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THE EFFECTIVENESS OF THE KAOLIN CLAY TOWARDS MONILIA FRUCTIGENA AND ALTERNARIA SOLANI UNDER LABORATORY CONDITIONS

Donyo Ganchev

Agricultural University – Plovdiv
Email: donyo@abv.bg

Abstract

Kaolin appears as odorless white to yellowish or grayish powder. Contains mainly the clay mineral kaolinite (Al2O3(SiO2)2(H2O)2), a hydrous aluminosilicate. It is used in the manufacture of china and porcelain and is also widely used in the production of paper, rubber, paint, drying agents, and many other products. It has a role as an excipient and an antidiarrhoeal drug and as an absorbent and filler including in some pesticide formulations. In this study, kaolin was tested under in vitro conditions against Monilia frucidena and Alternaria solani. The conducted tests have shown that it can be a potentially preventive fungicide against these plant pathogens. The substance was able to achieve full blocking of the germination of conidiophores and a growth of mycelium of Monilia frictigena at 0.1 % (m/v) concentration and at 0.3-0.5 % (m/v) towards conidiophores and mycelium of Alternaria solani.
Key words: kaolin clay, phytopathogens, antifungal activity

INTRODUCTION

The study of the antifungal possibilities of kaolin was conducted in the current research towards one of the most widely spread in the region of Bulgaria plant pathogens - Monilia frucidena and Alternaria solani. Kaolin appears as odorless white to yellowish or grayish powder. Contains mainly the clay mineral kaolinite (Al2O3 (SiO2)2(H2O)2), a hydrous aluminosilicate. It is used in the manufacture of china and porcelain and is also widely used in the production of paper, rubber, paint, drying agents, and many other products (National Center for Biotechnology Information, 2021). It has a role as an excipient and an antidiarrhoeal drug and as an absorbent and filler including in some pesticide formulations. (Ross & Kerr, 1930; Prasad et. al., 1991). Bulgaria is one of the countries rich in kaloin along with the United Kingdom, Germany, the Czech Republic, Spain, Malaysia, Pakistan, Vietnam, Brazil, etc. (Adamis et. al., 2005). The main use of kaolin (about 50%) is about the production of paper, but other uses include the following: ceramics (it is the main component of porcelain), toothpaste manufacture; it is an indigrient in different cosmetics products; also used as adsorbents in water and wastewater treatment, etc. (Murray, 2000). In medicine kaolin is used mainly for treating upset stomach and diarrhea (Pieszka et. al., 2016; Portnoy et. al., 1976; Alderman, 2005). According to the Safety Data Sheet of the product (according to Regulation (EC) No. 1907/2006) (Kaolin powder. Safety Data Sheet, 2019), kaolin is not a hazardous substance or a mixture according to Regulation (EC) No. 1272/2008, and no ecological problems are to be expected when the product is handled and used with due care and attention. Although not popular, it is used in farming as a spray applied to crops to deter insect damage, and in the case of apples, to prevent sun scald (Knight et. al., 2001; Alavo et. al., 2010; Lalancette et. al., 2005). Hoewever, some other studies show a direct antifungal (fungicidal action) and antibacterial action of the clay towards Aspergillus flavus, Saccharomyces cerevisiae (Shehu et. al., 2019), Brettanomyces and acetic acid bacteria in wine (Izquierdo-

Cañas et. al., 2018). The kaolin was founded to have a direct insecticidal action towards Callosobruchus maculatus under laboratory conditions (Vojoudi et. al., 2014)

The aim of the present study is to test the effectiveness of kaolin dust towards the plant pathogens Monilia frucidena and Alternaria solani in in vitro trials.

