

## The use of phosphates in forestry

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**Abstract.** Phosphite preparations are now an important alternative in plant protection against new, invasive pathogens of the genus *Phytophthora* and/or *Pythium*. It is crucial to intervene when alien, invasive oomycetes are carried to plantations or forest stands and attack fine roots via zoospores. The aim of this paper was to demonstrate the possibility of phosphite application to induce resistance to tree pathogens. Phosphate-based fertilizers have been used successfully in nurseries, where application is relatively easy by means of foliar sprays. The traditional fungicides, which are effective in combating fungi, however, fail to control oomycetes. Instead, they mask the disease, which, in turn, causes serious damage to seedlings after they have been planted in a suitable environment. Moreover, the number of effective fungicides available for forest plant protection has continued to decrease in the last decade. The effectiveness of the chemicals is reduced due to their frequent use and their similarity in terms of the active compound or the mechanism of action. Given the low diversity of active compounds, it is necessary to monitor the development of resistance of pathogens to fungicides by means of molecular biology (sequencing and quantitative PCR). Minimising the undesired side effects of chemicals on both, mycorrhizal fungi and pathogens can be achieved by strict adherence to rigorous security measures and, where possible, frequently changing the active compounds to alternatives such as phosphites. The significance of phosphate and phosphite uptake by trees is still a matter of debate, especially under field conditions. Nevertheless, phosphites are environmentally friendly compounds, which constitute an alternative or complement to the traditional chemicals (in accordance with the Directive on Integrated Plant management).

**Keywords:** Pathogens, plant protection, fertilizers

### 1. Introduction

Until recently, it was believed that phosphites interact toxically only in direct contact with pathogens (e.g. genus *Phytophthora*), and should be therefore applied directly on the site of the infection, and in relatively high concentration (Fenn, Coffey, 1984; 1985). However, detailed studies have shown that there is no relationship between the concentration of phosphites and its effectiveness, and the use of high concentration was completely non-toxic to fungi (Smillie et al. 1989). Further studies revealed that the application of phosphites reduces sporulation of organisms belonging to the genus of *Phytophthora*. As a result of their action, plant cell walls are altered and the amount of suppressors masking

the development of disease. The result of these studies was the creation of many fertilizers based on phosphite (Lovatt 1990). The research on the use of fertilizers in order to reduce the incidence of tree dieback was carried out in Germany in 2006–2009 by Jung (2008). In Poland in 2009, the Institute of Pomology and Floriculture was carrying out research on the use of the fertilizer named Actifos, which contains ammonium phosphonates in its composition, in plant protection (Orlikowski 2004, 2006; Korzeniowski, Orlikowski 2008; Muszyńska Orlikowski 2010). A similar work was also carried out at the Forest Research Institute (Tkaczyk et al. 2014).

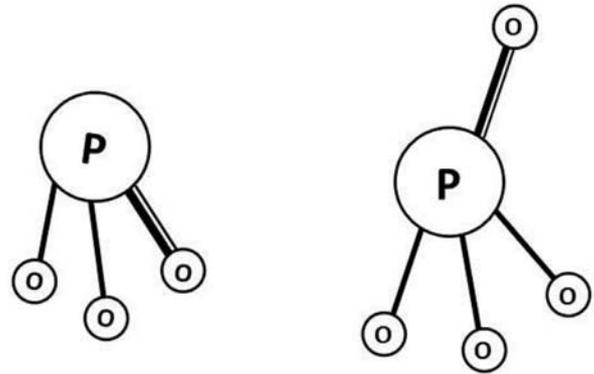
The purpose of this paper is to summarise the results of the research on phosphite in three aspects: as fertilizer,

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fungicides and bio-stimulators (resistance), and indicate the possibility of their use in forestry (e.g. in the context of the EU directive on integrated plant).

