 3 min of exposure to reduce the bacteria found in the bean seeds (Sánchez-Hernández *et al.*, 2015).

Figures 1 and 2 summarize the results found in the research. Figure 1 shows the spectroscopic characterization and laser light radiation effects on maize and barley seeds. It is possible to observe this in Fig. 1a and b, the changes

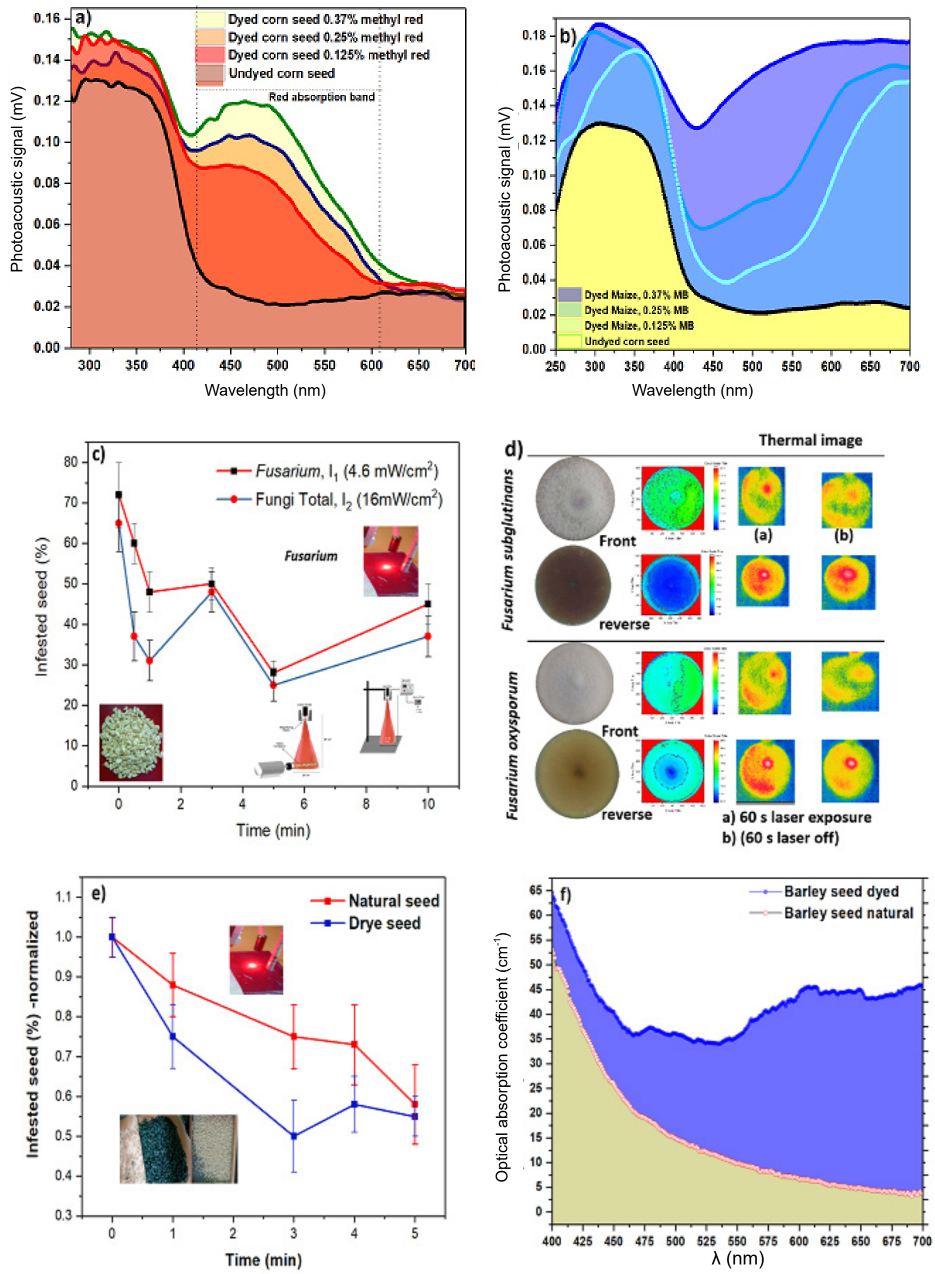

Fig. 1. Photoacoustic absorption spectra (PAS) and laser radiation effects in maize and barley seeds: a) PAS of maize dyed with methyl red dye, b) PAS of maize dyed with methylene blue, c) effects of red laser light (650 nm) on maize seed, d) effects of laser light on fungi (thermal images), e) effect of red laser light on barley seed, and f) effect of PAS on barley seed in its natural form and in its dyed form (methylene blue).

in absorption spectra due to the different typical dyes used as photosensitizer methyl red (Fig. 1a) and methylene blue (Fig. 1b), it may be observed how the addition of dye modifies the absorption of light at different dye concentrations (0, 0.125, 0.25, 0.37%). In this way, the effects of the radiation are modified as shown in Fig. 1c, the effects of the radiation on the quantity of seed infested by fungi are modified for both coloured and uncoloured seed conditions with the application of different exposure times of laser radiation.

The dyes produce changes in the absorption of the seed, as well as in its response to laser radiation, this also depends on the wavelength of the laser used. The response is modified when the study objects are coloured using artificial dyes, but also due to colour changes in the study object. Figure 1d shows the effects of laser light on *Fusarium* fungi, thermal images of this fungi are presented showing the effects of 60 s of irradiation with a red laser light. It was observed that the heating process was different depending on the colour of the fungus and the position of incidence of the laser light. At the point of incidence of the laser light, a different colour of the figure is reached, indicating a different level of heating of the object of study. Figure 1e and f, present the effects of laser light on the percentage of fungus infested seed and the photoacoustic absorption spectrum of the barley seed dyed with methylene blue and undyed (in its natural state). Seed colouring increased the fungal control effects on the barley seed.

Figure 2 a, b, c, and d show the micrographs at 1000x magnification of bean seeds soaked and treated with laser light. As can be seen the surface layer is affected by the treatment, this is apparent through observing the stereids or structural cells which are also known as the palisade, these are modified in the images Fig. 2a-d (control, irradiated with blue laser, irradiated with red laser and irradiated with green laser). This structure shows a thickening in the seed

