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Harnessing ozone for agriculture and the agrifood sector: an interdisciplinary roadmap from current applications to future perspectives

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A B S T R A C T

Ozone is increasingly recognized as a versatile and sustainable tool in agriculture and the agrifood sector due to its high oxidative potential and rapid decomposition into oxygen, leaving no persistent residues. Despite these potentials, critical knowledge gaps remain regarding optimal dosage protocols, crop physiological responses, species-specific ozone effects, the overall impact on soil fertility and a lack of harmonized regulatory framework hampering a wide adoption of this strategy. This article provides a critical roadmap, transitioning from consolidated applications to innovative research frontiers. Established uses include equipment sanitation, post-harvest shelf-life extension, and the effective removal of pesticide residues. Beyond these, the manuscript explores emerging applications such as field pest and weed control, where ozone exhibits mechanistic overlaps with conventional herbicides, and its role as a biostimulant capable of triggering beneficial responses in crops. Collectively, these applications offer significant potential for advancing agricultural sustainability. The integration of ozone with digital innovations, including precision agriculture and digital twins, is highlighted as a key driver to optimize delivery and minimize variability. Gaining further insight into these aspects is essential for to optimize application strategies. By adopting a One Health perspective and utilizing Life Cycle Assessment data, the study underscores ozone's potential to significantly improve the sustainability of agricultural systems. Ultimately, this work emphasizes the need for multidisciplinary research and policy tools to bridge existing knowledge gaps and ensure the safe, efficient, and sustainable adoption of ozone-based technologies in modern farming.

1. General introduction

Ozone (O_3) is a gaseous form of oxygen (O_2) composed of three atoms, known for its pungent smell, colourless, and its relative instability when compared to O_2 found in the atmosphere. This makes ozone a highly oxidative compound [1]. Ozone forms differently depending on the environmental context. In the stratosphere, ozone is produced through the reaction of O_2 with nascent oxygen (O), which results from the photodissociation of molecular O_2 by ultraviolet radiation. In the troposphere, ozone is generated from the reaction between volatile organic compounds (VOCs) and nitrogen oxides (NOx) under high solar radiation. Its strong oxidative capacity and rapid decomposition allow it to inactivate a broad range of microorganisms much faster than other oxidants such as oxygen, chlorine gas, permanganate, or hydrogen peroxide [2]. Thanks to these properties, ozone is not only a powerful disinfectant, but also a bleaching and deodorizing agent, which expands its possible fields of application. However, the high reactivity of ozone with organic matter, present in both air and water, leads to its rapid consumption, thereby reducing its concentration and, consequently, its effectiveness [3].

Ozone is widely used in various industrial applications. The treatment of drinking water with ozone is adopted by many municipalities due to its effectiveness in removing heavy metals (mostly iron and manganese), taste, and odour, as well as for disinfection. Ozone has also

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been proposed as an alternative to chlorine in the food industry for contamination control and, more recently, to extend the shelf life, following increasing restrictions on chlorine use. Despite only a few issues related to alterations in sensory attributes having been reported, this solution is still rarely adopted [3–5]. Additionally, ozone is being considered for cleaning applications in the food processing industry, although mainly at the research level. Surprisingly, despite its effective sterilization potential, its application for sterilizing medical devices has been scarcely investigated [2]. Moreover, the textile processing sector has already adopted ozone for both material processing and wastewater sanitation.

In agriculture, ozone has emerged as a promising technology due to its dual role: as a sanitizer, capable of controlling pathogens, pests, and post-harvest spoilage, and as a potential tool to improve crop management [6–11]. Its applications include *i*) seed and facility disinfection, *ii*) removal of pesticide residues, *iii*) shelf-life extension, and *iv*) stimulation of germination and growth. Despite encouraging results, numerous fields remain unexplored or only marginally investigated. These include, for example, a broader range of food industry applications, the definition of specific protocols for different purposes (disinfection, shelf-life extension, equipment cleaning), and the direct use of ozone in the field for pathogen and weed control. A critical challenge is represented by the correct determination of dosages: overdosing can cause detrimental effects on crops or soil organisms, while underdosing reduces effectiveness. Moreover, ozone exhibits significantly lower stability compared to atmospheric O_2, limiting its accumulation and thereby reducing long-term environmental risks. However, this same instability requires on-demand ozone generation through dedicated systems, which entails additional operational and economic costs [3]. These technical and economic constraints are part of broader open questions that will be further addressed in this manuscript. Beyond these aspects, the increasing attention to sustainable agriculture highlights the potential of integrating ozone-based technologies with digital innovations, life cycle assessment, and the *One Health* perspective, linking agricultural productivity to environmental and societal impacts.

Therefore, this manuscript aims to provide a critical overview of consolidated and emerging applications of ozone in agriculture, highlighting both benefits and limitations. The structure follows a progression from established uses to novel research frontiers, unresolved scientific questions, and interdisciplinary perspectives, with the ultimate goal of outlining future directions for safe and sustainable adoption of this tool. This review does not aim to propose ozone as a replacement for agrochemicals, but rather to highlight mechanistic overlaps and potential complementary applications.

2. Consolidated agricultural applications

This section examines the consolidated applications of ozone in agriculture, focusing on its roles in sanitation, pesticide residue removal, postharvest preservation, and crop development. Each subsection addresses the specific mechanisms, benefits, and limitations reported in the literature, providing a critical overview of current knowledge and practical outcomes.

2.1. Cleaning and disinfection

The use of ozone and its effectiveness in controlling soil-borne pathogens, nematodes, and insect pests, as well as in reducing post-harvest diseases and preserving food quality, has been thoroughly investigated [11–14]. Unlike conventional pesticides, ozone does not contribute to the development of biological resistance or environmental pollution, making it a promising alternative for disinfection and pest control, particularly in greenhouse settings [15]. For example [16,17], found that ozonized water with an ozone concentration of 3 and 4 mg L^{-1} was able to exterminate 100% of nematodes after 8 min of submersion, while other promising experimentations were run to eliminate aphids by Ref. [18]. Other studies reported the possibility of disinfesting soil from microorganisms [19], especially fungi [20]. Because of this, it is expected that ozonated water can have negative effects on pedofauna, but studies are still scarce, and it is expected that ozonated water treatments be tailored to individual crop and pedo-climatic conditions to avoid adverse effects [19]. Ozone has also been considered for disinfection and odour removals from livestock husbandry due to its fast action and the efficient spatial dispersion [21]. [22] observed a reduction in atmospheric evolution of 80% for ammonia (NH_3) and 77% for hydrogen sulphide (H_2S), with considerable improvement of air quality within breeding environments. These results highlight the potential of ozone not only in crop protection but also in improving animal welfare and environmental conditions in intensive farming systems.