MATERIALS AND METHODS

The plant pathogens cultures were received by isolation from infected fruits quince tree (Cydonia vulgaris) - for Monilia fructigena; from tomato plants (fruits) – for Alternaria solani. The kaolin dust was bought from Talloderma Ltd – a Bulgarian cosmetic company (THALLODERMA 2019). The initial starting concentration was 1.0 % and in the case of high or low effectiveness towards a given test pathogen, this concentration was increased or decreased in the next trials. Germ tube inhibition tests were conducted in order to determinate a possible protective activity of the tested substance (kaolin clay) towards Monilia fructigena and Alternaria solani. The fresh infected with the inspected pathogen plant parts was collected and incubated under the conditions of humid chamber with the purpose of stimulating the conidial sporulation of the plant pathogens. A conidial suspension was prepared with density 3*104 spores / ml. A microscopic slides kind “hanging drop” was sprayed with the solution of the potassium sorbate and after drying, 20µl from a conidial suspension was applied. The slides were incubated under the condition of humid chamber in thermostat and after 24-48 h. the number of the germinated conidia was counted with a light microscope (magnification 100x). The effectiveness was determinated by the formula of Abbot (Abbott, 1925). The tests for determination of the effect of the inspected kaolin clay to inhibit a development of mycelium of tested phytopathogens was by modified method of Thornberry (Thornberry, 1950). For this purpose, a potato – dextrose agar (PDA) was used and the tested substance was mixed with the growth media at the given concentration. Standard 10 cm diameter Petri dishes were used for this purpose. After the cooling of the mixture media/test substance, an inoculation disk from the pure culture from the tested pytopathogen was placed on it. The observation of the growth of the mycelium was conducted with a ruler on 3, 7, 10 and 14 day after the beginning of the tests by measuring the growth area around the inoculations disks by the ruler. Area Under Disease Progressive Curve (Simko & Pieph, 2012) was calculated and on the basis of these indexes the effectiveness was determinated by using the formula of Abbot. The statistical manipulation of the results (One-way ANOVA) was made with R Program Language for Statistical Computing (Team C., 2013).

RESULTS

Conidial Tests

The conducted tests with conidiospores show that the kaolin dust can achieve full blocking of conidiospores of Monilia fructigena germination at 0.1 % concentration and at 0.3 % concentration for Alternaria solani and (Fig. 1, Fig. 2, Fig. 3 and Fig. 4):