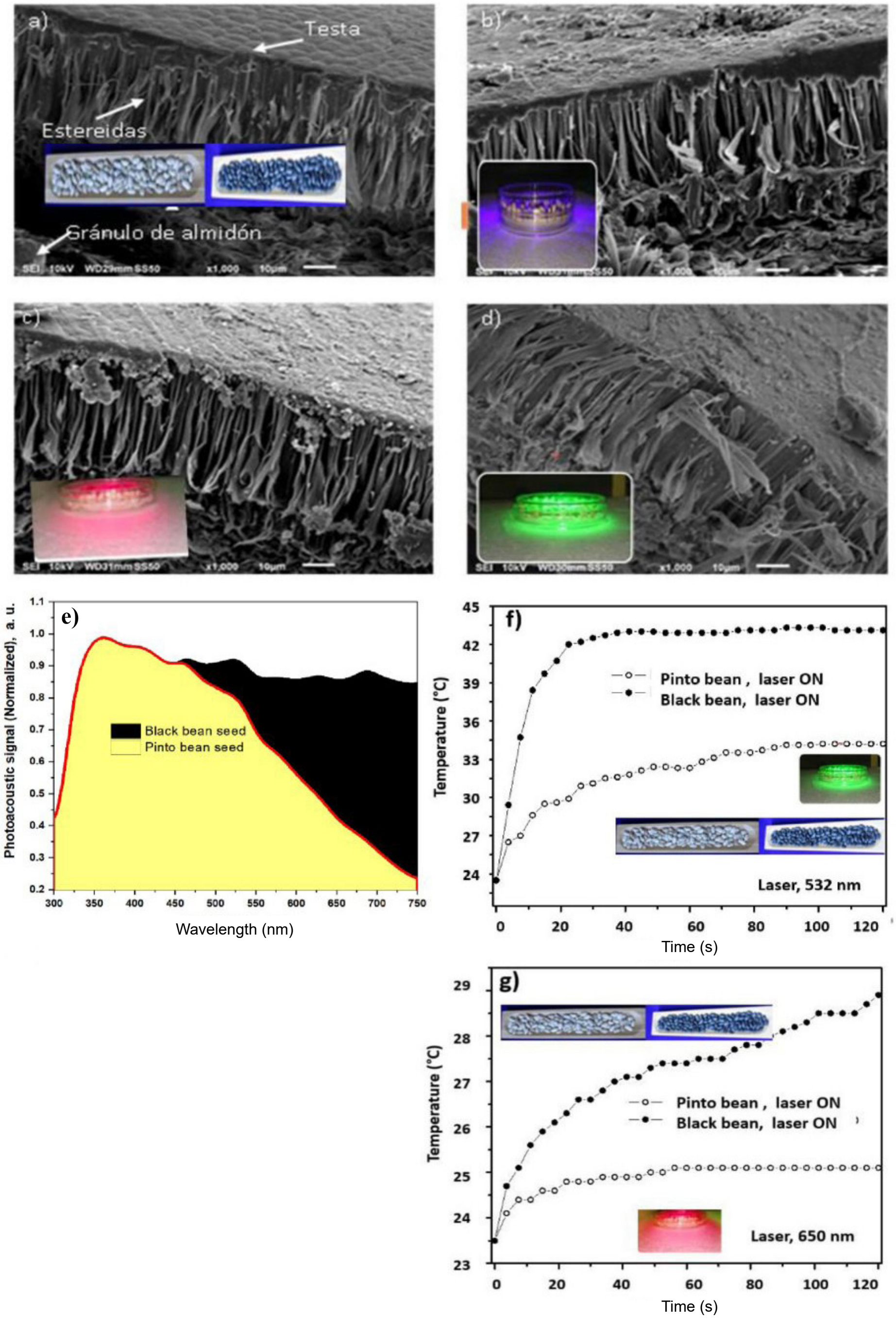

Fig. 2. Effects of laser light on bean seed: a) micrograph without radiation, b) micrograph radiated by a blue laser (408 nm), c) micrograph of seed radiated with a red laser (650 nm), d) micrograph of seed radiated with a green laser (532 nm), e) PAS of bean seed (pinto and black colour), f) thermal effect of two-colour bean seeds irradiated with a green laser (532 nm), g) thermal effect of seeds irradiated with a red laser (650 nm).

treated with a 532 nm diode laser light, while those treated at a wavelength of 408 and 650 nm are slightly thinner. The surface of the seed is also transformed; in the control sample there are capsules on the slightly rough surface, in the form of rhombuses. In the treatments, the capsules are not as noticeable as they are in the control, this may be due to thermal changes caused by the effect of the application of the diode laser light.

Thus, laser light has been reported to produce beneficial effects on seeds (pre-sowing stage) and the post-harvest stage. In this last stage, there are problems in several harvested products, one of the most relevant being strawberries. In the case of strawberry, the main cause of loss of quality and an appropriate appearance are gray mould caused by *Botrytis cinerea Pers*. and rotting by *Rhizopus stolonifer*. This has been reported by Ali *et al.* (2020), who used a 450 nm blue laser with an intensity of 1.3 mW cm^{-2} at three exposure times of 0, 3, 6 and 12 min. It was reported that the strawberry remained fresh until the fifth day according to the experimental conditions, thereby indicating a significant decrease in the deterioration rate caused by fungal microorganisms at 3 min of radiation exposure, although 12 min of exposure to radiation produced an inferior result as compared to the control samples. The control samples began to show evidence of the presence of fungi on the fourth day, as compared to the samples treated with radiation for 3 min, which showed similar signs on the seventh day. In addition, the authors also determined the ascorbic acid and anthocyanin content and antioxidant capacity. In the case of both ascorbic acid and anthocyanins, they had a tendency to increase as the days passed in all of the laser-treated samples, with greater differences in the case of anthocyanins, for example, on the second and fourth day the authors reported an amount of anthocyanins of around 45 mg 100 g^{-1} in the samples treated with 3 min of irradiation whereas in the control samples it was found to be around 35 mg g^{-1}. According to this publication, the antioxidant capacity decreased as time went on, but to a lesser extent in the laser-treated samples. For example, on the seventh day, while the laser-treated samples had an antioxidant activity of about 83%, the control samples had a value of around 72%.

A 532 nm Nd-YAG laser was used to irradiate wheat (*Triticum durum*) seeds under dry and wet conditions, they were kept in the dark for three weeks, Rassam *et al.* (2012) applied the laser treatment to control fungal infection in wheat seed and also to improve their growth and development. It was found that irradiation may be a viable alternative method for controlling fungal seed infection. Other studies have shown that a combination of Nd-YAG and diode lasers could have fungicidal effects on food, for example, light from 532 nm (Nd-YAG) and 660 nm (diode laser) lasers with powers of 100 and 120 mW, respectively, were employed for the purpose of eradicating *Aspergillus flavus* fungi, pistachios were inoculated with the fungus which became established in dry and humid conditions.

The samples received 0.5 J cm^{-2} for 7 days from each of the lasers and in combination, in doing so Saghafi *et al.* (2010) demonstrated that a combination of both lasers eradicates fungi from pistachios. The development of new irradiation approaches that can be used to eliminate harmful fungi from seeds, cereals, grains, and foods in general is especially useful in the case of the laser irradiation method. Unlike other methods such as the use of gamma rays or UV radiation, laser light does not cause changes in texture, nutritional content, and changes in physical appearance at low intensity levels and short exposure times. Thus, finding the right laser irradiation parameters to eliminate microorganisms from seeds and foods could benefit the health of the population and prevent the diseases of our times.

Another possible application of laser light within agri-food systems is in the maintenance of water quality. In agri-food systems, water may be a source of contamination and spread diseases to field crops, orchards, and greenhouses. Experiments with recycled irrigation water confirmed that UV Laser KrF excimer laser radiation (248 nm) with pulse operating characteristics of 20 ns, a pulse density of 1-2 mJ cm^{-2} and a power of 50-100 kW cm^{-2}/pulse is effective at inactivating the propagules of *Phytophthora* sp. (Pseudofungi). The doses of pulsed UV light applied were as follows: 0, 5, 10, 20 and 30 mJ cm^{-2}. With longer radiation times, the authors reported a higher level of disinfection of *Phytophthora capsici* (Banihashemi *et al.*, 2010).

In this way, it was found that there are several applications of laser light to ameliorate various problems of agri-food systems. In addition to this, there is also the ability of laser light to potentiate active elements such as fungicides or bactericides.

5. LASER BEAMS AS ENHANCERS OF ACTIVE ELEMENTS

Laser beams can be useful not only in the control of fungi directly in isolation or naturally in the case of agricultural seeds, water, soil, food, etc., but they can also be used to modify the active elements of the fungus itself (Geweely *et al.*, 2011) and those of plants with fungal properties (Aladjadjiyan, 2007; Ali *et al.*, 2022; Al-abedi *et al.* 2023) or indeed those of bactericides (Balkhyour *et al.*, 2021; El-Adly *et al.*, 2007; Zrig *et al.* 2022; Okla *et al.* 2022), it has been reported that laser light could be used to enhance their properties. However, when they are used for this purpose, it is still of interest to define the appropriate radiation parameters and also the optical and thermal characteristics of the biological element in question. The authors have tested their effects on different types of fungi at different exposure times using a laser diode. It has been shown that the effects of each biological element vary for the same irradiation parameters and experimental conditions. Geweely *et al.* (2011) reported this in their study designed to potentiate the pigments of different types of fungi. According to their research, the authors reported that the control of certain fungi (A. *nidulans, F. moniliforme, P. Purpurogenum, P. herbarum*) occurred as a function of the applied exposure times. The laser light eliminated the evaluated fungi at an exposure time of 15 min. The fungus *Penicillium purpurogenum* was found to be the most resistant to laser light and the main pigment producer among the fungi evaluated. These possible differences were associated with different pigment components such as melanin which produce different laser light absorption capacities.

The potential application of laser light to induce better properties in the study object has been reported for example in the ajwain, *Trachyspermum* (*T.*) *ammi*. He-Ne (632 nm) and He-Cd (460 nm) lasers with a power of 5 and 16.2 mW were employed at exposure times of 5 min and the distance between the laser light and the seed was 12 cm. The authors report that laser treatment significantly increased the antibacterial properties of ajwain sprouts against food pathogens: *Staphylococcus aureus, Listeria monocytogenes* and *Bacillus monocytogenes, Bacillus cereus* and *Salmonella* spp. The effects depended on the type of laser used, the seed species and the type of bacteria. It was found that the He-Ne laser influenced antibacterial activity, it was improved to a statistically significant extent in the ajwain species (*T. ammi, Chakwal*) which was evaluated against *Staphylococcus aureus* (*T. ammi*) and *Listeria monocytogenes* (*Chakwal*). The properties of the sprouts that were improved using laser light were coumarins and secondary metabolites which are reported to be antibacterial, antiviral, and antifungal (Balkhyour *et al.*, 2021).

