Ozone applications in agriculture and agrifood

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This presentation briefly reviews the current state of art of emerging applications of ozone in agriculture and food processing.

There are many applications for ozone as an alternative to chemicals. In the field or in greenhouses, this molecule is effective in protecting crops. For example, Venturia inaequalis is the cause of the main apple disease which is named apple scab (Bowen et al., 2011). This disease causes the appearance of black spots on leaves and fruits. If these symptoms appear early in the season, the fruits develop in an abnormal way. Later contamination results in unsightly stains that prevent commercialization. In all cases, farmers face significant economic losses. Currently, about sixteen chemical treatments are spreaded by year in France to protect orchards against this fungus. However, V.inaequalis has developed resistance to several molecules such as dodine, kresoxim methyl, myclobutanil, or thiophanate-methyl as well as their mixture (Chapman, Pathology, Lafayette, & Sundin, 2011). Moreover, this chemical management is today decried because of environmental and sanitary risks.

Ozone can be considered like a good alternative (Guzel-Seydim, Greene, & Seydim, 2004). This molecule is well-known for its disinfectant capacity and its low remanence compared to conventional fungicides. A lot of studies have already been done to verify ozone antifungal abilities against fungal spores (Antony-Babu & Singleton, 2009; Sharpe et al., 2009). As V.inaequalis as concerned, we have previously shown the anti-fungal effect of ozone dissolved into water against conidia of this pathogen, thanks to in vitro tests and experiments carried out on artificial leaves. However, after its arrival on vegetal material (leaf, flower, or fruit), spore quickly germinates and interacts with the plant and the environment. In our knowledge, no experiment has been already done on V.inaequalis response face to ozone application when the treatment is done on inoculated baby apple trees. That is why, before considering a transfer to the field, it is necessary to evaluate the anti-fungal effect of water loaded with ozone on young plants contaminated by V.inaequalis under controlled conditions.

Our works showed that, the water loaded with ozone has an anti-fungal effect against development of V.inaequalis spores on young plants in controlled conditions. Ozonated water performances observed on baby plants are less remarkable than those seen on artificial leaves. Some ways can be explored to improve the efficiency like by increasing ozone dose or by adding adjuvants to maintain the liquid on the vegetal surface.

Ozone could be an interesting molecule at the service of the grapevine, from the nursery to the vineyard. Different experimentations have been performed and have showed promising results: i) ozone gas application during 34h to 25 days on grafted grapevines in nursery has led to the reduction of fungal developments on the plants. ii) Ozonated water has also been tested in nursery. Irrigation with ozonated water have stimulated the roots growth in number and weight and have reduced the time needed for buds to break. Plants soaked in ozonated water have a similar recovery rate than the plant treated with fungicide treatment. iii) Another study investigated whether ozonated water could be used to control conidia dispersal of the esca-associated fungus Phaeoacremonium aleophilum. Fungal development after artificial inoculation was significantly reduced by 50% in planta with ozone treatment. iv) Finally, ozonated water can be interesting for the control of downy mildew in the vineyard. For all these applications, the ozone molecule has proved its usefulness to produce healthy grapevines and replace or reduce the use of conventional plant protection products.

Applied dissolved in water or in gaseous form, it also reduces pathogens and phytosanitary products present on fruits and vegetables, increasing shelf life, reducing storage losses and consumer exposure to residual phytosanitary products.

During storage, microbial contamination of fruits and vegetables can cause serious economic losses. Fruit growers and industrials are therefore looking for solutions capable of effectively reducing pathogenic microorganisms and improving the fruit preservation while leaving little or no residue in the product (Aslam et al, 2020). Among the emerging strategies, ozone has shown favorable results on extending shelf life of fruits (Barboni et al., 2010 ; Tabakoglu and Karaca, 2018 ; Skog and Chu, 2001).

Our studies investigated the effect of continuous ozone injections during cold storage period on apples, plums and melons as a non-persistent alternative to chemicals. Fruits were placed in three downscale chambers of 12 m3 with or without gaseous ozone at two different concentrations. The experiments on apple were conducted on different varieties (Ariane and Rosy Glow). Apples were stored for periods ranging from 1 to 6 months under ozone and ultra-low oxygen atmosphere at 1.5°C. Plums were also stored in the same atmosphere conditions during 5 weeks at 2°C. On melon, experiments were conducted under ozone in normal cold-storage conditions at 8°C during 24h to 3 weeks. Fruit quality was assessed just after ozone treatment and after a storage at room temperature by physico-chemical and microbiological analysis. On apple, these evaluations were completed with biochemical tests, and sensorial analysis.

These works confirmed the results previously obtained by showing a reduction of the number of infected fruits in the ozone chambers, corroborated by the microbiological analysis. A reduction of up to 90% of the fungal flora was observed on melons and apples with the highest ozone dose. Above certain ozone concentrations, phytotoxicity occurred. Furthermore, results of the sensory analysis differentiated ozone and control samples with a tendency of control samples being blander.

Banana is the most important exported fruit in the world and therefore travels long distances between the packinghouse and the consumer. During these phases, bananas are vulnerable to a variety of post-harvest diseases, including fungal pathogens. The major one is crown rot disease which is caused by a fungal complex that includes Colletotrichum musae, one of the most pathogenic fungi (Ewané et al., 2012; De Lapeyre de Bellaire and Mourichon, 1997). Nonetheless, the effectiveness of the storage measures is not always guaranteed, and the management of these fungal diseases still relies on the use of chemical fungicides, which are currently strongly criticized for their health and environmental risks. Ozone (O3), due to its oxidizing and antimicrobial properties as well as its low persistence, is an interesting candidate for this type of application (Gutiérrez et al., 2018; Karaca and Velioglu, 2014). Ozone can be used as gas or dissolved in water as ozonated water for post-harvest sanitization (Pandiselvam et al., 2021; Tzortzakis, 2016). Within the framework of the ATMOZFR project, a laboratory container equipped for the post-harvest treatment of fruits and their preservation with ozone has been built. After sea shipping in Ivory Coast, this reefer was dedicated to banana fruit treatment for one year.

This mobile tool allows to perform experimentation on fruits for various applications from harvest to market. To conclude on these first experiments, the results show that bathing bananas in ozonated water from 2 to 10 minutes reduced the crown rot area of bananas significantly when compared to washing in regular water. In addition, storing bananas in a low O2 atmosphere with stable ozone concentration for 10 days reduces the crown rot area compared to a controlled atmosphere without ozone. Nevertheless, with the highest concentration, symptoms of phytotoxicity were observed. Therefore, an application of ozone at higher concentration during a short time could be interesting.

Finally, applications on fresh produce such as meat and fish are also widespread and will be briefly presented. Ozone also offers the food industry an alternative or complement to cleaning and sanitizing agents.

Better clarification of ozone's regulatory status worldwide for agricultural and food applications would greatly facilitate its adoption. Indeed, ozone is recognized as GRAS in the USA and authorized for direct contact with all types of food matrix. In Europe, ozone disinfection of food products is not authorized. At the same time, ozone is recognized as a biocidal product for treating water (swimming pool, bottled water, drinking water, cooling water).

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