In the context of equipment sanitation in food processing, ozone has shown promising results, particularly in reducing *Brettanomyces* populations, thereby improving wine quality. Its application appears to be a valuable alternative for cleaning various types of processing equipment, contributing to reduced water usage and enhanced environmental performance [23].

Ozone is highly soluble in water at 20 °C and exhibits optimal sanitizing efficacy at concentrations between 0.5 and 2.0 mg L^{-1}, particularly within a pH range from 6.0 to 8.5. Despite its short half-life (∼20 min) and residue-free decomposition, ozone must be managed with caution due to its corrosive properties and the health risks associated with exposure to concentrations exceeding 4.0 mg L^{-1} [24,25]. Moreover, the mode of application (aqueous wash, fog, gas phase, soil drench) plays a decisive role in both efficacy and risk mitigation [12].

In agricultural and food applications, large-scale adoption is strongly linked to consumer acceptance, which requires maintaining sensory and organoleptic qualities within market standards. To advance the use of ozone in postharvest treatment, research should clarify the underlying metabolic pathways involved in shelf-life extension, ensuring that nutritional and quality attributes remain intact [11]. At the same time, unresolved issues remain, particularly regarding optimal dosage and potential non-target effects on soil biodiversity, which will be further discussed in Section 4.

While current findings are promising, further studies are needed to define protocols that optimize the balance between product longevity, safety, and quality assurance.

2.2. Removal of pesticide residues

Ozone has been observed to oxidize and reduce the presence of diverse types of pesticides on different fruits and vegetables surfaces, including oranges, grapes, apples, strawberries, raspberries, and blackcurrants [26–30]. It also appears that ozone can penetrate plant tissues, potentially enhancing its activity. However, there are no in-depth studies on the actual efficiency or the underlying mechanisms of this penetration process. In citrus matrices [31], reported that chlorothalonil residues adsorbed on oranges were completely removed after 5 min of ozonation using an ozone concentration of 4 mg L^{-1} in a water solution. A recent study on cauliflower demonstrated the significant potential of ozone to effectively degrade various pesticide residues present on the crop surface by around 60% [8]. Although other agents, such as chlorine and hydrogen peroxide, have demonstrated higher efficacy, the authors underscore the significance of ozone owing to its comparatively lower environmental impact and safety profile. This is attributed to its rapid decomposition into O_2 molecules within 5 to 15 min post-application, minimizing residual chemical presence. In grapes [32], showed that washing table grapes with ozonated water (3 mg L^{-1} for 5 min) before cold storage reduced multiple pesticide residues up to 100%, compared to SO_2 control (20–60% reduction). Moreover [33], achieved 100% degradation of pyraclostrobin on table grapes with gaseous ozone treatment. In apples, repeated exposure to ∼1 mg L^{-1} gaseous ozone during cold storage has been shown to reduce fungicide residues significantly, though quantification varies depending on the compound and treatment duration [34]. In addition [35], showed that repeated exposure to 1 mg L^{-1} ozone during cold storage reduced fungicide residues by up to 79%, highlighting its potential for postharvest preservation in apples. Conversely, other investigations revealed pesticide-specific resistance. Boscalid residues in grapes are only partially degraded under comparable conditions, indicating that molecular structure plays a crucial role in ozone susceptibility [31,36].

The use of ozone as a tool to reduce pesticide residues has also been investigated within the food processing industry [9]. reported significant reductions of pesticide residues on grape juices, with residual contaminant loads decreasing by up to 50%. Comparable findings were reported in tomato juices, where carbosulfan, oxamyl, and methomyl residues were completely degraded within 15–30 min depending on the initial pesticide concentration [37]. However, in some fruit juices, ozone exposure altered phenolic profiles and induced colour loss [38,39], underscoring the need to balance safety gains with potential quality impacts. Nevertheless, better results have been observed using UV-C lights, highlighting the need for further investigations on the topic. Taken together, these case studies emphasize that the success of ozone treatment is context-dependent, governed by treatment parameters (concentration, exposure time, temperature), commodity characteristics (surface area, wax layers, tissue compactness), and the chemical structure of the pesticide. Future research should therefore prioritize the optimization of these parameters and explore hurdle approaches, such as combining ozone with UV-C irradiation, ultrasounds, or pulsed electric fields, to enhance degradation efficiency while preserving nutritional and sensory quality [40].

2.3. Postharvest shelf-life extension

The main proposed mechanisms by which the ozone use could extend the shelf life of products are primarily the reduction of microbial load on the product surface, by inactivating spores, insect control through the reduction of populations, and the decrease of the respiration rate of fruits and vegetables following the degradation of ethylene [41–43]. The main challenge is linked to the inactivation of spores and eggs, representing the most difficult phases to kill pathogens and insects [23]. Depending on the species, developmental stage, and type of product to be preserved, different exposure times, ozone concentrations, and delivery methods (such as water or air) may be required. However [44], report that ozone may have negative effects on quality, particularly when applied at high doses on fresh products. In this context [7], suggested a dosage ≤50 mg L^{-1} to maximize the control against insects and microorganisms while avoiding food quality decrease, preserving equipment and human health. Yet, these represent broad generalizations that necessitate targeted assessments according to the specific agricultural product, the nature of the harmful agent, and the characteristics of the operational environment. A significant scientific effort is currently underway to evaluate the potential of ozone in the meat industry. Recent studies have demonstrated promising effects of ozone in enhancing meat shelf life through the reduction of pathogens such as *Salmonella* spp. and *Escherichia coli* [45]. Future perspectives also have to point to integrated approaches, where ozone could be combined with other non-thermal technologies such as UV-C, high-pressure treatments, or pulsed light, thereby enhancing microbial inactivation while mitigating negative impacts on food quality.

2.4. Seed germination and crop growth

[15] reported a positive effect of seeds germination at low concentration of ozone exposure but only for a few species (wheat, maize, tomato, beans, and cucumber). Nevertheless, the concentration of ozone results to be an aspect to be investigated in depth to tailor dosages on crop-specific needs. Similarly [46], observed a slight improvement in photosynthetic activity in lettuce plants treated with ozone at the seed or early seedling stages. A study conducted in Spain evaluated the effect of ozone, delivered through the circulating solution, on chili pepper growth [47]. The results indicated a positive impact on growth, likely due to improved uptake of N and K. However, these observations were also associated with reduced root system development, highlighting the need for further research to determine the optimal ozone concentration that supports crop growth without adverse effects. The positive effect of ozone was also investigated on baby red chard in a floating hydroponic system in Chile [48]. Results suggested that ozone increased micronutrients uptake (iron and manganese) and yield production. A study conducted in Russia [49] reported the positive effect of ozone as a suitable strategy to improve wheat germination. The study reported a threshold of 6 g m^{-3} of ozone, beyond which no significant increase in the seeds germination rate was observed.