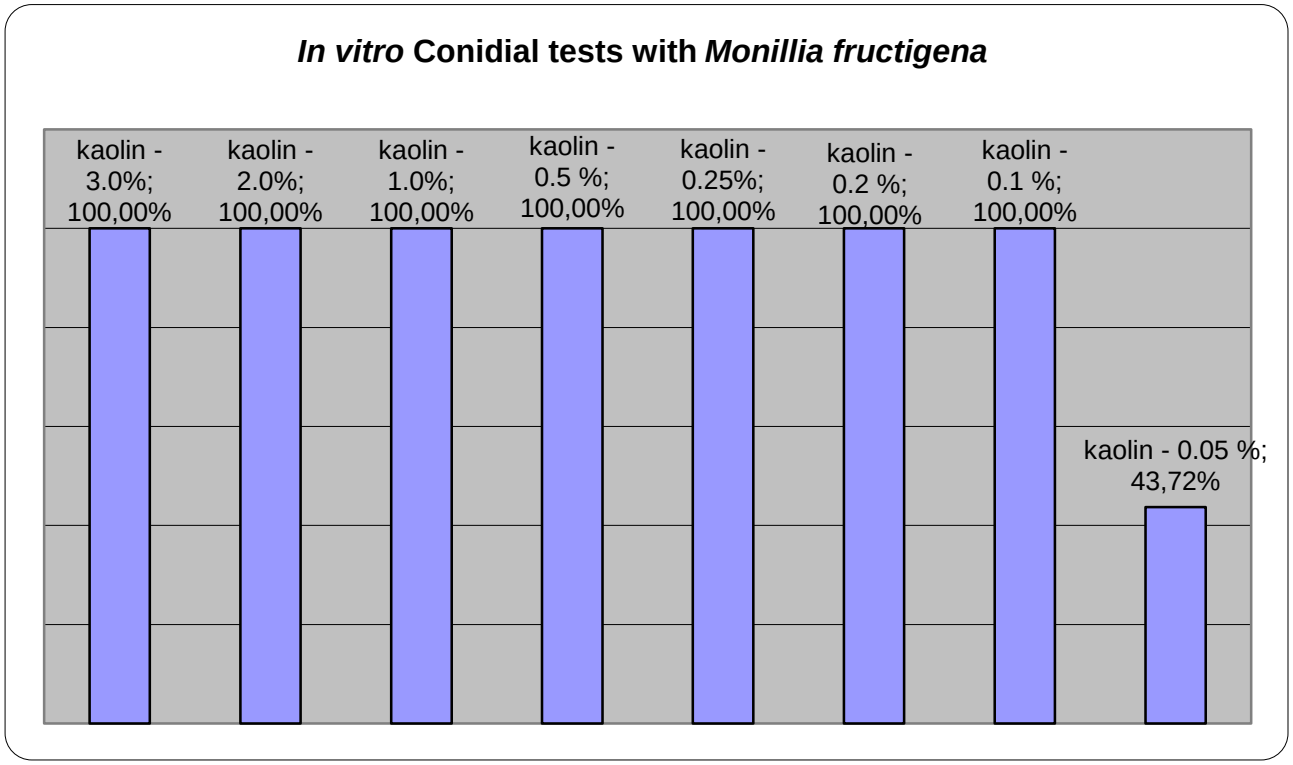

Fig. 1. Effectiveness of the kaolin dust towards conidiospores of Monilia fructigena ($p<0.05$)

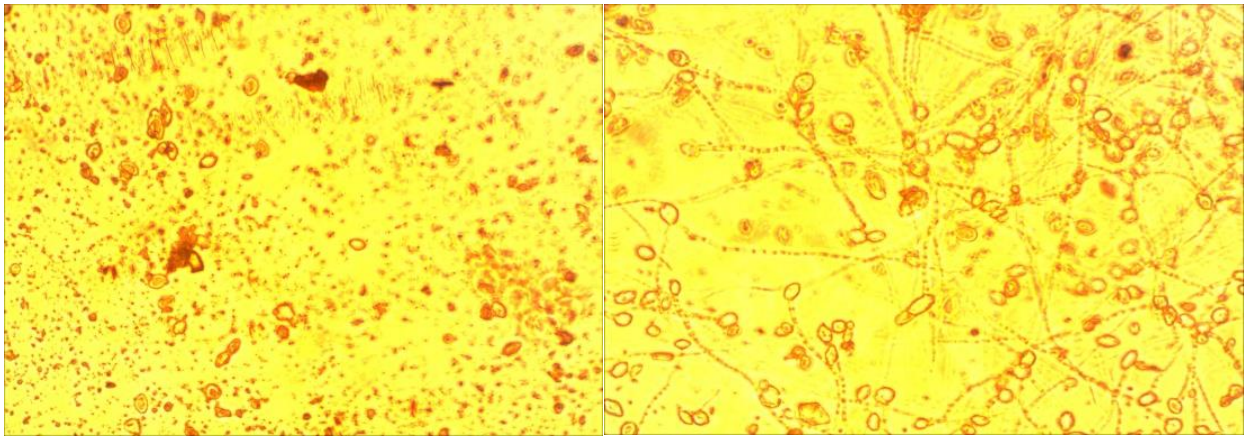

Fig 2. Monilia fructigena conidial suspension in vitro test. On the left – control variant; on the right – the kalolin water solution at 0.1 % (m/v) concentration