The effects of laser light combined with 6-benzylaminopurine for the conditioning of foods such as sprouts have been investigated and, due to promising results, have been considered in a proposal to improve nutraceutical values and also to improve sanitary quality. Zrig *et al.* (2022) reported that they can be used to modify antimicrobial activity and bioactive compounds in flax seed sprouts (*Linum usitatissimum*). A more pronounced effect was achieved when combining the two conditioning treatments as compared to using them independently. It should be noted that secondary metabolites were increased for some compounds with laser irradiation alone. Increased levels of bioactive metabolites such as the phenolic and fatty content in flaxseed shoots in response to laser light have been attributed to antibacterial enhancement. It should be noted that the presence of phenolic compounds, as well as fatty acids and lignans, are essential for the antibacterial activity of *L. usitatissimum,* in this way the use of laser irradiation could be used to potentiate their antibacterial power.

Potentiating the bacterial activity of bioactive compounds and improving food quality allows for the preservation of shelf life in fresh foods. Thus, irradiation with laser light has been reported to potentiate nutraceuticals and improve the quality of products that would allow for the preservation of food quality. Laser treatment on freshly cut products has been reportedly used for this purpose directly. Wen *et al.* (2023) recently reported studies demonstrating the potential of light from laser diodes to preserve the quality of fresh produce (*Solanum tuberosum*). The authors applied two wavelengths of diode lasers, at 450 and 660 nm using five exposure times (0, 10, 20, 30, and 40 min) and they placed the laser at a separation distance of 30 cm from the potato samples. It was found that by using the blue laser (450 nm) at an exposure time of 30 min, the quality of the sliced potato is maintained for a greater number of days. In colorimetric tests, fewer differences in colour were found between the samples at the beginning of the study and several days after storage in potatoes treated with laser light in relation to untreated potatoes, thus the extension of their shelf life was evident. The antioxidant activity was also increased and thus the laser light maintains the quality of freshly pre-cut produce.

6. INTERACTION BETWEEN PLANTS/MICROORGANISMS AND LASER TECHNOLOGY

The interaction between plants and microorganisms that were stimulated with laser technology has been reported in several studies. Thus, the application of laser technology to plant inoculants for the purpose of facilitating plant growth and the development and enhancement of disease bio-control is another promising area for the application of this technology. Clover seeds have been incubated using *Rhizobium* bacteria treated with He-Ne laser radiation (632.8 nm) and 8 mW cm^{-2} at exposure times of 5, 10 and 15 min. The best results that were achieved in terms of plant growth variables such as height, root, dry weight and the number of nodules were found after 5 min of laser exposure (El-Raouf Ahmed *et al.*, 2023). In other studies using laser radiation with different exposure times being applied to *Rhizobium* inoculants; the authors have pointed out the importance of defining the appropriate radiation times for the germination of clover seeds (Abd El-Raouf *et al.*, 2023). It was found that the longer the laser exposure time of the *Rhizobium* bacteria, the lower the germination percentage. This highlights the importance of defining the radiation parameters when laser applications are made in agriculture for different purposes, as was pointed out in the scientific literature.

Maslova *et al.* (2020) studied the biocontrol of cucumber plant pathogens *in vitro* using He-Ne and semiconductor lasers applied to the antagonistic bacteria *Bacillus subtilis* (BsP) and *Pseudomonas fluorescens* (PfV) in order to enhance the production of metabolites. The radiation parameters of the lasers were in the power range of 2-2.5 mW cm^{-2} and had exposure times of 15, 30, 60, 120, 240, 480, and 960 s. The authors found that laser radiation increased the number of cells in the bacterial suspensions by 120 and 80% at 240 and 480 s for PfV and BsP respectively. A greater increase in the photosynthetic activity of cucumber seedlings *Cucumis sativus* was also related to a higher concentration of the applied bacterial metabolites.

Other laser treatments on fungi such as an arbuscular mycorrhizal fungi (AMF) inoculum have been used. Okla *et al.* (2022) found that the application of laser light together with a mycorrhizal inoculation on *Pelargonium graveolens* significantly improved plant biomass and nutrient content. The treated samples also showed an increase in the amount of essential oils and secondary metabolites as compared to the untreated samples.

In addition, laser light and mycorrhizae increased antibacterial activities and reduced the incidence of brown spots caused by the *Septoria* fungus. Thus, the combination of laser light and mycorrhizae increases both shrub health and nutritional effects. Similarly, studies reported by Dłużniewska *et al.* (2021) indicate that seed stimulation and inoculation with AMF reduced the incidence of *Septoria* leaf spot. However, there was only a slight reduction in root rot by *fusarium* fungus.

Laser light may be applied to micro-organisms such as bacteria and fungi that benefit plant growth; this strategy could be important in the practical application of plant growth and disease control for agricultural production systems in the open field and in greenhouses, and also to enhance the process of germination which mainly occurs under abiotic stress conditions. The synergy of these physical and biological methods forms a part of the trend for the improvement of agri-food systems, with both of them being both ecological and sustainable approaches.

Tables 1 and 2 summarize the advances that have been made in the effects of laser light, these have been documented in the present literature review, and reflect the scientific findings occurring in different parts of the world over time. This work documents the beginnings of the application of lasers for the control of organisms and microorganisms to the work being carried out in the present day. It presents the effects of high and low intensity pulsed and continuous laser light on organisms such as insects and microorganisms such as bacteria, fungi and viruses and also as a strategy to potentiate metabolites and the antimicrobial activity of plants. These results are presented to confirm the possible use of laser irradiation as a pesticide in agri-food systems as a viable strategy which may be used to attain sustainability as long as the research is carried out in a transdisciplinary way to approach the problems of each community, region, climate, soil, *etc*. which each have their own specific problems with regard to the type of organisms and microorganisms which must be controlled.

Figure 3 shows the number of papers found in this review of literature. These are classified according to the UVC, UV-B, UV-A, blue, green, red, NIR, MIR and FIR regions of the spectrum and the control attained over insects (a), bacteria (b), fungi (c), and viruses (d), respectively.

Also, a number of published articles are related to the applications of laser light increasing, *e.g*. those related to the level of metabolites and antimicrobial activity (Fig. 3e) and in general, the number of papers published in the 1960´s to the present day in the topic of laser light applied in organisms and microorganisms has been increasing (Fig. 3f). It may be observed that the most studied microorganism category was fungi, and the least studied was viruses. According to the literature reviewed, it is possible to observe in Figure 1f that the studies reported which were carried out for the benefit of agri-food systems began in the 1960s, with applications directed at the control of various organisms. Although studies involving laser light began with bacteria, applications designed to benefit agri-food systems began to boom in the 1990s, with one of the microorganisms of greatest interest being fungi. Insect control is one of the serious problems in the sector, at present it involves the use of agrochemicals for many agri-food systems with all of their inherent risks. Thus, intervening in such systems with proposals such as the use of lasers will be useful for the years to come.

7. DISCUSSION AND FUTURE PROSPECTS

According to the literature review, it has been found that lasers have favourable effects which allow for their use in agri-food systems for the control of organisms and microorganisms. In the case of insects, the technology is relevant since insects adversely affect the development of crops and the agrochemicals that are used to control them harm both farmers and the environment. The insects that are targeted for extermination are found among crops and in the soil, flying insects and other insects are found in leguminous plants or in stored grain. The wavelength regions of laser light that can be applied range from the UV-C region, blue visible, to the far infrared region (FIR), laser light in the visible region (blue, green and red light) has also been used in pulsed or continuous mode, while in the UV-C, NIR, MIR and FIR range it was only used in pulsed mode. In general, the laser light is applied by emitting a pulsed light wave, while the visible light wave (blue, green and red light) is emitted by lasers that produce light in both a pulsed and continuous form, with the continuous wave being more predominant. The main insects that have been shown to be vulnerable to laser light are: a) UV-C light has been used to combat mites (*Tetranychus urticae*); b) by means of blue light *Anopheles stephensi*, larvae, B*emisia tabaci*, *Tribolium castaneum* and *Trogoderma granarium;* c) by green light *Anopheles stephensi,* aphids (*Acyrthosiphon and Rhopalosiphum padi),* whitefly*, Bemisia Tabaci,* weevil Khapra (*Trogoderma granarium*), ants (*componotus consobrinus*); d) by red light: mites, greenhouse whitefly, green aphid, ant, fungus gnats, greenfly, fungus gnats (Diptera: Sciaridae); e) MIR and NIR mosquitoes *Anopheles stephensi*, cowpea beetle and f) the FIR range of light has been used to combat mosquitoes *Anopheles stephensi* and aphids (*Acyrthosiphon* and