Despite the moderately widespread use of ozone in agriculture, the limited available literature consistently highlights the need for more in-depth research to accurately assess the actual potential of this tool. Several application areas remain unexplored or have only been marginally examined, representing significant opportunities for innovation in agricultural production systems.

3. New frontiers for agricultural applications and research perspectives

This section explores innovative and less conventional applications of ozone in agriculture, focusing on its potential beyond consolidated uses. The aim is to highlight emerging research directions, ranging from field pest and weed control to biostimulant effects and post-climatic event treatments, while also considering technological integration and sustainability. By outlining these new frontiers, the section seeks to identify both opportunities and knowledge gaps that must be addressed for ozone to become a reliable and scalable tool in future farming systems.

3.1. Field pest and disease control

The use of ozone may represent a promising strategy in agriculture for enhancing the effectiveness of treatments aimed at controlling pathogens. Due to its strong oxidizing properties, ozone could also contribute to managing infestations directly in the field by reducing or inactivating populations of harmful agents like yeasts, fungi, bacteria, viruses, and other parasites [50]. An example is represented by its application in grapevines at the cluster level. This approach is being pioneered by a European Union H2020 project (OXIR – GA n° 873055, https://www.oxir.eu/work-packages/) started in 2020. In this project, the potential of ozone for controlling pest and pathogen pressure was evaluated over two years on several crops like lettuce, basil, blackberry, strawberry, hemp, muskmelon, tomato, and grapevine. In all these crops, ozonated water significantly reduced fruit-level infestations when applied alone. However, the highest efficacy was achieved in combination with a fungicide (Serenade ASO®, *Bacillus subtilis* QST 713). An important observation was that the ozone treatment on grapes helped preserve the natural microbial biodiversity associated with leaf surface and grape clusters. Other studies have been run to extend the fruit life cycle using both ozonated atmosphere with ozone concentration of 100-300 μg L^{-1} (apple, peaches, and grape) or 5-10 mg L^{-1} (dried figs), or water with concentration of ozone of 1.5-10 mg L^{-1} (citrus fruits, peaches, nectarines, and table grape), 0.3-1 mg L^{-1} (oranges and lemons), or 350 μg L^{-1} (strawberries), with different times of exposure and temperature [41,51–58]. Ozone demonstrated to be effective also in controlling infestations of the primary fungal pathogens affecting chestnut in post harvesting [59,60]. Currently, stocks of chestnuts are commonly subjected to immersion in water at room or lukewarm temperature, a treatment originally developed in Italy in the 1930s [61]. This treatment can control insect infestation, killing especially the larvae of *Curculio* spp. and *Cydia* spp. and molds without influencing nut quality. A similar treatment, inappropriately called "sterilization", consists of submerging the nuts in water at 48–52 °C for 35-45 min followed by rapid cooling in tap water for about 15 min [62]. This method is also able to kill insect larvae but does not eliminate the mycoflora contaminating the fruits [61]. Other studies introduced the use of hot air-assisted radiofrequency treatments [63], or a combination of biological and physical methods [64], or the use of a range of chemical treatments [65] to eliminate contaminating microorganisms. Yet, chestnuts exposed to a continuous flow of air with 300 μg L^{-1} of gaseous ozone showed a marked reduction in microbial population [66–70] which allowed storage at 2 °C to be considerably extended without negative effects on fruit quality parameters [70].

3.2. Integration with agrochemicals

The adoption of ozone in agriculture can contribute to increased agrochemical efficiency, especially fungicides, while improving the overall sustainability of agricultural practices (OXIR Project, Work Package #2, https://www.oxir.eu/work-packages/). In this context, encouraging outcomes can be achieved through the integration of ozone with conventional agrochemical applications, indicating the potential for effective pest and disease control while concurrently reducing the required agrochemical dosages. This combined approach not only enhances treatment efficacy but also promotes more sustainable and environmentally responsible agricultural practices.

3.3. Weeds control with ozonated water

Ozone, once it enters plant tissues through stomata [71,72], acts as a strong oxidant, reacting with lipids, proteins, nucleic acids, and carbohydrates, thereby triggering lipid peroxidation, loss of membrane integrity, and damage to proteins and DNA [73]. It also stimulates the production of reactive oxygen species (ROS), activating plant defence responses such as phytoalexin synthesis, lignification, and the hypersensitive response [74]. Interestingly, several of these effects resemble the action of herbicides [75]. For instance, pro-oxidant herbicides such as paraquat and diquat exert their toxicity by diverting electrons from photosystem I and generating large amounts of ROS, causing membrane lipid peroxidation and necrosis, an outcome strongly reminiscent of ozone-induced oxidative stress [76]. Likewise, photosystem II (PSII) inhibitors such as atrazine or diuron impair electron transport, reduce CO_2 assimilation, and enhance ROS accumulation, paralleling the ozone-mediated inhibition, at the level of physiological outcomes rather than shared molecular target sites, of photosynthesis, stomatal dysfunction, and premature senescence [77]. Moreover, the activation of the phenylpropanoid pathway under ozone exposure, with increased synthesis of lignin, flavonoids, and stilbenes, indirectly recalls the involvement of aromatic amino acid–derived secondary metabolism, which is also affected by glyphosate through interference with the shikimate pathway, although through distinct biochemical targets and mechanisms [78]. In this sense, although ozone is not a herbicide *per se*, it induces a phytotoxic syndrome that mirrors the mechanisms of different herbicide classes, combining oxidative burst, photosynthetic impairment, and interference with secondary metabolism. This mechanistic overlap highlights how plants perceive and respond to ozone in ways that are functionally similar to their responses to xenobiotics, suggesting that ozone stress physiology could serve as a useful comparative model for understanding herbicide action and resistance.