## 2. Phosphites used as fertilizer

Phosphates, as salts of phosphoric acid  $H_3PO_3$  (orthophosphoric III), differ from phosphoric acid  $H_3PO_4$  (orthophosphoric V) with one oxygen atom less (Fig. 1). This difference gives phosphites greater mobility in the soil and in plant tissues than phosphates and greater ability to penetrate the plants through leaves, stems and roots, and thus are more easily retrieved and transported through the xylem and phloem (Guest, Grant 1991). Thoughts about the use of phosphonates by plants as a phosphorus source are divided. Thao and Yamakawa (2010) cites studies of MacIntire et al (1950), which showed that phosphites are a source of phosphorus. Thao and others (2008) explored the usefulness of phosphite as a fertilizer in crops of spinach. They showed that in a situation where delivered to plants both forms of phosphorous and phosphite in sufficient doses, the symptoms of lack of phosphorus were not observed. However, when the only source of phosphorus was replaced by phosphite, this led to a reduction of growth of spinach. A similar relationship was observed in crops of *Brassica rapa* var. *perviridis* (Thao et al. 2008; Thao Yamakawa, 2009, Thao Yamakawa, 2010). Other researchers confirmed that phosphite may be applied as a source of phosphorus in sweet potato tissue cultures (Hirosse et al. 2012). In studies of Moor et al (2009), the use of phosphites as fertilizers for strawberries did not affect the plant growth or increase their fruiting. Avila et al (2012b), on the example of corn, demonstrated that an application of phosphites as fertilizers reduces the possibility of phosphorus uptake in its pure form, regardless of the nutrition with phosphorus. Replacement of a quarter dose of phosphorus fertilization by phosphite led to a reduction in the production of plant biomass showing deficiency of phosphorus. This effect was not observed in the experimental variant, when the plants are properly nourished. Similar studies on the effect of foliar application of phosphite on corn growth was carried out by Schroetter et al (2006) who demonstrated that inadequate supply of plant in phosphorus led to their stunting, or even dying. This effect was much smaller when the plants were properly nourished with all necessary nutrients. In studies of Ratjen and Gerendás (2009) courgettes, which showed a deficiency of phosphorus, were severely damaged in the variant, which uses phosphite fertilization. Other studies on the effects of phosphite fertilization on the biomass yield of bean (Avila et al. 2012) showed that at the time when the phosphite is added in small amounts, no significant changes in the plant biomass was observed, whereas increasing



**Figure 1.** The Phosphite ionic compound (left) and the phosphate ionic compound (right), modified by Guest, Grant (1991)

the dose would cause its significant decrease. These tests were carried out on both plants having as well good as poor nutritional status of phosphorus. In a variant of phosphorus deficiency in plants the foliar fertilization was applied, which led to their damage (Avila et al. 2012). Generally, it is believed that the phosphites are very stable chemical and are not transformed to phosphorus that can be used by plants as a food source (Ouimette, Coffey, 1989). This is a single bond between carbon and phosphorus, which is resistant to hydrolysis and is not readily biodegradable (Othake et al., 1996; Kelderer et al. 2006). So far, no plant enzyme capable of oxidising phosphites to the phosphorus form was found, although isolated bacteria from the roots of avocado have the ability to oxidise phosphites in plant tissues (Brunings et al. 2012). Also, bacteria of similar characteristics were located in the soil (Stoven et al. 2007). They can include species such as *Escherichia coli*, *Klebsiella aerogenes*, *Agrobacterium tumefaciens* and bacteria of the genera *Pseudomonas* and *Rhizobium*. However, this process is dependent on a number of environmental factors: temperature, soil moisture and pH, but the period of phosphites oxidation can last from a few weeks to even years (McDonald et al., 2001a).

## 3. Phosphites used as a fungicide

In the 1970s more attention was paid to phosphite as a potential means for protection of plants (Barchietto et al., 1989; Martin et al. 1998; McDonald et al., 2001b). Then, Rhône-Poulenc Agrochimie, a French company, introduced a new fungicide to protect the plant against downy mildew and diseases caused by organisms of *Phytophthora* genus (Fenn, Coffey 1987; Landschoot, Cook, 2005; Ann et al. 2009; Wojdyła et al. 2010; Wojdyła et al. 2011). The product comprised fosetyl-aluminum as an active substance. The action of preparations containing in its composition