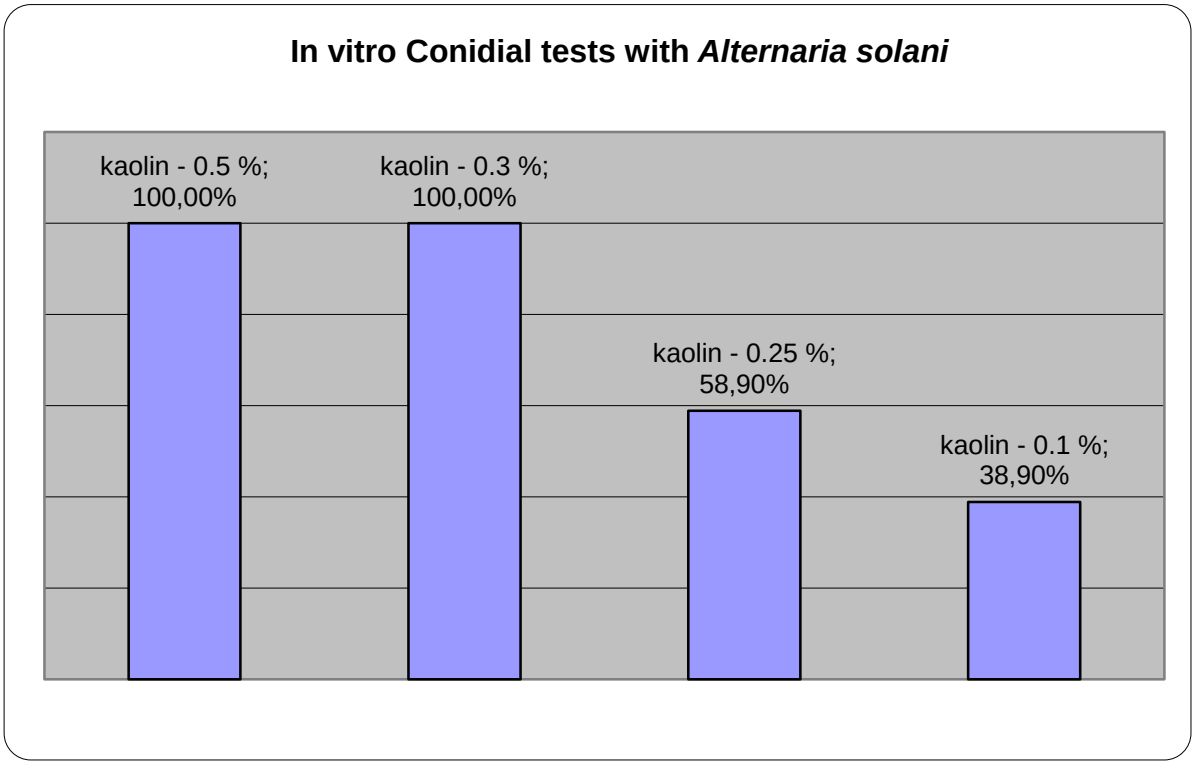

Fig. 3. Effectiveness of the kaolin dust towards conidiospores of Alternaria solani ($p<0.05$)

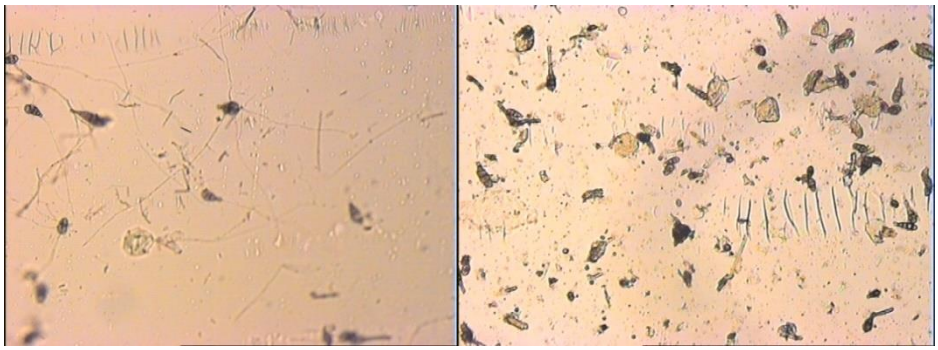

Fig. 4. Alternaria solani conidial suspension in vitro test. On the left – control variant; on the right – kalolin water solution at 0.3 % (m/v) concentration

Thornberry Tests

In the conducted Thorberry tests, again kaolin was completely effective at 0.1 % concentration against mycelium of Monilia fructigena (Fig.5 and Fig.6) and 0.5 % concentration against mycelium of Alternaria solani (Fig.7 and Fig. 8). However, at 0.25 % concentration the clay achieve over 80 % (84.9 %) effectiveness towards the same plant pathogen.