Considering these similarities, ozone could also be employed as a means for targeted weed control. Specifically, the use of ozonated water at specific ozone concentrations (in the order of tens of mg L^{-1}) and applied under high-pressure spraying conditions (from 70 to 80 until 400-500 bar) have been proposed and explored in preliminary and pilot-scale experiences as a potential approach to damage weed vegetation across various contexts, including plant nurseries, horticulture, orchards, open-field cultivation, and even urban environments. Treatment aimed at weed removal from the sidewalks not only enhances urban decorum and aesthetics but may also potentially contribute to surface sanitization or reduction of organics (pollen, unburned hydrocarbons, etc.) and organisms (including *Escherichia coli*) originating from humans and animals that can be windblown to increase the particulate matter (PM) concentration and danger. In urban environments, ozone-based treatments could be further developed and explored for application not only to roads and sidewalks, but also along railway tracks and potentially as a mosquito control measure within drainage systems, although these applications would require specific experimental validation under real operating conditions. Moreover, the rapid degradation of O_3 to O_2, along with its reduced likelihood of inducing classical target-site resistance mechanisms, as observed for single-site herbicides, makes this strategy a promising and sustainable approach for managing weeds and reducing PM danger and annoying insects. However, it should be noted that all these mechanisms of action are clearly non-selective, meaning that ozone can damage both target and non-target plant and/or insect species. This intrinsic lack of selectivity highlights the importance of focusing on the application practice as the key driver of selectivity, rather than on the compound itself. For this reason, weed control strategies based on ozone should adopt delivery methods that restrict exposure, such as localized spraying on weed foliage, soil injection under plastic mulch, or confined spot treatments. As an example of spatial confinement, research on drip-applied fumigants has shown that when the chemical is delivered beneath plastic mulch, its distribution is restricted to the treated bed, which improves weed control efficacy while simultaneously reducing off-target risks [79]. More recently [80], demonstrated that ozone-induced free radical solutions, when applied through precision spraying, significantly reduced pest and weed pressure on Chinese cabbage while minimizing non-target effects.

3.4. Use as a biostimulant

Ozone penetrates plant tissues through the stomata and migrates by molecular diffusion until it reaches the cells, where the damage occurs [81,82]. However, the occurrence of damage mainly depends on the amount of ozone penetrating the tissues. In fact, while ozonated water with a concentration of 10-20 mg L^{-1} of ozone can dry out most of the herbaceous plants, at lower concentration, ozone may function as a biostimulant. In grapevines, for instance, ozone has been shown to act as a signalling molecule capable of inducing pigment biosynthesis. Empirical observations conducted in a commercial winery in the Emilia Romagna region, Italy (Bucchi R., 2025, personal communication) have shown that ozone-treated grapevines exhibit increased pigmentation intensity. A study of [83] reinforces these observations: low-dose gaseous ozone treatments (10 mg L^{-1} for 1 min) increased total polyphenols by 79% and antioxidant potential (ABTS + 55%, DPPH + 66%) in the leaves of the ornamental plant *Kalanchoe daigremontiana*, accompanied by higher superoxide dismutase and catalase activities, thus stimulating defence metabolism without phytotoxicity. Similarly [84], demonstrated that ozonated water soaking enhanced flower growth and markedly boosted flavonoid and rosmarinic acid contents in the aromatic plant *Agastache rugosa*, while increasing antioxidant enzyme activities. These results suggest that ozone is a molecule with hormetic properties, that is, capable of producing a biphasic response in which at low doses it stimulates beneficial effects while at high doses it is harmful. Because of this, it would seem that exposures to sub-lethal concentrations stimulate secondary metabolism and antioxidant pathways. [85], in horticultural crops, further highlighted that controlled ozone application can improve phytochemical profiles, mechanical properties, and storage resilience. Furthermore, treated grapevines tend to retain green coloration for a longer period after harvesting (Bucchi R., 2025, personal communication), suggesting a delayed onset of senescence. In addition, the canes of ozone-treated vines exhibit greater structural firmness, which may be indicative of prolonged nutrient mobilization toward lignified tissues. This, in turn, could enhance the accumulation of carbohydrate reserves, thereby potentially improving vine vigour and productivity in the subsequent growing season.

Collectively, these pieces of evidence align with the concept of ozone as a sustainable biostimulant rather than a purely phytotoxic agent. Nevertheless, the precise mechanisms underpinning these responses remain only partially understood. While enhanced antioxidant activity, activation of defence enzymes, and stimulation of secondary metabolic pathways such as phenylpropanoid biosynthesis are consistently reported [86–88], the molecular signalling cascades linking ozone perception to these downstream effects are still unclear. This lack of mechanistic resolution underscores the need for more integrative studies combining physiological, biochemical, and omics approaches to fully disentangle how ozone shifts from being a stressor to acting as a biostimulant.

3.5. Treatments after climatic events

During the already mentioned OXIR Project, ozone applications have been explored as a post-injury treatment for hail damage in several crops (https://www.oxir.eu/work-packages/), but it is important to note that there is limited research on its effectiveness and potential benefits. In fact, ozone can both benefit and harm plants depending on the concentration and application method. Further research is needed to determine if ozone can be a practical and beneficial tool for mitigating the effects of hail damage on crops. Here, we speculate on both beneficial and detrimental effects:

Beneficial – 1) disinfection: ozone can function as a sanitizer by reducing microbial growth and secondary infections [11], being potentially used to reduce the risk of secondary infections occurring when hail damage exposes plant tissues to the environment; 2) stress tolerance: studies suggest that ozone, at specific concentrations and application methods, can enhance plant natural defence mechanisms against oxidative stress [75]. *Detrimental* – 1) increased oxidative stress: high concentrations of ozone can exacerbate oxidative damage in plants, leading to further injury and potentially hindering recovery [74]; 2) photosynthesis inhibition: exposure to relatively high concentration of ozone can reduce the rate of photosynthesis, impacting the plant ability to produce energy and recover [89]; 3) premature senescence: ozone can accelerate the plant aging process, leading to premature leaf drop and reduced lifespan [90].

Therefore, for post-hail ozone applications, a series of factors should be taken into account:

- *Concentration and application method.* The effectiveness and safety (acceptable risk when properly managed) of ozone applications are highly dependent on the concentration used and how it is applied. Relatively high ozone concentrations can be harmful.
- *Crop type and growth stage*. Different crops and their growth stages respond differently to ozone exposure. Careful consideration is therefore required when determining the appropriate application strategy.
- *Timing of application.* The timing of ozone application after a hail event is crucial. Waiting a few days to assess the extent of damage and allowing for some natural recovery may be beneficial.
- *Insurance adjusters.* Since many farmers have hail damage insurance, we recommend contacting the insurance adjuster before making any decisions about post-hail treatments.

More research is therefore still needed to determine economic viability and optimal application methods for using ozone as a post-hail treatment for crops.

3.6. On-farm technological development and integration with digitalization

The evolution of on-farm ozone applicative solutions both as a sanitizing agent and for the control of phytopathogenic agents must be framed within the strategies to identify alternatives that aim to replace or reduce the significant impact of agrochemicals currently in use. From this point of view, many similarities can be identified with what has been proposed so far in the field of application regarding the use of devices emitting UV-C radiation. Here, the technological replacement is clear and well-defined: a physical process as an alternative to chemical treatment. In the case of ozone treatments, we can speak of an *intermediate solution*: physical means are used to produce, when necessary and in *real time*, a control factor (ozone) that then acts chemically.

