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Phytopathogenic fungi in seeds of the genus *Pinus*, stored in a gene bank

Hongos asociados a semilla del género *Pinus*, almacenada en banco de germoplasma

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Abstract

Phytopathogenic and saprophytic fungi present in seed with more than one year of storage were identified in the gene bank of the CONAFOR Jalisco delegation of the pine species: *Pinus douglasiana*, *P. devoniana*, *Pinus hartwegii*, *Pinus oocarpa* and *Pinus pseudostrobus*. The mycoflora present, its level of infestation and phytopathogenicity were identified. Physiological and sanitary quality was determined. The 5 species analyzed presented at least 4 genera of fungi: *Fusarium*, *Penicillium*, *Aspergillus* and *Rhyzopus* have been reported as storage fungi that cause seed deterioration. It is advisable to take measures to ensure sanitary quality, considering that the seed stored in the GDBs are the safeguard of forest genetic resources.

Phytopathogenic fungi in seeds, Seeds *Pinus* stored, *Pinus* genebank

Resumen

Se identificó la presencia de mico flora patógena presente en semilla con más de un año de almacenamiento en el banco de germoplasma de la CONAFOR delegación Jalisco de las especies de pinos: *Pinus douglasiana P. devoniana*, *Pinus hartwegii*, *Pinus oocarpa* Schiede y *Pinus pseudostrobus*. Con el objetivo de identificar la micoflora presente, su nivel de infestación y fitopatogenicidad. Se determinó la calidad fisiológica y sanitaria. las 5 especies analizadas, presentaron al menos 4 géneros de hongos: *Fusarium*, *Penicillium*, *Aspergillus* y *Rhyzopus* han sido reportados como hongos de almacén causantes de deterioro de semilla. es recomendable tomar medidas que aseguren la calidad sanitaria, considerando que la semilla almacenada en los BGF son el resguardo de los recursos genéticos forestales.

Hongos en semilla forestal, Semilla de *Pinus*, Bancos de germoplasma

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Introduction

Forest ecosystems provide products and services that contribute to human well-being, including food, timber, timber, medicinal plants, poles, firewood and fodder, among others, and environmental services such as the maintenance of water sources, biological diversity, climate regulation and carbon sequestration (Nordon, 2014).

Forest regeneration is the basis for species renewal and continuity; thus, production, dispersal, seed germination and recruitment are among the most important processes in the distribution and abundance of plant species (Nathan and Muller-Landau, 2000; Wang and Smith, 2002). Within temperate forests, the most abundant genera are oaks or Quercus spp. and pines Pinus spp. There are about 46 species of pines in Mexico (Sánchez-González, 2008); species widely used in their geographical distribution for their multiple uses.

Forest germplasm is a valuable and limited resource that is influenced by three factors: collection, management, and conservation, which are strongly related, as their effects are cumulative, with an increasing impact on seed quality (Vargas et al 2004). Since ageing is a natural and irreversible process, the higher the initial quality of the seeds and the better the storage conditions, the lower the rate of ageing and deterioration. In Mexico, the National Forestry Commission (CONAFOR) established the National Programme for the Management of Forest Genetic Resources in 2004, which sets out objectives and actions aimed at the conservation and sustainable management of forest genetic resources. Actions include the production of seeds of known origin or with genetic improvement (stands, seed areas and orchards) and germplasm banks (BGF). The BGFs have technical personnel and the necessary equipment to carry out the processes of collection, processing, storage and conservation of forest germplasm under controlled conditions of temperature and humidity, as well as the analysis of its physical and biological characteristics, with the purpose of conserving its germination potential. However, no analysis of sanitary quality or determination of fungal flora associated with the seed is carried out.

Fungi that lodge in seeds cause different types of damage; if the infection is very severe, the damage can lead to embryo death. With mild infections, seeds do not lose their germination power; however, their vigour may be affected. Fungi that have invaded seeds in the field become active again when the seeds are about to germinate, due to the high humidity present in the soil; some of these cause seed rot and seedling wilt. Many fungi do not cause problems to seeds and seedlings during germination and emergence, but are capable of causing the development of foliar, stem or fruit diseases, which reduce the quantity and quality of harvests (Moreno, 1995).