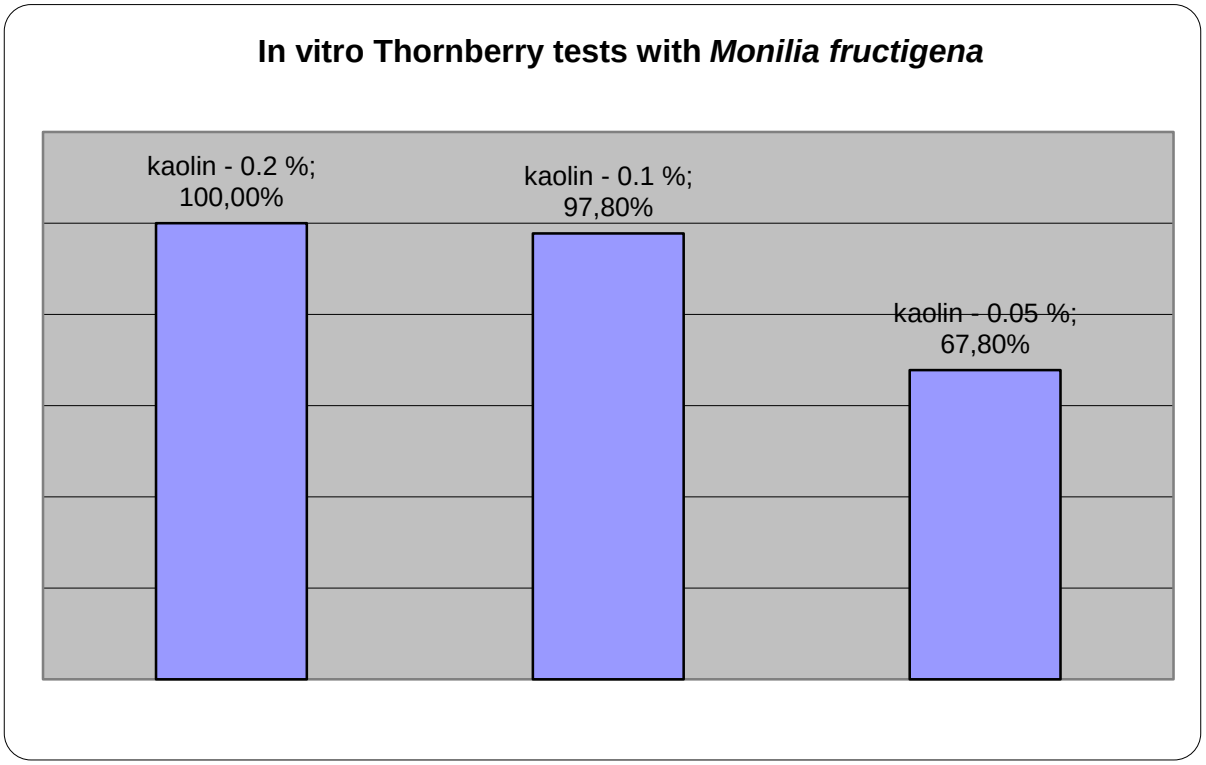

Fig.5. Effectiveness of the kaolin dust towards the mycelium of Monilia fructigena ($p<0.05$)

Fig.6. In vitro Thornberry tests with Monilia fructigena. In the left – control variant; in the right – kaolin at 0.2 % (m/v)