The main similarities between ozone-based approaches and UV-C treatment solutions concern:

1. No persistent residues in the environment, eliminating the well-known problems of contamination from drift phenomena.
2. Use aimed more at reducing the number rather than at replacing agrochemical treatments, thus recognizing these processes as having a more prevalent role in supporting sanitation.
3. More flexible supply chain of the production factors to be used, with consequent simplifications in storage management; in the case of ozone, procurement involves the purchase of oxygen cylinders of generally limited volume (15-25 L), similar to those used in the medical-health sector and in any case generally sufficient to meet, based on the ozone concentrations currently considered most appropriate (60-80 g m^{-3} of water), the requirements of an entire production season.
4. The need to operate with dedicated equipment, which is normally separate from that used for chemical treatments.
5. Providing application procedures that, although not entirely free from risks to the health of operators, do not require special protective solutions, generally resolvable with appropriate shielding at the level of the machines used or by wearing normal personal protective equipment.
6. Application cases are still rare, often limited to experimental activities with prototypes, with commercial solutions mainly focused on greenhouse cultivation or post-harvest agri-food activities, with applications on stationary equipment dedicated to sanitization or sterilization activities.

The main interest in ozone obviously concerns the possibility of using it in mobile field processes to achieve the advantages mentioned at the abovementioned points 1. and 2. In this regard, this solution has a considerable advantage over UV-C treatments. The latter, in fact, offers the best results with treatments carried out at night (sunlight inhibits the effects of UV-C radiation) and operating with a *stop&go* mode according to a prescriptive map, to better dose the amount of energy to be delivered according to the level of risk evaluated through previous crop monitoring activities. By contrast, there are no such limitations in this regard for ozone treatments, which can be applied at any time of the day, even in a preventive manner (provided that site-specific application strategies are adopted, for example using volumetric sensors to detect canopy volume in *real time* and adjust water doses accordingly). A further advantage of ozone over UV-C (which requires specific machines) is that it can be adapted to sprayers already in use for conventional treatments, simply by integrating an ozone generator *(ozonator)* into them (Fig. 1).

This is a device that converts oxygen into ozone through electrical discharges (*via* a dielectric medium), as schematically represented in Fig. 2. During operation, therefore, the generator must be connected to an oxygen cylinder. The gas produced is then mixed with the water to be distributed by: *i)* bubbling inside the sprayer tank, or *ii)* near the nozzles, exploiting the aerosol formation processes at their outlet (Fig. 2). However, the latter solution still presents many uncertainties, due to both the difficulties of achieving a uniform mixture and the findings on crops, which in various situations have suffered damage to plant tissues.

Fig. 1. Trailed-coupled sprayer of the firm Caffini (Patú, Verona – Italy) equipped with an ozonator of the firm Ecofarm Storti (Verona – Italy).

4. Open questions

Despite increasing interest in ozone applications in agriculture, several unresolved questions remain that limit its large-scale adoption. These concern not only the technical definition of optimal dosages and application methods, but also fundamental issues related to soil health, atmospheric persistence, environmental trade-offs, operational constraints, and the absence of tailored regulatory frameworks. This section addresses these open challenges, highlighting the critical knowledge gaps that must be filled to ensure safe, efficient, and sustainable use of ozone in agriculture.

4.1. Definition of the optimal concentration for weed and pests control

The potential of ozone use for the control of weed has already been outlined in previous sections, but the definition of its optimal concentration remains uncertain. While some studies suggest that concentrations between 10 and 20 mg L^{-1} can inhibit weed growth effectively, it is unclear how these thresholds vary across distinct species and environmental conditions. Further research is needed to establish standardized protocols that maximize efficacy without compromising crop health or non-target organisms. The use of water as a medium for delivering ozone could potentially be enhanced by substituting it with specific oils, which may improve ozone efficacy, particularly against harmful insect pests. However, the non-specific mode of action of ozone raises concerns and necessitates targeted studies to evaluate these effects comprehensively. The lack of literature regarding this aspect underscores the need for further research to elucidate the actual contribution of ozone to weed and pest management. Defining specific dosages and application methodologies could be crucial in mitigating the environmental impacts of the agricultural sector. Furthermore, the absence of synthetic residues aligns ozone with the principles of organic farming.

4.2. Effect on overall soil fertility

The effects of ozone on soil biodiversity also warrant deeper investigation. Although moderate ozone exposure appears to exert limited immediate harm to beneficial soil microorganisms, the long-term impacts on microbial community structure and function remain poorly understood. Since ozone penetration with a common trailed sprayer is generally limited to the top 5 cm of soil [91], risks are concentrated in the surface layer, where microbial activity and organic matter are highest [92,93]. In this regard, inappropriate ozone dosages or excessive application methods risk triggering a trade-off that adversely affects soil biodiversity. This underscores the imperative for further studies to identify optimal application protocols that preserve, rather than compromise, soil microbial health. Concerns persist about potential disruptions to nutrient cycling and overall soil health, especially under repeated or high-dose ozone treatments. Similarly, the influence of ozone on soil organic matter (SOM) dynamics is not fully elucidated. While the high oxidation potential of ozone indicates that it may accelerate organic matter mineralization and nutrient release, the extent to which this affects soil C stocks and fertility over time is unknown. In this respect, the strong non-selective oxidative capacity of ozone poses a serious concern when applied directly to soil. In fact, beyond its impact on living microbial communities, it may also alter the pool of non-living SOM, a cornerstone of soil fertility. Excessive oxidation could accelerate SOM mineralization, leading to C losses and reducing long-term soil health. While ozone represents an efficient tool for supporting agricultural systems, it is essential to characterize potential risks and establish specific protocols that ensure the preservation of soil fertility. This issue is particularly relevant within the framework of the European Union “Soil Mission”, which explicitly promotes the protection and regeneration of soil biodiversity and organic matter level as fundamental assets for sustainable agriculture and climate resilience [94]. Recent updates (2023–2024) of the *Mission Soil agenda* further stress the need to monitor biodiversity responses to innovative technologies, and the use of unconventional inputs such as ozone is certainly included among these.