Table 1. Laser in the control of insects in agri-food systems

Laser type	Operating characteristics	Insect	Affecting system	Reference
		Continuous wave laser		
Laser diode	444, 527, and 640 nm 1 s, 10 W cm^{-2}; 3 s, 4 W cm^{-2} and 4 s, 4 W cm^{-2}	White fly (*Bemisia tabaci*)	Bean cultivation (*Phaseolus vulgaris* L.)	Zaidem *et al*. (2023)
Laser diode	532 nm 200 mW 10, 15 and 20 s	Ants (*Camponotus consobrinus*)	Live in the soil Predators that can eat grains	Rashid *et al*. (2021)
Semiconductor Laser	450 nm 10 W 0.5, 1, 2 and 3 s	Larvae (*P. rapae antifeedant*)	Pests in cruciferous plants	Li *et al.* (2021)
CO_2 and others	532 nm, 1.070 nm and 10.6 µm (<100 ms) 0, 20, 40, 60, 80 and 100 mJ cm^{-2}	Aphids (*Acyrthosiphon pisum*) (*Rhopalosiphum padi*)	Plants (*Vicia faba,* L. *cv. Aquadulce, Triticum aestivum Linnaeus*)	Gaetani *et al*. (2021)
Laser model (supetek)	405 nm 110 W (0, 10, 20, 30 and 40 min)	*Weevils:Tribolium castaneum* and *Trogoderma granarium*	Stored flours	Ubaid (2016)
Diodo Laser	532 nm 30, 45, 60 and 75 s	Beetle: Khapra, *Trogoderma granarium* (*Everts*) *Coleoptera Dermestidae*	Stored cereals	Amaar and El-Refai (2015).
		Pulsed laser		
Laser Diode Integrated UV-A LED light trap	445 nm 10 s each 200 ms	Fungus gnats (*Diptera: Sciaridae*)	Mushroom and mushroom crops	Nair *et al*. (2023)
Excimer lasers (UV-C)	248 nm 1 to 4 min with dose 5-80 kJ m^{-2}	Mite (*Tetranychus urticae*)	Plants in citrus orchards	Gala *et al*. (2021)
Nd:YAG Laser	1 064 nm 10, 20, 30 s 5 pulsos /s (300, 360, 420 and 480 nm)	Cowpea beetle	Legume seeds such as chickpea, *etc*.	Rashid *et al.* (2018)
Diodo laser	445 nm 670 mW (< 25 ms)	*Diaphorina citri* (1), and *Anopheles stephensi* (2)	Citrus crops (1) and a malaria vector (2)	Mullen *et al*. (2016)
CO_2 and others	Green laser (532 nm), FIR laser (10, 600 nm), NIR and MIR (976, 1064 and 1470 nm) 2-45 ms	Mosquitoes (*Anopheles stephensi*)	Agricultural production systems and carriers of malaria	Keller *et al.* (2016)
Ruby laser	694 nm	Mites: *Panonychus ulmi* and *Tetranychus urticae*; Greenhouse whitefly *Trialeurodes vaporariorum*; Green alphid: *Mysus persicae* and *Psylla pyricola*	Greenhouse host plants: pear (*Pyrus communis*), (*Medicago sativa*) and rye grass (*Bromus* sp.)	Wilde (1967)

Table 2. Laser used in the control of microorganisms in seeds and food

Object of study	Experimental criteria	Radiation parameter	Significant Findings/Results	Reference
Radiation source: He-Ne laser				
Fungi				
Seed Soybean (*G!ycine max* L.)	Seed natural and dyed with methylene blue, methyl red and carmine	λ: 632.8 nm, P:7.5 mW, t: 1, 3, 6 and 10 min	↓ Number of seed-borne fungi at 3 min of irradiation. Pretreatment of the seeds with methylene blue, methyl red and carmine enhanced the laser effect. *Rhizoctonia solani, Alternaria tenuissima, Cercospora kikuchii* and *Colletotrichum,* at 10 min.	Ouf and Abdel-Hady (1999)
Seed: red clover (*Trifolium pratense* L.)	Dressings: Funaben, Sarfun and Super-Homai	λ: 632.8 nm, P:40 mW I: 0, 3, 6 mW cm^{-2}, t: 0.1 s. 1, 3 and 5 irradiation rounds	↓ *Phoma* at intensities of 3 mW cm^{-2} x 1 and 3 mW cm^{-2} x 3. ↓ *Penicillium* at 6 mW cm^{-2} x 1 and 6 mW cm^{-2} x 3. ↑ *Alternaria* at 3 mW cm^{-2} x 3; 3 mW cm^{-2} x 5 and 6 mW cm^{-2} x 5.	Wilczek *et al.* (2004)
Seed: alfalfa (*Medicago sativa* L ssp. sativa)	Martin's substrate was used to identify the mold strains. Fungal mycelium developed was transferred to PDA culture medium	λ: 632.8 nm, P: 40 mW I:3, 6 mW cm^{-2}, t: 0.1 s. 1, 3 and 5 irradiation rounds	↓ *Penicillium* fungus: in 3 and 5 radiation cycles was eliminated. ↑ *Alternaria* fungus; in 1 radiation cycle the fungus was stimulated (in seeds coated with Super-Homai).	Wilczek *et al.* (2005)
Seeds: (*Brassica napus*)	Infection severity test was applied	λ: 632 nm, P:24 mW, I:1 mW cm^{-2} t: 0, 30, 60, 90 and 120 min	↑ Higher resistance to *P. lingam*-inoculation were in 30-90 min.	Starzycki *et al*. (2005)
(*Trifolium pratense* L.)	A day before seeding was applied the radiation laser.	λ: 632.4 nm, I:40 mW, t: 0.1 s	↓ Fungal diseases in the seeds infected	Ćwintal and Sowa (2010)
Product: Brown rice snack bars	1 mL of the *Aspergillus flavus* spore suspension was added onto the surface of the brown rice snack bars. *Litsea cubeba* vapor and helium-neon (He-Ne) laser was applied	λ: 633 nm, P:7 mW, t: 1 min	↓ Growth of *Aspergillus flavus.* ↑ Shelf life, from 10 days control to 25 days with the hybrid treatment applied. laser treatment without essential oil vapor was ineffective against mold growth. mold growth.	Suhem *et al.* (2015)
Fruit: Caraway (*Carvi fructis*)	The caraway fruits were irradiated in the dark	λ: 632.8 nm, P:30 mW, t: 1, 3, 5, 10 and 15 min	↓ Elimination completely of fungi spore. ↑ Essential oil %, ↑ Germination %.	El-Raie *et al.* (2017)

↓: Decrease, ↑: increase; λ: wave length, P: emitting source power; t: exposure time to laser light, I: intensity.

Table 2. Continuation

Object of study	Experimental criteria	Radiation parameter	Significant Findings/Results	Reference
Radiation source: He-Ne laser ↓: Decrease, ↑: Increase				
Fungi				
Seed: Soybean (*Glycine max* L.)	Not discoloration, mycelium deposits and insect or mechanical damage	λ: 632.8 nm, I: 2 mW m^{-2} t: 3 times x 9 s	↑ Microorganisms as *Botrytis cinerea, Aspergillus flavus* and *Rhizoctonia solani.*	Klimek-Kopyra *et al.* (2020)
Product: *Vanilla planifolia* Jacks	Susceptibility to *Fusarium oxysporum*.	λ: 632.8 nm, I:2 mW m^{-2} t: 30, 60, 90, 120, and 150 s	↑ Highest survival rate at 60 s, Laser can be a promising alternative to induce a "hormetic effect".	Fernández-Valdez *et al*. (2023)
Diseases				
Seed: *Brassica napus*	Seedlings inoculated with P. *lingam*	λ: 632 nm, P: 24 mW I: 1 mW m^{-2} t: 30, 60, 90, 120 min	↑ Higher resistance to *Phoma lingam*-inoculation were in 30-90 min.	Starzycki *et al.* (2005)
Bacterial				
Seeds: tomato	Seeds were disinfected with 1 per cent solution of formaldehyde to destroy bacteria	λ: 632.8 nm, Power: 30 mW t: 15-375 s	Laser treatment of infected tomato seeds did not destroy phytopathogenic bacteria *Corynebacterium.*	Dobrowolski *et al.* (1996)
Seed: mustard, cauliflower, and turnip (*Brassica* cultivars)	The seeds were soaked in distilled water for 2 h.	λ: 632 nm, Power: 5 mW t: 5 min s	Mustard Sprouts ↑ Antibacterial activity against *E. coli, Bacillus subtilis* and *Pseudomonas aeruginosa*. Cauliflower and turnip sprouts ↑ Antibacterial activity against *Salmonella* spp. and *Sarcina lutea*.	Almuhayawi *et al*. (2021)
Radiation source: Diode laser				
Fungi				
Seeds: Rice (Giza 159 cv.)	Seeds were not colored. Some tests were carried out with isolated fungi.	λ: 630-660 nm, P: 1 mW, t: 0, 15, 30, 45, 60 s	↓ *Pyricularia oryzae*, *Helminthosporium,* and *Fusarium* sp.	Moustafa *et al.* (2004).
Grain: Corn (*Zea mays* L.)	Seeds were dyed before the irradiation. by soaking in red methyl for 25 min disinfected for 3 min in a 10% solution of sodium hypochlorite and rinsed with distilled water.	λ: 655 nm, P: 27.4 mW, t: 0, 30, 60, 180, 300, 600 s	↓ Total Mycoflora At all times, the percentage of seed infected by any type of pathogen, decreased when comparing the results with the control samples. The time of radiation had greater differences than the intensity of radiation, the best time for both intensities was 5 min.	Hernández Aguilar *et al*. (2005)