phosphonates against Oomycetes (genera of *Phytophthora*, *Pythium*, *Peronospora*, *Bremia* and *Plasmopara*) was reported by many authors (Pankhurst et al. 1998; Aberton et al., 1999; Dobrowolski et al. 2008; Amiri et al. 2009; Ann et al. 2009; Hardy, 2009; Wilkinson et al., 2001a). Ouimette and Coffey (1989) observed *in vitro* the influence of five different compounds (having in their composition the phosphite) on 34 isolates belonging to nine different species of *Phytophthora*, and Wilkinson et al (2001b) under the same conditions studied the influence of potassium phosphite for 71 isolates of *Phytophthora cinnamomi*. Generally, the sensitivity of pathogens to phosphites was growing with increasing concentration of the preparation, although some isolates responded poorly to increasing doses. Reducing the development of *Phytophthora plurivora* and *P. cinnamomi* through the use of phosphites *in vitro* described Fenn and Coffey (1984). This mechanism was related with the growth reduction of pathogen cells and its sporulation (Griffith et al. 1990; Niere et al. 1990; Soulie et al., 1995; Davis, Grant 1996; Jackson et al. 2000; Matheron 2000; Wilkinson et al., 2001c). Coffey and Joseph (1985) noted the impact of phosphite preparations on the growth and development of *P. plurivora* and *P. cinnamomi*, including the significant inhibition of mycelial growth, reducing the formation of oospores (up to 97%) or two- to threefold reduction in the number of produced zoosporangia and halve the production of chlamydospores. This phenomenon is associated with the disturbance of pathogen metabolism by phosphite (Griffith et al., 1990; McDonald et al., 2001). Smillie et al (1989b), while testing the effect of phosphites against *P. cinnamomi* on lupine, *P. nicotianae* on tobacco and *P. palmivora* the papaya, also confirmed that a suitable concentration of the phosphite can protect plants against these pathogens.

#### 4. Phosphites used as a stimulator (resistance and growth)

Pscheidt and Ocamb (2013) observed that the concentrations of phosphite in plants that managed to protect themselves against infection were lower than these, which were considered to be needed to inhibit the growth of pathogens *in vitro* and therefore they hypothesised that there are plant defense systems interacting with the phosphites to reduce the growth of pathogens. The activation occurs at the time of contact with the pathogens. In addition, to direct influence on the growth and pathogens, the phosphites reduce the production of suppressors masking the presence of pathogens and thus accelerate the defense responses of plants (Yamada et al. 1989). The contact of plant tissues with pathogens in the presence of phosphites starts the plant defense reactions, for example phytoalexins synthesis in plant cells (Guest,

Bompeix 1990). The response of resistant or susceptible plant to infection is similar. The only difference is in the rate of formation of phytoalexins. The resistance mechanisms of the species are hereditary, and after recovering are gradually disappearing. The amount of produced phytoalexins depends on the age of the plant, temperature, oxygen content in the air and quantity of the inoculum (Fig. 2).

Further processes occurring in infected cells in the presence of phosphites are: ethylene biosynthesis, the increase of plant resistance to stress conditions, the increase in the activity of phenylalanine lyase and lignin biosynthesis (Guest, Bompeix 1990). The plant is able to isolate infected cells from the others (thanks to phytoalexins), which leads to reducing the spread of disease (Daniel, Guest 2006; Singh et al., 2003).