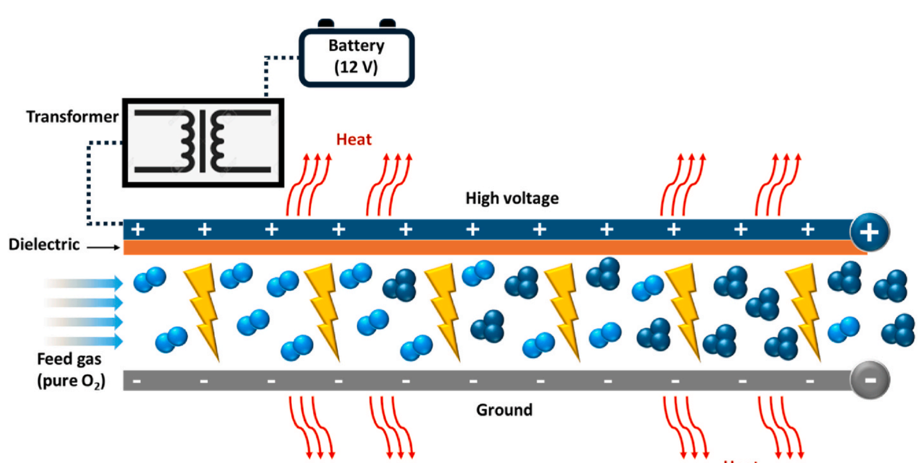

Fig. 2. Schematic of the ozonator module operating principle.

4.3. Role as a biostimulant

As mentioned, beyond its biocidal role, ozone appears to have biostimulant effects like stimulating greater photosynthetic activity, with potential increases in nutrient uptake and improvements in stress responses [83–85]. Yet, the underlying physiological mechanisms and the consistency of these effects across crop species remain largely speculative and require further empirical validation. As a matter of fact, only a few studies report positive effects, particularly on seed germination and secondary metabolite accumulation, highlighting the need for systematic investigation. Given the widespread use and growing interest in biostimulants, it is crucial to determine whether ozone fits this category and to characterize its effects on crops. While ozone may enhance yield performance and resilience to biotic and abiotic stresses, its actual potential must be rigorously established. Evidence-based clarity is essential to avoid speculation and define how this tool can effectively contribute to future residue-free agriculture [95].

4.4. Regulatory aspects for human, crop, and soil risks

The atmospheric residence time of ozone prior to its decomposition into O_2 is short, typically ranging from minutes to hours. However, variability in environmental factors such as temperature, humidity, and reactive compounds can complicate predictions of ozone persistence and its ecological impact. Clarifying these dynamics is essential for evaluating the environmental risks associated with ozone application. In this sense, legal limits for human exposure to gaseous ozone vary by countries and contexts (workplaces or outdoor environment). Several regulatory bodies have established exposure thresholds for occupational activities, which differ from ambient air-quality standards and depend on the exposure duration and workload. In the United Kingdom, a time-weighted average of 200 μg per cubic meter of air (μg m^{-3}) has been adopted for an 8-h working-time, with short-term exposure limits of 600 μg m^{-3} for 15 min [96]. In the USA, the limit is from 100 to 200 μg m^{-3} (depending on work intensity) as an 8-h time-weighted average [97,98]. EU has not adopted a Union-wide occupational exposure limit, but has left the possibility of legislating on this matter to each member state; therefore, for example, an 8-h time-weighted average limit of 200 μg m^{-3} is valid in Austria, Denmark, France, Hungary, and Sweden; the limit is 150 μg m^{-3} in Poland, 120 μg m^{-3} in The Netherlands, and 100 μg m^{-3} in Finland, Latvia, and Romania [99]. In Italy, the Decree 81/2008 indicates that workers cannot be exposed to more than 200 μg m^{-3} for 8-h time-weighted average and not more than 600 μg m^{-3} twice a day over 15 min [100]. Instead, in Europe, the outdoor air quality limits range from 120 μg m^{-3} as a maximum daily 8-h mean, not to be exceeded for more than 25 days per year [101], while in USA the limit is 140 μg m^{-3} as the fourth-highest daily maximum 8-h concentration, averaged over three years [102]. The lack of harmonized global legislation poses a significant risk of creating heterogeneous outcomes and systemic disparities. In less regulated realities, cost-containment strategies may drive the adoption of inefficient equipment. This introduces the risk of fluctuating ozone concentrations, which could prove hazardous to human operators, agricultural productivity, and the integrity of soil ecosystems. Yet, it should be also considered that the pungent ozone odour can be smelled at an air concentrations of 40-100 μg m^{-3} [103,104], but significant health effects on humans usually appear above 200 μg m^{-3} for prolonged periods [105]. [98] has assessed the ozone concentration limits in the air in terms of symptoms and effects on human health as reported in Table 1.

Given these toxicological thresholds and the potential risks to human health, regulatory aspects become critical to ensure the safe use (i.e., acceptable risk when properly managed) of ozone in agricultural settings. Although there are ambient air quality directives in the European Union that set threshold values for ozone exposure on vegetation to protect crops, these are not tailored to direct applications of ozone in agricultural systems [106–111]. In particular, there is a lack of crop- and soil-specific regulatory thresholds that consider ozone's effects on non-target organisms and soil health [112]. Without such frameworks, large-scale adoption of ozone treatments in agriculture runs the risk of unintended environmental or health consequences. Thus, international agencies such as European Food Safety Authority (EFSA), US Environmental Protection Agency (EPA), FAO, and WHO have a potential role in defining safe use protocols that include exposure limits, application methods, monitoring guidelines, and environmental risk assessments. A harmonized framework remains a prerequisite for large-scale adoption. Consequently, further scientific efforts are imperative to provide regulatory agencies with robust, data-driven evidences. Such efforts are essential to inform and direct a harmonized, large-scale legislative response.

Table 1
Ozone concentration (μg m^{-3}) in the air: symptoms and effects on human health.

Ozone concentration	Symptoms and effects
20-40	Odour threshold
200	Distinctive odour, burning nose and throat
400-1000	Sight problems after a few hours of exposure
1000	Severe tracheal problems
2000–4000	Headache, chest pain, persistent feeling of thirst, and severe burning chest. Prolonged exposure leads to poisoning
10,000–20,000	Feeling of choking. Pulmonary oedema
30,000–40,000	Small animals perish after 2 h
>100,000	Humans go into a coma after inhaling ozone for 1 h

*1 μg m^{-3} is roughly equal to 2 μg L^{-1} (Modified from Ref. [98]).