↓: Decrease, ↑: increase; λ: wave length, P: emitting source power; t: exposure time to laser light, I: intensity.

Table 2. Continuation

Object of study	Experimental criteria	Radiation parameter	Significant Findings/Results	Reference
Radiation source: Diode laser↓: Decrease, ↑: Increase				
Fungi				
Seed: Wheat	Pre-treatment of grain	λ: 627 nm, P: 0.95 mW, t: 0, 2, 5, 6, 10 s	↓ Laser light dries the grains and reduces moisture. ↓ Significantly reduces the number of seed-born fungi, such as *Penicillium, Fusarium* and *Alternaria*. The pretreatment of grain is a very important process for its subsequent storage.	Franjo Jović (2006)
Seeds: Maize (*Zea mays* L.)	Seeds dyed by red methyl	λ: 655 nm, P: 27.4 mW, t: 0, 30, 60, 180, 300, 600 s	↓ More than 60% of seeds infected with *Fusarium* sp. fungi and total fungi (*i.e.*, any genus of fungi). Treatments at 300 s recorded the lowest percentage of *Fusarium*-infected seeds.	Hernández-Aguilar *et al*. (2011)
Seed: Barley (*Hordeum vulgare* L.)	Seed condition: natural and dyed with methylene blue.	λ: 650 nm, P: 27.4 mW, t: 0, 60, 120, 240, 480 s	↓ Associated natural mycobiota, in both conditions, finding the highest percentage of decrease in the dyed seed. The best time was at 2 min, followed by 3 and 5 min.	Pérez-Reyes *et al*. (2015)
Fruit: Strawberry (*Fragaria* × *ananassa*)	Strawberries were washed with tap water (to remove dust) and then immersed in distilled water for 30 min.	λ: 450 nm, P: 100 mW t: 0, 3, 6, 12 min	↓ Fungal at 3 min ↑ Shelf life	Ali *et al*. (2020)
Radiation source: Argon laser				
Fungi				
Seed: Soybean (*Glycine max* L.)	Seed were well developed, not discoloration, mycelium deposits and insect or mechanical damage.	Laser (Argon) λ: 514 nm, I: 5 mW m^{-2} t: 3 times x 3 s	↑ A significant increase in the frequency of occurrence of *Aspergillus flavus* ↓ The saprotrophic fungus *Rhizopus nigricans.* ↓ *Gliocladium roseum.*	Klimek-Kopyra *et al*. (2020)
Disease resistance				
Radiation source: SHG Nd:YAG laser				
Seed: Hard wheat (*Triticum durum*)	Two groups of irradiated seeds (dry and wet).	λ: 532 nm, P: 20 mW t: 0, 1, 5, 10, 15 min	↑ Resistant to fungi infection. Modifies resistant to fungal infection of hard wheat seeds when treated in wet condition.	Rassam *et al*. (2012)

↓: Decrease, ↑: increase; λ: wave length, P: emitting source power; t: exposure time to laser light, I: intensity.

Table 2. Continuation

Object of study	Experimental criteria	Radiation parameter	Significant Findings/Results	Reference
Radiation source: Kr-F laser (Excimer UV laser) ↓: Decrease, ↑: Increase				
Fungal				
Food: Fruit (apple, Kiwi, oranges, Lemons, Nectarines. Peaches, Pears, Raspberries, Table grapes)	Fruits were inoculated with different fungus	λ: 248 nm, P: 20-40 mW E (per pulse): 0.1-100 mJ cm^{-2} pulse: 20 ns f: 100 Hz	↓ *Alternaria alternata, Botrytis cinérea, Fusarium oxysporum, Fusarium roseum, Monilinia fructicola, Penicillium expansum, Penicillium digitatum, Phytophthora citrophthora, Rhizopus stolonifer*. Fungi on fruit were controlled with less than 500 mJ cm^{-2}, except for *Aspergillus fungus*, which required a higher energy density level to be controlled (1900 mJ cm^{-2}).	Lagunas solar *et al*. (2006)
Bacteria				
Food: Goat milk	Aliquot of 1 ml of inoculant (*E. coli*) was added to the 50 ml milk samples. 3 irradiation regimes were applied	λ: 248 nm, E (energy per pulse) = 410 mJ t: 1, 8 s.	↓ 6 log reduction of the initial microbial population (*Escherichia coli*)	Kasahara *et al*. (2014)
Food: Milk	Culture strains: *E. coli*, *Salmonella*, Aureus, among others were used to inoculate	λ: 248 nm E: 0.5 to 2 mJ cm^{-2} t: 1, 5, 7, 14, 28, 56, 114 s	The bacterial content of the milk can be adequately controlled.	Smith *et al.* (2002)
Radiation source: CO_2 laser				
Fungi				
Vegetal: carrots	Samples were left at room temperature for observation on days 0, 2, 5, 10, 10, and 18 days	λ: 10.6 µm E: 1 kW t: 4 s	↓ Decreased mold growth on carrots. ↑ Shelf life increased.	Watson *et al*. (2007)
Fruit: Tangerines (*Citrus reticulata* Blanco)	*Penicillium digitatum* spores were applied to the surface of the fruit before and after etching it	λ: 10.6 µm t: 45 µs	↓The amount of *penicillium* spores was reduced	Sood *et al.* (2008)

↓: Decrease, ↑: increase; λ: wave length, P: emitting source power; t: exposure time to laser light, I: intensity.

Table 2. Continuation

Object of study	Experimental criteria	Radiation parameter	Significant Findings/Results	Reference
Radiation source: CO_2 laser ↓: Decrease, ↑: Increase				
Bacteria				
Vegetal: Carrots and potatoes	Carrot and potato discs were sterilized, completely and partially dried and inoculated with *E. coli* bacteria.	λ: 10.6 µm E: 1 kW t: 2-10 ms	↓ *E. coli* At the 10 ms exposure time, a reduction of 5 Log and 3 Log occurred for the partially dried and dried samples.	Watson and Stewart-Tull (1999)
Fruit: Orange peel	Fruit were packed following standard commercial protocol, which included waxing with 5 ppm thiobendazole. Fruit stored at 10°C.	λ: 10.6 µm, E: 0.578 W t: 35 µs	↓ 2.4 log cycles of inactivation (*Salmonella spp.*)	Danyluk *et al.* (2013)
Fresh fruit: Apple peel	Samples of 1 cm^2, which were contaminated with *E. coli.*	λ: 10.5 µm t: 60, 120, 180 s	↓ Inactivation of *E. coli* K12 with minimal damage to fruit surface	Chee *et al.* (2015)
Radiation source: Hybrid methods				
Seed: cucumber (*Cucumis sativus*)	Seeds of the maternal line (G-3) of the cucumber hybrid cultivar *Pobeda* were used.	λ: 632 nm Power: 20 mW laser + gamma rays Dose:10 Gy	↑ Quality	Cholakov *et al*. (1996)
Seed: pistachios	Cultures of *Aspergillus flavus* were grown and stored on pistachios at 23ºC.	Dose: 0.5 J cm^{-2} λ: 532 nm (Nd:YAG) Power: 100 mW λ: 660 nm, Power: 120 mW	↓ *Aspergillus flavus* Combination of a green and a red laser beam optimal effects on pistachio mold fungus eradication.	Saghafi *et al*. (2010)
Seed: Soybean (*Glycine max* L.)	Seeds treated with Micorrizal inoculum. The order of application of lasers was Ar first, followed by He-Ne.	(Argon) λ: 514 nm, I: 5 W m^{-2} (He-Ne) λ: 632.8 nm, 2:5 W m^{-2} t: 120,180 s	↓ Average *Fusarium oxysporum* disease index in the plants.	Dłużniewska *et al*. (2021)

↓: Decrease, ↑: increase; λ: wave length, P: emitting source power; t: exposure time to laser light, I: intensity.