#### 5. Possibilities of phosphate fertilizers application in forestry

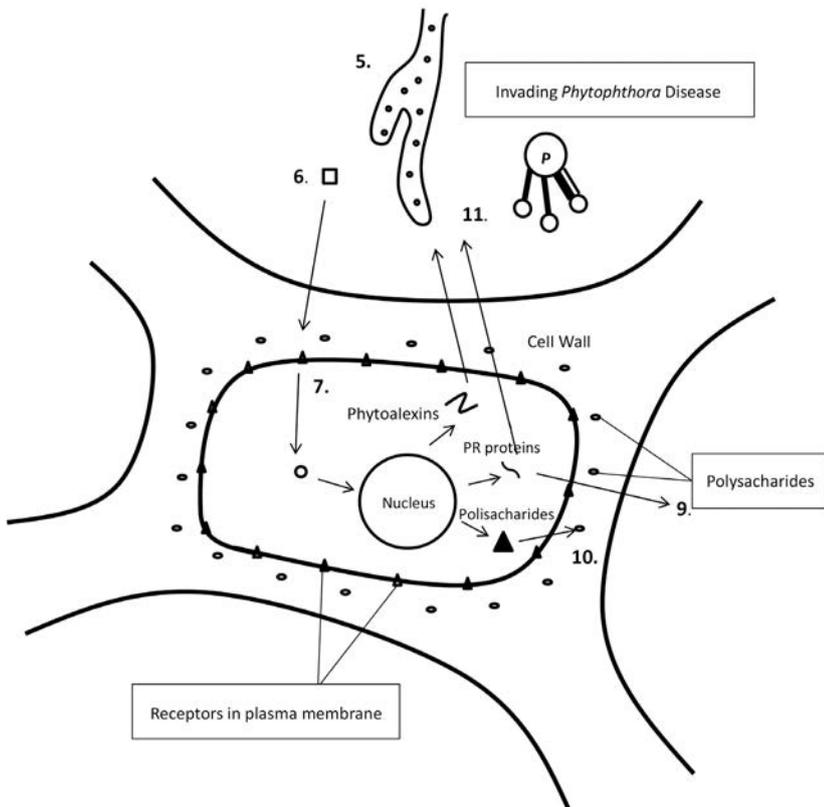
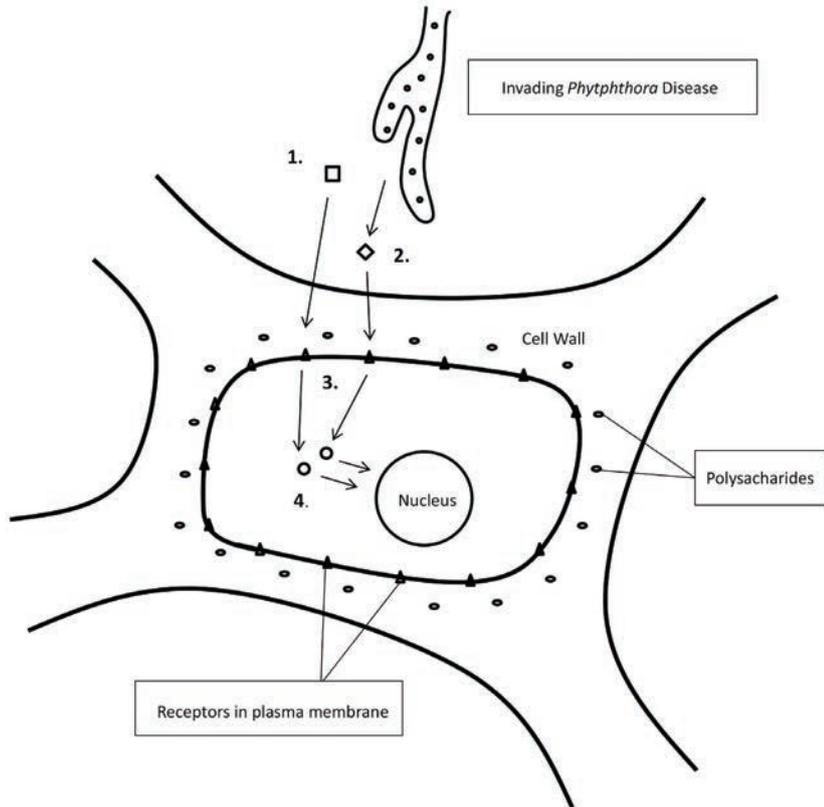
Initially, the test results using phosphites were not as good as with the use of phosphoric acid (MacIntire et al. 1950). Until the 1980s, the research on the use of phosphoric acid as a fertilizer in agriculture have been abandoned and focused on the clarification of its impact on improving the growth and health of plants. Because of the above-described characteristics, the phosphites are of great help in reducing the negative effects of soil-borne pathogens, attacking the fine roots of plants. In addition, in a current situation of a limited number of plant protection products approved for use in forestry, the use of phosphite is an important alternative solution in the context of integrated pest management (IPM). The Forest Research Institute on the order of the National Forests successfully completed a series of experiments in nurseries and within the scientific project EU Life + performed experimental spray-diseased oak stands in the Krotoszyn Plateau (western Poland). Preliminary results are encouraging, and their summary will occur in 2017, after the analysis of the crowns and roots of trees surveyed. If the use of phosphites proves to be a successful method in the experiment, it will attract a particular interest of the foresters to protect valuable adult oak forests, attacked by alien, invasive, pathogenic oomycetes.

#### Conflict of interest

The authors declare no potential conflicts.

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**Figure 2.** Reaction of a plant cell to *Phytophthora* spp., without phosphite (a) in the presence of phosphite (b) (Guest, Grant 1991):

1. Some molecules from the disease are recognised directly.
2. *Phytophthora* disease masks its recognition with suppressors.
3. Recognition fails at host-cell interface.
4. Only a weak signal goes to cell nucleus, this delays the plants' defence response.
5. Pathogen is affected by phosphite.
6. Suppressors either under or not produced.
7. Recognition of disease by plant cell.
8. Phosphite encourages defensive molecules, such as phytoalexins and PR proteins, to attack the pathogen directly.
9. Defensive molecules send alarm signals to cells that have not yet been attacked.
10. Polysaccharides strengthen the cell wall adding additional protection.
11. Pathogen is limited or killed by plant response.

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## Authors' contribution

M.T. – The concept and submitting work to develop text of the article, manuscript preparation, preparation of drawings; K.K., J.A.N., T.O. – The concept and submitting work to develop text of the article; JS – Review of the literature, developing text of the article.