4.5. Environmental costs of its production and use

The environmental costs of ozone production and use raise important questions regarding energy consumption and sustainability. While ozone generation demands electrical input, often dependent on the energy source, comprehensive analyses of the trade-offs between reduced agrochemical usage and the carbon footprint of ozone production are still lacking. Optimizing these parameters is critical to determine whether ozone application constitutes or not an environmentally friendly strategy. In this context, studies employing Life Cycle Assessment (LCA) methodology can provide valuable insights and contribute to a clearer understanding of the subject. According to an LCA (ReCiPe method – [113]) conducted on a scenario formulated on the basis of data obtained by the manufacturer during experimental tests (Caffini S.p.A., personal communication), taking as a reference one year of operational monitoring data (own data) of a hillside vineyard under average climatic conditions of central-southern Europe, a reduction in ecotoxicity indicators was observed. In particular, this entails a reduction in the values of the indicators relating to terrestrial, marine, and freshwater toxicity of 40.3%, 23.7%, and 32.7%, respectively [113]. As regards improvements in impact categories typically linked to the consumption of fossil fuels by internal combustion engines to power processes in the field, there are only minor changes (i.e., reduction lower than 5%), as the potentially lower number of conventional treatments can be offset by the longer operating times required for the ozonation phases and the greater volumes of water to be distributed. Even the possible use of larger tanks to reduce distribution times would require more powerful tractors with higher hourly consumption, with a consequent balancing of these indicators [114–116]. These considerations directly connect to the matter discussed in Section 5 (Interdisciplinary aspects), where LCA-based analyses highlight the potential toxicity reductions achievable with ozone adoption. Moreover, it should be highlighted that these unresolved questions underscore the critical need for systematic, multidisciplinary research efforts that integrate expertise from fields such as agronomy, pedology, soil chemistry, soil ecology, crop physiology, environmental science, microbiology, and engineering. Only through such comprehensive investigation, benefits, limitations, and potential risks associated with the use of ozone in agricultural systems can be fully elucidated. This holistic understanding is essential to optimize application protocols, ensure environmental safety, and harness ozone full potential as a sustainable tool in modern agriculture.

4.6. Operational and management aspects

The volume of water distributed during ozone treatments is generally 2-3 times higher than that normally applied in chemical treatments, even exceeding 0.70 m^3 ha^{-1} (Caffini S.p.A., personal communication). In fact, to achieve a more uniform distribution of spray within the canopy, it may be desirable to exceed the leaf dripping threshold, since in this case any potential drift of the product to the ground does not result in residue impacts due to the rapid ozone degradation. However, this entails: a) greater water consumption during the season; b) longer times for refilling and preparing the machine, also due to the need to carry out the ozonization process, which takes about 20-50 min m^{-3}, depending on the physicochemical characteristics of the starting water in terms of impurities, temperature, and pH, as well as the cleanliness of the tank.

Potentially, the same sprayer used for chemical treatments, once equipped with an ozonator, could be used for ozone treatments. However, practical experience suggests using two independent sprayers, precisely to prevent any residues of active ingredients remaining in the tank from compromising the performance of the process in terms of time and oxygen consumption. In terms of energy, the sprayer with ozonator requires negligible additional power. The laboratory staff of the Caffini S.p.A. (personal communication) has estimated an energy demand in the range of 0.08-0.20 kWh m^{-3}, supplied as 12 V direct current, which can be easily generated directly on board the implement.

To date, however, comparative field trials have not yet been able to identify clear advantages of ozone-based treatment strategy over conventional chemical-based ones. Under controlled conditions, the advantages are obviously easier to identify and replicate, but in field environments, because of spatial and climatic variability, the operator is faced with "chaotic" conditions (typical of open systems) in which the inaccurate or untimely implementation of management decisions often invalidates the occurrence of the expected potential performance improvements. Similar situations can be found, for example, in applications involving both the UV-C treatments and electrostatic sprayers. These operational uncertainties must also be considered in parallel with ongoing digitalization and precision-agriculture efforts (see Section 3.6), where ozone integration with sensor-based and automated systems could help mitigate variability and improve field efficacy.

5. Interdisciplinary aspects

Addressing global challenges such as enhancing agricultural sustainability, reducing chemical inputs, and promoting the use of renewable resources requires a comprehensive upscaling of cross-sectoral impacts. Integrating ozone-based technologies into broader agricultural and environmental management systems presents a promising avenue for achieving these goals. By considering the interconnected effects across sectors, including agriculture, public health, and environmental conservation, research can elucidate how ozone applications might contribute to systemic improvements in sustainability metrics while simultaneously mitigating reliance on synthetic agrochemicals. To realize the full potential of ozone technology, further investigation into advanced utilization techniques and methodologies is imperative. This includes the refinement of existing ozone generation and application systems to improve efficacy, safety, and cost-effectiveness. In parallel, advances in mechanical application and distribution systems are essential to fully assess the real potential of ozone in agriculture (Fig. 3). Ozone can be delivered in different ways, including gaseous injection into soil, aqueous or oily sprays, fogging, or drip irrigation, and each approach presents specific strengths and limitations in terms of efficacy, selectivity, and safety [12,79].

5.1. The digital frontier

Recent developments in precision agriculture allow these application methods to be enhanced with digital tools, such as automated sprayers equipped with sensors, robotics for site-specific treatment, and IoT-based monitoring of ozone concentration in real time. Integration with machine learning algorithms can support decision-making on dosage, timing, and application mode, adapting treatments to crop type, developmental stage, and environmental conditions [117,118]. Artificial intelligence can also be used to model the dynamics of ozone decay and interactions with plant tissues or soil microbiota, enabling predictive control of applications to maximize benefits while minimizing risks. In this context, the concept of the digital twin represents a particularly powerful innovation in agriculture [119]. Digital twins, defined as virtual replicas of agricultural systems that continuously integrate *real-time* sensor data, can simulate ozone dynamics, predict plant physiological responses, and optimize treatment strategies before field application, thereby reducing uncertainty and minimizing risks of over- or under-dosing. These tools can complement precision agriculture by providing a virtual testing environment that supports decision-making and improves efficiency [10]. Such advances align closely with the *One Health* paradigm, which emphasizes the interconnection between

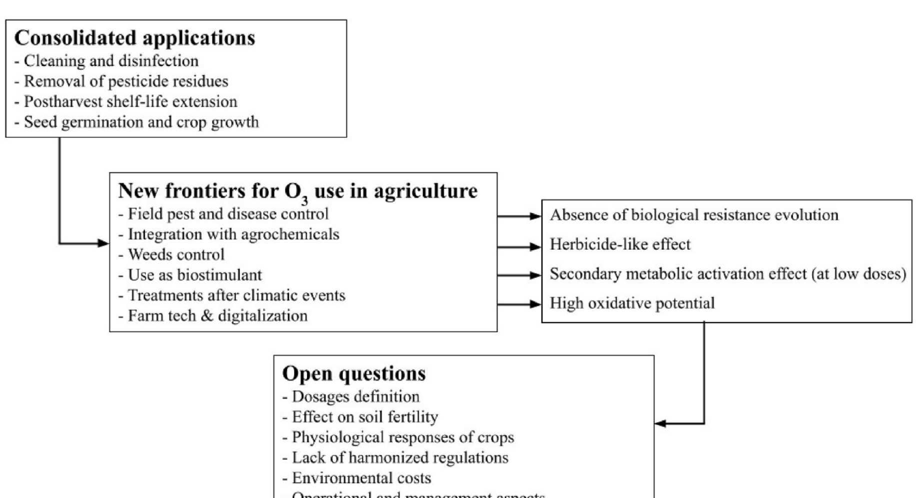

Fig. 3. Conceptual framework for ozone application in agriculture.