Table 2. Continuation

Object of study	Experimental criteria	Radiation parameter	Significant Findings/Results	Reference
Radiation source: Hybrid methods ↓: Decrease, ↑: Increase				
Fungi:	Fungi treated in insolation	Diode Laser λ: 670 nm, P: 5 mW Ar laser λ: 514 nm	↓ Fungi *Aspergillus ochraceus, Chaetomium funicolum, Penicyllium lividum* and *Trichoderma polysporum*	Dobrowolski *et al.* (1997)
Laser for the increase of secondary metabolites and antimicrobial activity				
Seeds: Ajwain (*Trachyspermum ammi*)	Seeds were soaked for 2 h in distilled H_2O	Laser He-Ne λ: 632 nm P: 5 mW, t: 5 min	↑ Levels of phenols, flavonoids, coumarins ↑ Increased antibacterial activity	Balkhyour *et al.* (2021)
Volatile Oil: Fennel (*Foeniculum vulgare* var. dulce)	Photosensitizer (toluidine blue)	Laser He-Ne λ: 632 nm P: 7.3 mW, t: 5, 10, 15 min.	↓ Decreases microbial activity (*Escherichia coli, Serratia marcesence, Klebsiella pneumoniae*). Photosensitizing dye and essential oil are inactivating bacterial.	El-Adly *et al.* (2007)
Seed: Linseed (*Linum usitatissimum* L.)	Nutrient solution was supplied at the beginning of experiment	Laser He-Ne λ: 632 nm P: 5 mW, t: 5 min.	Laser He-Ne + 6-Benzylaminopurine ↑ Increased antimicrobial activity. ↑ increased antioxidant capacity, fat acids, phenols, and flavonoids	Zrig *et al.* (2022)
Plant: *Pelargonium graveolens*	Sterilized seeds, co-cultivated with *Rhizophagus*	Laser He-Ne λ: 632 nm P: 5 mW, t: 5 min. + Mycorrhiza Inoculated	↑ Enhance the phytochemical content (phenol, flavonoids, alkaloids). ↓ Inhibition of *E. coli* , *Streptococcus salivarius*	Okla *et al.* (2022)
Fruit: Strawberry (*Fragaria* × *ananassa*) (*Fragaria* × *ananassa*)	Laser light followed by nanoparticles treatment), then stored for 12 days at 10°C and 85 to 90% RH.	Diode laser λ: 450 nm P: 100 mW I: 1, 3 mW/cm^2 Laser + guava leaf-based chitosan nanoparticles	↓ Combined treatment eliminates fungus.	Ali *et al.* (2022)
Leaves Extract (*Eucalyptus spp.*)	Leaves was washed with tab water and distilled.	Semiconductor Laser λ: 450 nm P: 50 mW, t: 20 min.	↑ Enhances antifungal activity of chloroform extract from eucalyptus.	Al-abedi *et al.* (2023)

↓: Decrease, ↑: increase; λ: wave length, P: emitting source power; t: exposure time to laser light, I: intensity.

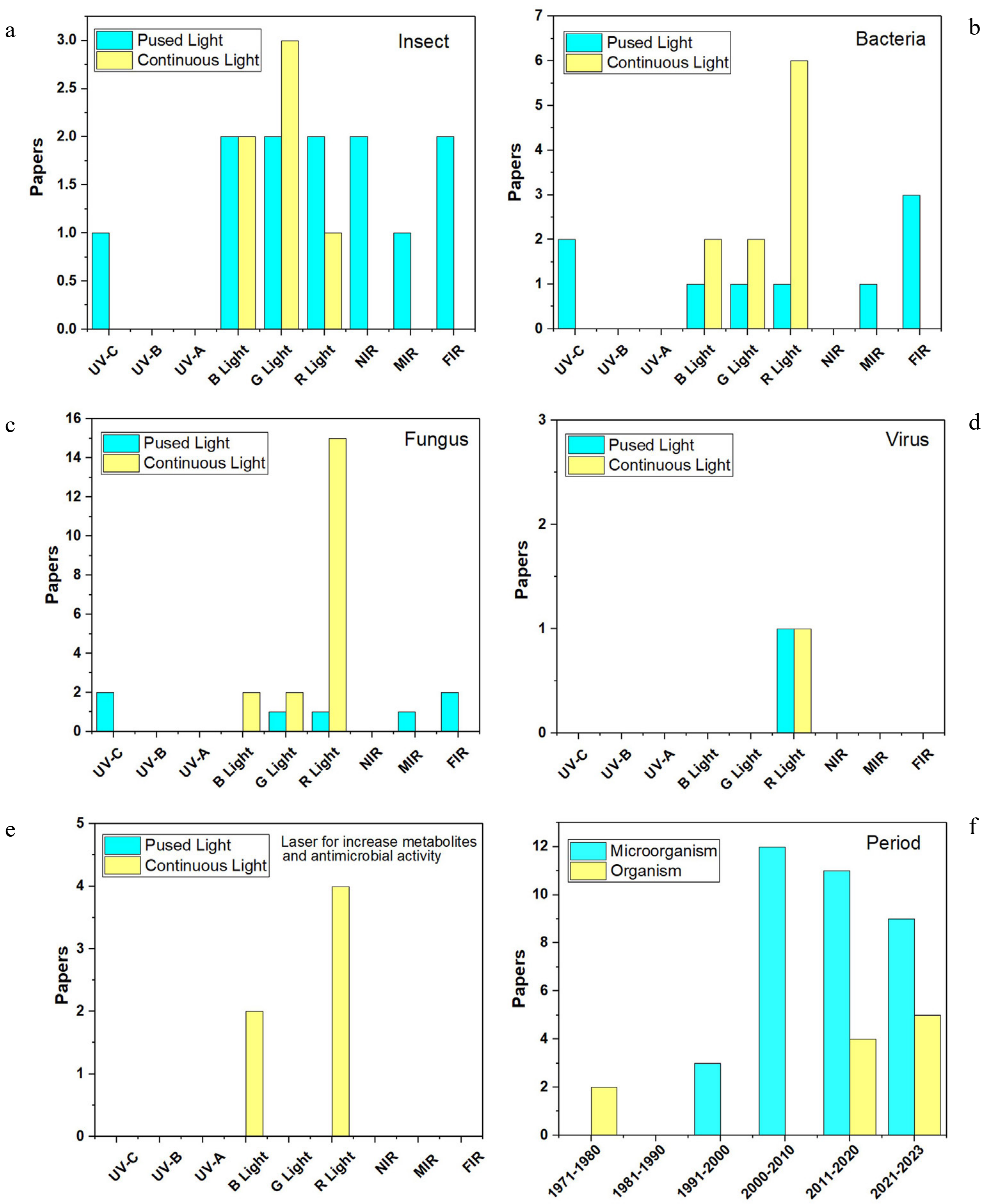

Fig. 3. Articles published related to the applications of laser light to control microorganisms and organisms of agri-food systems, more specifically: a) insects, b) bacteria, c) fungus, d) viruses, e) lasers for increasing metabolite and antimicrobial activity, f) Articles published by decade.

Rhopalosiphum padi). The main insects that have been annihilated and controlled according to the literature are illustrated in Fig. 4.