The objective of this work was to identify the diversity of pathogenic mycoflora present in seed with more than one year of storage in the germplasm bank of CONAFOR delegation Jalisco of the following pine species: Pinus douglasiana Martínez; Pinus devoniana Lindl.; Pinus hartwegii Lindl.; Pinus oocarpa Schiede ex Schltdl. and Pinus pseudostrobus Lindl. their level of infestation and phytopathogenicity.

Methodology

The physiological quality analyses were carried out in the seed laboratory of the germplasm bank of CONAFOR Jalisco state management, as well as in the seed laboratory of the University Centre of Biological and Agricultural Sciences (CUCBA) of the University of Guadalajara. Sanitary quality was analysed. Fungal quantification was performed by inducing mycelial growth and sporulation by incubation under the blotting paper or plotter protocol (Cimmyt, 2010) incubated at 27°C at 12-hour intervals of light and dark. Fungal identification was with the keys of Barnett and Hunter, 1998 and Cimmyt, 2010.

Results

The initial quality of the stored seed was low as the standard germination percentages obtained are within the range of 22 to 69 percent, taking into account that the seed was stored with a germination higher than 80%, it is considered with a considerable deterioration, Table 1.

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Species	Humidity (%)	Purity (%)	Feasibility (%)	Germination (%)
Pinus douglasiana	6.4	97.6	64	65
Pinus hartwegii	8	96.1	25	22
Pinus devoniana	6.9	99	64	54
Pinus oocarpa	11.3	89.2	66	69
Pinus pseusdutro	10.8	99.6	49	44

Table 1 Initial seed quality

In the 5 species analysed, at least 4 genera of fungi were present, of which: Fusarium sp., Penicillium sp., Aspergillus niger and Rhizopus, have been reported as storage fungi causing seed deterioration, generating abnormal seedlings and embryo death (Correa et al., 2012).

Species	Pathogenic flora encountered	% of infestation
Pinus douglasiana	*Aspergillus nigger*	18
	Penicillum	20
	Fusarium sp.	5
	Rhizopus ehrenb	12
Pinus hartwegii	*Aspergillus nigger*	40
	Chaetomium globosum	35
	Corynespora cassiicola	60
	Penicillium spp.	50
	Phoma westend	25
Pinus michoacana	*Aspergillus nigger*	20
	Chaetomium globosum	10
	Lasiodiplodia theobromae	5
	Penicillium	18
Pinus oocarpa	*Aspergillus nigger*	5
	Rhizopus ehrenb	4
	Corynespora cassiicola	12
	Phoma westend	8
Pinus pseudustrobus	*Fussarium moniliforme*	65
	Fusarium poae	48
	Lasiodiplodia theobromae	25
	Phoma westend	20
	Rhizopus ehrenb	15

Table 2 Fungal species identified on pine seeds

Fungi transmitted by conifer seeds can be classified into: saprophytes or weak pathogens; phytopathogens that infect and kill the seed embryo; pathogens of primary importance in seedlings; and phytopathogenic field fungi such as Fusarium spp. whose pathogenicity occurs in adult stages of the plant when conditions are conducive (Flores, 2010). Although most of these fungal genera are considered saprophytes, some of them do not always cause direct seed damage, but it is recognised that when the incidence is very high, seed vigour and viability tend to decrease (Mittal et al., 1990).

Although the management of seed in a germplasm bank ensures its conservation through low humidity and temperature, which prevents the development of microorganisms or pests, Campo-Arana et al. 2014, after analysing native forest seed from Colombia in germplasm banks, emphasise the importance of applying treatments to reduce the fungal load present, as well as the need to review the protocols for seed collection and storage.

Conclusions

Due to the incidence and pathogenicity of the fungal species found, as well as the loss of physiological quality of the seed, it is advisable to take measures to ensure its sanitary quality, especially considering that the seed stored in FGDBs are the safeguard of forest genetic resources.

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