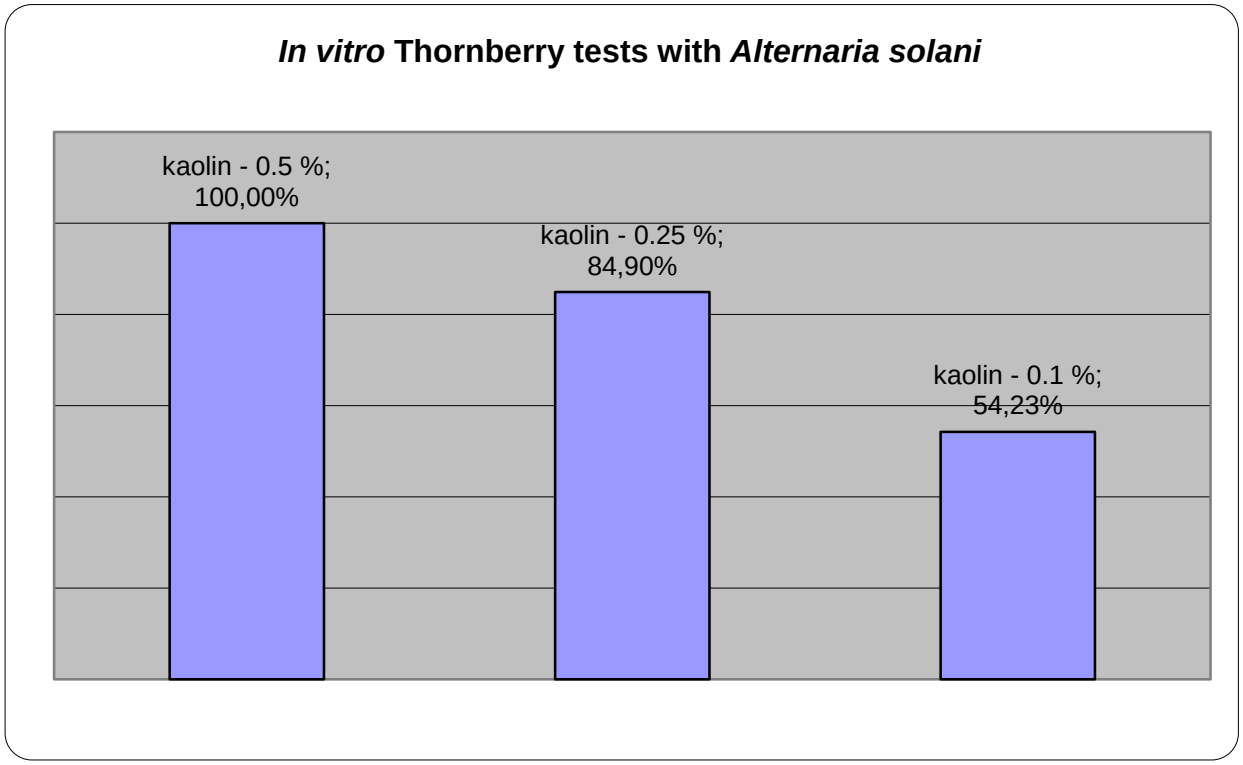

Fig. 7. Effectiveness of the kaolin dust towards the mycelium of Alternaria solaini ($p<0.05$)

Fig.8. In vitro Thornberry tests with Alternaria solani. In the left – control variant; in the right – kaolin at 0.25 % (m/v)

Just like in the conidial suspension trials, kaolin shows full effectiveness towards the mycelium of Monilia fructigena at 0.1 % (m/v) concentration. However, towards the mycelium of Alternaria solani, the 100 % effectiveness was possible at 0.5 % (m/v) concentration.

DISCUSSION

Conducted in vitro tests (conidial and radial growth assays) show that kaolin have a potential to be an effective preventive pesticide in the agriculture, not only as a physical barrier against insects and sun burn, but with a direct fungicidal action towards plant pathogens at

concentrations which are completely comparative with the commercial synthetic fungicides. The substance was able to achieve full blocking of the germination of conidiophores and the growth of the mycelium of Monilia frictigena at 0.1 % (m/v) concentration and at 0.3-0.5 % (m/v) towards the conidiophores and the mycelium of Alternaria solani.

CONCLUSION

Kaolin as a natural material is extremely abundant, easy to be produced, stored, cheap and available, easy for making water pesticide solutions. Plus the fact that it is completely non toxic for humans and environment, there is a big potential in the development of plant protection products on the base of kaolin that can be applied in the commercial agriculture as well as in the organic farming and the integrated pest management.

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