In relation to bacteria, it has been reported that the laser light wavelengths most commonly used are those in the visible light region (blue, green and red light) followed by those in the MIR and NIR and finally those in the UV-C region. The type of wave most frequently applied is the continuous wave in the visible region, while in the UV-C, MIR and FIR regions, they have only been applied in the form of a pulsed wave. It should be noted that in the FIR light range, CO_2 lasers have been used, which according to some authors have a pulse time in the order of microseconds. The bacteria controlled by means of these light regions include a) UV-C light emitting excimer laser (248 nm) used to control *Escherichia coli, Listeria monocytogenes, Salmonella choleraesuis, Yersinia enterocolitica, Staphylococcus aureus, Aeromonas hydrophilia, and Serratia marcescens*; b) by blue light: *Escherichia coli, Staphylococcus aureus* and *Candida albicans*; c) Green light: the controlled bacteria are *Escherichia coli, Staphylococcus aureus and Candida albicans*, d) Red light: *Escherichia coli, Staphylococcus aureus, Candida albicans, Arthrobacter globiformis, Bacillus macerans, Sarcina lutea*, *Chromobacterium violaceum, Arthrobacter atrocyanus, Pseudomonas aeruginosa, Saccharomyces cerevisiae, Staphylococcus aureus, Listeria monocytogenes, Salmonella* spp., *Bacillus subtilis* and *Pseudomonas aeruginosa*); e) MIR light has been used

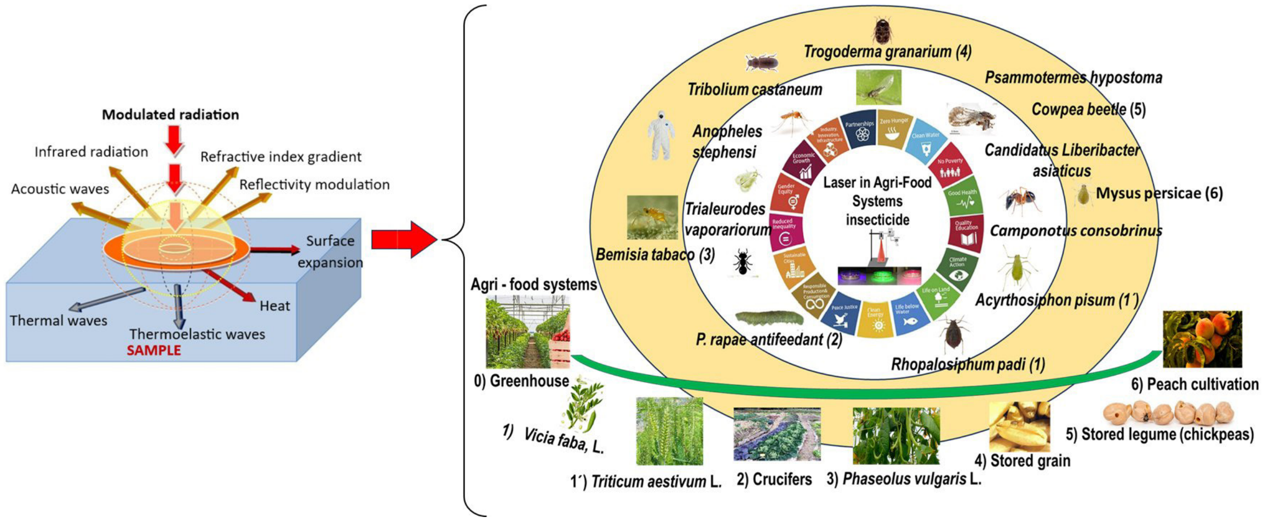

Fig. 4. Lasers in agri-food (insecticide).

on *Pseudomonas aeruginosa* and finally; f) FIR light produced by a CO_2 laser has been applied to control *E. coli* and *Salmonella* spp.

The most reported on microorganisms in the literature with regard to the application of laser technology for their control are fungi. According to these publications, laser light is regarded as a sustainable strategy in agri-food systems to achieve fungal control. The lasers that have been discussed in the literature and used for the above-mentioned purpose emit light in the UV-C, in the visible region (blue, green, and red) and also in the MIR and FIR regions. The use of lasers emitting light in the red region are most frequently reported, and among them the He-Ne laser followed by diode lasers. The light wave type used is predominantly continuous but in the case of the UV-C, MIR and FIR region it is pulsed. The types of fungi controlled by the light regions used are as follows: a) UV-C: *Alternaria alternata, Botrytis cinerea, Fusarium oxysporum, Fusarium roseum, Monilinia fructicola, Penicillium expansum, Penicillium digitatum*, and a pseudo fungus (*Phytophthora* spp.); b) blue light: *Botrytis cinerea, Rhizopus stolonifer*; c) Green light: *Aspergillus ochraceus, Chaetomium funicolum, Penicyllium lividum, Trichoderma polysporum, A. flavus, Rhizopus nigricans, G. roseum;* d) red light (*Aspergillus ochraceus, Chaetomium funicolum, Penicyllium lividum, Trichoderma polysporum, Rhizoctonia solani, Alternaria tenuissima, Cercospora kikuchii, Colletotrichurn truncatun, Pyricularia oryzae, Helminthosporium, Fusarium* sp., *Fusarium oxysporum, Aspergillus* spp., *Penicillium* spp. *Alternaria spp. Cladosporium* spp., *Rhizopus* spp., *Trichoderma* spp. *and Helminthosporium* spp., *Helminthosporium, A. flavus, E. nidulans, A. terreus, Penicillium, A. niger, Rhizopus*; e) MIR and FIR: *Penicillium digitatum.*

In the case of laser applications concerning viruses in agrifood systems, it was found in this literature review that, compared to the insects, bacteria, and fungi, these are the least studied. They have only been targeted using the red region and favourable results were achieved in the control of the tobacco mosaic virus and to increase resistance to viral disease using He-Ne lasers, in both cases low intensity lasers were used. In the visible range, the most frequently used type of light wave is the continuous wave, while in the red regions UV-C, MIR and FIR only pulsed wave is used. It is worth noting that its use could be considered a trend in the coming years in the face of emerging viruses. Svyatchenko *et al.* (2021) evaluated the antiviral activity of photodynamic therapy against SARS-CoV-2 *in vitro*. The authors used a laser (662 nm) emitting a continuous wave with doses of 16 and 40 J cm^{-2} thereby indicating the high level of effectiveness of photodynamic therapy to inactivate SARS-CoV-2 when accompanied by low concentrations of photosensitizers such as added dyes. Thus, further research will be required in the coming years with regard to applications in agri-food systems prone to attack by emerging viruses.

Finally, lasers have been applied as a strategy to increase the production of metabolites and to enhance the antimicrobial effect of plants. The lasers applied to date emit light in the red and blue regions, a continuous, low intensity beam of light was used.

Reducing the use of chemical pesticides in agri-food systems is essential. Therefore, it is a priority in the scientific community to continue to research and define the optimal irradiation parameters of laser light. But also, it is essential to establish the appropriate instrumentation and technologies for each application that is achievable in terms of cost, accessibility, ease of use, and portability for the real situations which arise in the agri-food system. The optimal laser technology can be used and acquired by an agricultural producer or consumer, it is available at affordable prices, it is easy to use and produces reliable results to ameliorate urgent problems. In this way, transdisciplinary research is necessary in order to consolidate the development and application of innovative and clean tools such as those that may be generated from the field of physics through the application of electromagnetic waves, in this case with laser light sources (Zaidem *et al.,* 2023). The transdisciplinary approach to research allows for the results of research to be brought closer to the realms of production. To the extent that more results and further adaptations to real-world agri-food systems are added over time, this could be a viable strategy for the sustainability of communities (Hernández-Aguilar *et al.*, 2020; 2022). In most developing countries, this type of technology is not yet being used, nor was the research focused perspective introduced in the 1970s. However, to the extent that it is adopted, the research will most probably be based on the context of its eventual application and then the technology will be adjusted to the exact needs of the user or group of potential users. In addition to this, the integration of disciplines and different specializations, from electronics experts to experts in optics, engineering, researchers, agronomists, phytopathologists, soil scientists, bacteriologists, *etc.* is required in order to jointly advance in terms of proposals and demonstrations adjusted to the needs of the real world.

In general terms, certain technologies from the field of physics show promise and will continue to be investigated, but increasingly this will occur in combination with other disciplines and eventually replace or at least reduce the use of chemical methods. As has already been addressed in this literature review, with a wide range of applications such as those that have been covered being used to control organisms (insects) and microorganisms (bacteria, fungi, viruses, *etc*.). One current trend is the use of hybrid methods in order to take advantage of the benefits and applications of laser technology, this has been addressed in the relevant research with respect to different laser wavelengths and physical methods (fixed and variable electromagnetic fields, electric currents, ozone, plasma, vacuum application, UV radiation, gamma radiation, sound, ultrasound, *etc.*) but without disregarding biological proposals (with bioactive elements being potentiated by laser radiation), osmoconditioning (the seeds are moistened in an osmotic solution (sugar, polyethylene glycol, glycerol, mannitol) over different periods of time, the seeds are left to dry in the air before sowing), hydro-conditioning (moistening of the seeds for different periods of time, the seeds are left to dry and then planted) and nano-conditioning (nanoparticles (zinc oxide, iron oxide, titanium dioxide, silver, gold) are used to provide the adequate use of water and nutrients) as all of these methods do not harm the environment or endanger the health of producers, their families, or consumers.