humans, animals, plants, and environmental health [120]. They also resonate with the European Union *Mission Soil* initiative, which explicitly promotes the regeneration of soil biodiversity and organic matter as key drivers of sustainable agriculture and climate resilience [94]. By embedding ozone-based strategies within a *One Health* framework, their benefits can be assessed not only in terms of agricultural productivity, but also regarding broader impacts on ecosystem services, environmental safety, and public health. In this perspective, within a context of increasingly intense and unpredictable climate change [121], ozone-based strategies may contribute to the development of antifragile agricultural systems [122], capable not only of resisting or recovering from disturbances, but of improving their functional performance through repeated exposure to environmental stresses.

These innovations collectively pave the way for more efficient, selective, and sustainable use of ozone within the broader framework of digital and climate-smart agriculture. Such interdisciplinary approaches facilitate real-time monitoring and adaptive management, maximizing benefits while minimizing unintended impacts.

5.2. Global Regulatory harmonization

From a regulatory perspective, understanding and harmonizing requirements across different national and international frameworks is essential for the widespread adoption of ozone-based agricultural interventions. Currently, only a limited number of countries have established specific regulations governing the use of ozone in agriculture and related sectors, while international standards are still under development. Investigating the regulatory landscape, including safety thresholds, permissible concentrations, and environmental impact assessments, will be critical to ensure compliance and foster public trust. Collaborative efforts among policymakers, scientists, and industry stakeholders could drive the establishment of unified guidelines, accelerating innovation and adoption on a global scale. Collectively, these interdisciplinary considerations highlight the need for integrated research efforts that bridge technological development, regulatory policy, and systemic sustainability goals, thereby unlocking the transformative potential of ozone technologies in addressing pressing global agricultural and environmental challenges.

5.3. Environmental benefits and climatic uncertainties

From an environmental assessment perspective (in accordance with LCA approaches, ReCiPe 2016 Midpoint), the main impact category in which the greatest improvements can be expected from ozone-based treatment strategies is that relating to toxicity indicators. In this context, reductions of up to 50% in agrochemicals consumption can be expected, together with a possible reduction in the number of treatments carried out during the season. However, there is still considerable uncertainty regarding the robust and stable adoption of ozone-based treatment strategies in open-field pest management. The main reason lies in increasing climate variability, which complicates management decisions regarding the ability to synchronize the timing of interventions with the development stages of plant pathogens. This difficulty risks favouring planned defence plans over targeted preventive plans implemented with technologies alternative to chemical approaches (including the aforementioned solutions with UV-C and electrostatic sprayers).

6. Possible limitations

Despite the potential of ozone-based technologies to support the transition towards sustainable and environmentally-conscious agriculture, several limitations currently act as bottlenecks to its widespread adoption. These primarily concern dosage optimization and economic feasibility [15]. Due to the species-specific nature of ozone, its use as a biostimulant or pest-control agent necessitates rigorous evaluations to define appropriate dosages based on the target species and specific application goals. Furthermore, the economic implications of infrastructure adaptation are critical factors. While aqueous ozone can be distributed using conventional equipment such as atomizers, ozone's inherent instability precludes storage and transport, necessitating integrated on-site generation. This requirement represents a potential point of criticality for the agrifood sector, as the production process must remain economically viable to ensure long-term sustainability.

7. Future research and perspectives

To order to overcome the main challenges related to the large-scale adoption of ozone in the agrifood sector, future research and perspectives must prioritize three main pillars: (1) comprehensive investigations into the physiological responses of target and non-target organisms to ozone exposure; (2) the calibration of standardized dosing protocols tailored to specific applications, ranging from herbicidal and pest control to biostimulatory effects; (3) advancement of on-site next-generation machineries for generation and distribution.

These priorities highlight the necessity of multidisciplinary scientific efforts to ensure a holistic understanding of ozone's potential, thereby facilitating the transition toward a more sustainable and resilient global agrifood system.

8. Conclusion

Ozone applications in agriculture represent an interesting and effective alternative to traditional chemical formulations and energy-intensive strategies. Despite its proven performance in cleaning, disinfection, pesticide residue removal, shelf-life extension, and crop growth support, the exploration of new frontiers (pest, disease, and weed control; use as a biostimulant; recovery tools after adverse climatic events) require further scientific efforts. These efforts are essential to establishing ozone as a cornerstone of sustainable agriculture, yet the transition from laboratory success to field-scale application carries profound technical and ecological implications. Specifically, the challenge lies in defining precise concentration thresholds that maximize oxidative efficiency without triggering phytotoxicity or detrimental stress responses in the plant. Furthermore, the urgent need to understand both short- and long-term impacts on soil microbial communities is critical, as any disruption to the balance of soil fertility could offset the environmental gains of reducing synthetic chemical inputs. Evaluating the full life-cycle costs, including the energy requirements for on-site ozone generation and the durability of delivery systems, remains a priority for ensuring economic viability. Ultimately, the current lack of standardized international regulations and clear legislative guidelines acts as a systemic barrier, preventing the widespread integration of ozone into certified organic or integrated pest management protocols and slowing the transition toward a truly residue-free food production system. Overcoming these challenges will require not only technological innovation, but also harmonized regulatory frameworks, stakeholder collaboration, and long-term field validation. Nevertheless, it should be clear that the use of these new technologies requires greater technical awareness from operators. Their technological development should proceed in line with the trends imposed by digitalization to promote the precision-oriented approaches required by smart agriculture practices (information management, automation, site-specific dosages). If effectively integrated, ozone has the potential to become a transformative tool for sustainable crop protection, bridging technological progress with environmental and societal goals.

CRediT authorship contribution statement

Leonardo Verdi: Writing – review & editing, Writing – original draft. **Gilmo Vianello:** Supervision, Conceptualization. **Simone Orlandini:** Writing – review & editing, Writing – original draft. **Youry**

Pii: Writing – review & editing, Writing – original draft. **Pasqualina Sacco:** Writing – review & editing, Writing – original draft. **Fabrizio Mazzetto:** Writing – review & editing, Writing – original draft. **Stefano Cesco:** Writing – review & editing, Writing – original draft. **Giuseppe Corti:** Writing – review & editing, Writing – original draft, Supervision, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Data availability

No data was used for the research described in the article.

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