Taking all of the above into consideration, it is necessary to continue to research the synergistic effects of hybrid physical methods and also biological methods in order to develop applications in agri-food systems and to take action towards the achievement of sustainable development, but above all, the paramount aim should be the improvement of the quality of life of the population that is being adversely affected by the environmental and health consequences of the use of agrochemicals. International committees, such as those formed by the WHO, have noted that food irradiation can reduce the risk of microbiological foodborne diseases. In this research, pulsed or continuous wave laser irradiation has been presented for the treatment of not only finished food products, but of different stages in its production within agri-food systems, because in this way, in addition to the reduction of foodborne diseases, successful treatments would also reduce the incidence of disease due to the use and consumption of agrochemical residues, improve the quality of the air we breathe and also reduce losses in the development of crops and in the post-harvest stage.

The research reported shows that laser technology has the potential to strengthen the path towards sustainability with abundant evidence providing proof that such technology could provide a viable alternative to the chemical pesticides that are currently being used in agri-food systems. Consolidated research has already been conducted, however, research in this field is continuing so that it can be applied in the real world, in different countries. In addition to continuing to evaluate the effects of laser irradiation on different organisms and microorganisms, its mechanisms of action are being studied on an ongoing basis.

In the possible hypotheses concerning the mechanisms of interaction of laser light and biological objects, it is viable to argue that it is due to the substances contained in biological objects with light absorption bands, namely the chromophores (Hernández-Aguilar *et al.*, 2016). Photoreceptors are found in several biological objects, including seeds, insects, bacteria, fungi, *etc.* (Smith, 2000; Hernández-Aguilar *et al.*, 2010). These are modified through photosensitization using the molecules of some types of dye, which not only modifies the optical component of the seed, but also the thermal component. In this way, both of the proposed mechanisms of action have a simultaneous presence in the phenomena of laser radiation.

In this way, the various specimens with their respective characteristics (physical, chemical, optical, thermal, photothermal, *etc*.) first absorb light energy and then transform it into chemical energy and heat (Rassam, 2010; Hernández-Aguilar *et al.*, 2015a; 2015b; Jamil *et al.,* 2013). In the case where the production of heat plays an important role, the radiation dose of the laser light on the object of study is important, this involves the radiation intensity and the duration of exposure to continuous light or a pulsed light wave, in addition to the pulse size.

In the proposed mechanisms of action involving the interaction of laser light with the objects of study, both optical and thermics components have been reported. For example, when a laser light is applied for seed treatment, the absorption capacity of the light is modified depending on the characteristics of the light, but it also depends on the optical component associated with this object of study. Thus, if a 650 nm laser is applied, it will have a different optical penetration length as compared to the application of a 450 nm laser (Hernández-Aguilar *et al.*, 2011b; Pérez-Reyes *et al.*, 2015). According to the literature, depending on the colour or the dyeing status of the seed, the penetration length increases as a function of increasing wavelength. Thus, this parameter is modifiable by dyeing the seed or the biological object. The laser then causes changes in the tissue, in the case of insect changes may even occur in terms of colour, shape and in the appearance of some deformities, especially when high power pulsed lasers are used (Rashid *et al.*, 2018). On the other hand, there are also roles for low intensity lasers which could be used in the agricultural sector and in cereals or legumes being stored for later use in the food industry, lasers could be used to reduce their moisture content and thus improve their state of preservation.

One model which relates the optical and the thermal component is the model proposed by Rosencwaig and Gersho (1976) which operates through the optical absorption coefficient β and the thermal component through the thermal diffusivity value α and the thermal diffusion coefficient a_s, in the case of thermally thickened samples ($a_s l_s >> 1$) obtained by Poulet *et al.* (1980). In some studies that have been used to determine the absorption coefficients of biological samples, the best effects have been related to samples with higher absorption values, such as those reported in insects and seeds. In the studies of Gaetani *et al.* (2021) it was reported that when three lasers were employed to emit wavelengths of 532, 1 070 and 10 600 nm different coefficients of absorption were achieved (10^{-3} cm^{-1} at 532 nm, 10 cm^{-1} at 1 070 nm and 1000 cm^{-1} at 10.6 µm), where the best results were attained using the laser with the highest optical absorption coefficient.

According to the literature, it is important to note that some authors have reported the use of dyes to obtain better results. In bacteria, toluidine blue dye has been used mainly in conjunction with a He-Ne laser (Macmillan *et al.* (1966); for the green laser the dyes used in the biological objects have been safranin dye, rhodamine (Kohmura *et al.*, 2020), and when YAG lasers were employed, crystal violet dye was used (Tatsuno *et al.*, 2021), to target viruses (ruby laser and blue dye) (Hamrick and Cleary *et al.*, 1968) and for fungal control, in the case of a He-Ne laser and laser diode, the following dyes (methylene blue, methyl red and carmine, and methylene blue and methyl red) have been used (Ouf and Abdel-Hady, 1999; Hernández-Aguilar *et al.*, 2011a; Pérez-Reyes *et al.*, 2015; Sánchez Hernández *et al.*, 2015). The use of dyes allow for the modification of the radiation intensity and effect for the control of microorganisms and organisms. The photosensitization of biological objects has been reported to be one of the key factors for the control of organisms (insects) and microorganisms (bacteria, viruses, fungi). In this sense, the laser is emerging as both an environmentally and human friendly technology that could be used in the coming years in the agri-food systems of developing and developed countries as a strategy for sustainable development.

It is possible to envisage the potential use of lasers for fungal control in the near future. As can be seen in Fig. 5, fungus control could be achieved with inexpensive lasers as their light is mainly located in the red region of the visible spectrum, followed by the green and blue region. The result of the application of this technology is the potential control of fungi that afflict agri-food systems. One of the main causes of losses in such systems is fungal contamination. Also, people's health is affected by food contaminated with fungi, and it has even been found that fungi in food is linked to some types of cancer.

In this way, laser technology could be widely used in agri-food systems, it is potentially useful for both farmers and/or producers and also for the consumer, as well as benefiting sustainability.

8. CONCLUSIONS

The application of laser beams is a sustainable technology that is increasingly being applied as a pesticide in agri-food systems. The evidence shows the following possibilities of laser technology in agri-food systems: 1) To annihilate and combat insects, 2) to eliminate bacteria, 3) for the control or elimination of fungi, 4) to potentiate the active elements used in fungal control.

Concerning the annihilation and control of insect populations; laser light has the potential to degrading their tissue structure and has better effects when there are more pigments. Laser beams are applied in a pulsed form (from ms to s) or with a continuous intensity (from 1s to 40 min), both have been used to annihilate certain insects (whiteflies, ants, aphids, beetles, mites and pear psyllas), during the storage of cereals or leguminous plants.

In relation to their applications involving bacteria, laser beams have been used to eliminate bacteria which have a great impact on human health (*Escherichia coli*, *Salmonella*). The most frequently used lasers beams are those with (blue, green and red light) followed by those which employ the MIR, NIR and UV-C regions. Visible light beams have been applied as continuous waves, while those in the UV-C, MIR, and FIR regions have mainly been applied in the form of a pulsed wave.

In relation to fungal applications, the laser beams which are more frequently used are those in the UV-C, visible region (blue, green, and red lights), and also those in the MIR and FIR regions. Laser beams in the red-light region are most frequently reported, and among them the He-Ne laser followed by diode laser is most often used. The continuous light mode is predominantly used in the visible region and in the case of the UV-C, MIR, and FIR region the pulsed mode is used. Fungi which affect crops and human health, such as *Fusarium* and *Aspergillus*, have been controlled by using laser irradiation. Fungi are the most frequently studied microorganisms and, the least studied ones are viruses with regard to their laser resistance.

The effects of light on the control of organisms and microorganisms are enhanced using pigments and/or dyes. When using He-Ne, green and YAG lasers the dyes used for the control of bacteria include toluidine blue, safranin dye, rhodamine, and crystal violet dye. In the case of viruses, a blue dye has been used, in conjunction with ruby laser beams. In applications where lasers were used as fungicides, He-Ne lasers and laser diodes were applied together with dyes such as methylene blue, methyl red, and carmine. Finally, laser light was found to enhance the metabolites and antimicrobial activities of some plants.

Conflicts of Interest: The authors declare no conflict of interest.

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