THE INFLUENCE OF STORED PERIOD ON THE LEGUMES SEEDS IN THE CONTROLLED ATMOSPHERE UPON MICROMYCETES VIABILITY

INFLUENȚA DURATEI DE PĂSTRARE A SEMINȚELOR DE LEGUMINOASE PENTRU BOABE ÎN CONDIȚII DE MEDIU CONTROLAT ASUPRA VIABILITATII MICROMICETELOR

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Abstract. *The legumes seeds micoflora (pea and bean) which are stored the certain period in the controlled conditions (T=+4°C and relative humidity 70%) are decreased. In many cases the viability of the inocul depend of its age. The seed aging degree could generate a change in the relation parasite-seed pursuant of longevity different of the contamination micoflora. The experimental biologic material is represented by breeds and local landraces coming from different pedoclimatic areas and stored a time period at T=+4ºC. The used researchs methods were macroscopic analyze and CGA (potato-sucrose-agar). The obtained results were realized trough quantification the analyzed elements. The epiphyte and endophyte micromycets associated of the seed were evaluated trough the colonies counting and attack frequency which was estimated in percents. The effectuated analyses were processed through statistical methods in order to emphasize of some significant regressions, the study showing that of the micromycets evolution is hard influenced of the storage period increase of the analyzed seeds.*

Rezumat. *Micoflora semintelor de leguminoase pentru boabe (mazarea si fasolea) pastrate anumite intervale de timp in depozit cu atmosfera controlata (T=+4ºC si umiditate relativa a aerului 70 %) se reduce considerabil, viabilitatea inoculului depinzand in majoritatea cazurilor de virsta acesteia. Gradul de imbatrinire a semintei poate produce o schimbare in relatia cu parazitul ca urmare a diferentei de longevitate a micoflorei ce o contamineaza.*

Materialul biologic de experimentare s-a constituit din probe de seminte din soiuri si populatii locale colectate din diferite zone geografice si depozitate o perioada de timp la T=+4ºC. Metodele de cercetare aplicate au fost analiza macroscopica a semintei si metoda placilor de agar (CGA).Rezultatele obtinute s-au bazat pe cuantificarea elementelor analizate, micromicetele epifite si endofite asociate semintei s-au evaluat prin numararea coloniilor iar frecventa atacului s-a exprimat in procente. Determinarile efectuate s-au prelucrat statistic pentru evidentierea unor regresii intre diferite elemente, studiul reliefind faptul ca evolutia micromicetelor este influentata puternic de cresterea duratei de conservare a semintelor speciilor analizate.

INTRODUCTION

In our agriculture country, the bean and the pea are tradition cultivation by high nutrient value of the seeds who constitute the primary source of protein in the food of humans and animals to disposed of the majority locality of the favorable pedoclimatic conditions (Hulea A, 1973). However, the acquired productions on the big areas and on farms rural didn't upen the potential biologic level of the sorts cultured or presented fluctuations of the year at else. A one main cause of these situations may be the infection degree seeds. In the majority occurrence the existing inocul worn of the seed is viable in mayoralty of these age, on the fresh seeds been present in big percent, the infection reduced from the year at else or after many conserved years. The seed aging degree could generate a change in the relation parasite- seed pursuant of longevity different of the contamination micoflora (Raicu C, 1978). Departed from these situations this study dignifies the duration role of keeping about the micromycets longevity existing on the beans seeds and the stored peas at temperate four degrees plus certain period of time.

MATERIAL AND METHOD

The experimental biologic material is represented by breeds and local landraces who belong to two legumes species Phaseolus vulgare si Pisum sativum. Of the midst's samples of fresh seeds and the stored seeds thus took:

- a. for the bean took:- seventeen of the fresh seed which from three samples belong of the Ami, Astra, Avans breeds and fourteen locale landraces .-nine stored samples of the locale landraces.
- b. for the pea took:-twelve fresh samples and ten stored samples of the locale landraces.To make possible of this phytopatological study of mycromicets evaluation on the legumes seeds it was utilize the following research methods: the seeds macroscopic analyze; the Ulster method (Mallone; Muskett, 1964).

Beyond analyzed the macroscopic, was study seeds by Ulster method utilize the potato- sucrose- agar of the nutritive medium. The Petri dishes with seeds were incubated for 7 days at T=22°C, In the last three days was expose to bulb with fluorescent light for twelve hours per day. Beyond seven days of incubation the micromycets colonies existing on the seeds it was determine macroscopic and the conidiene forms by effectuated the microscopic slide.

RESULTS AND DISSCUSIONS

The experimental results have a statistic character indicating the isolated fungus genus with afferent regressions lines influenced by the seeds storage duration.

The studied biologic material belong of the species *Phaseolus vulgaris (table 1)* characterizing through:

-on the fresh bean seeds were identified 18 genus of the micromycets, with 592 colonies from which: 5 parasite genus: *Colletotrichum lindemuthianum, Isariopsis griseola, Sclerotinia sclerotiorum, Fusarium roseum, Rhyzoctonia solani;* 13 saprophyte genus: *Cladosporium herbarum, Alternaria alternata, Stemphylium botryosum, Epicoccum sp, Acremoniella verucosa, Acremoniella atra, Rhyzopus sp., Trichotecium roseum, Trichoderma viride, Penicillium sp., Stachybotrys atra, Papularia arundinis, Chaetomium sp.*

- on the seeds which were stored during the year 1992 were identified 6 genus of the micromycets, with 28 the colonies, such as: *Cladosporium herbarum, Alternaria alternata, Stemphylium botryosum, Epicoccum sp., Rhyzopus sp, Trichoderma viride* .

Table 1

Action mode of the pathogen and saprophyte mycromicets on the bean cultivar seeds studied in the two experimenthal conditions

The experimenthal conditions / The isolated genus of the micromycets	The fresh seeds from 2006	The stored seeds from 1992
	The number of the colonies isolated	
The parasite genus		
Colletotrichum lindemutiamum	54	0
Isariopsis griseola	27	0
Sclerotinia sclerotiorum	18	0
Fusarium roseum	19	0
Rhyzoctonia solani	5	0
The saprofite genus		
Cladosporium herbarum	9	3
Alternaria alternata	149	1
Stemphylium botryosum	30	6
Epicoccum sp.	28	2
Acremoniella verucosa	116	0
Acremoniella atra	10	0
Rhyzopus sp.	46	6
Trichotecium roseum	37	0
Trichoderma viride	26	10
Penicillium sp.	3	0
Stachybotrys atra	3	0
Papularia arundinis	9	0
Chaetomium sp.	3	0
Total	**592**	**28**

After fourteen storage years the parasite mycromicets were disappeared being identified only six saprofite mycromicets with a low colonies number: *Alternaria alternata, Cladosporium herbarum, Stemphylium botryosum, Trichoderma viride, Epicocum sp., Rhyzopus* sp. In the figure no. 1, it observe that the regressions lines shows the link between the stored period and the evolution of these six mycromucets. The action of the *Alternaria alternata* is much more influenced by the increasing of the storage period, then the action of the other micromycets.

Thus, through the increasing of the seeds storage period, the infection rate with *Alternaria alternata* is reduced much more.

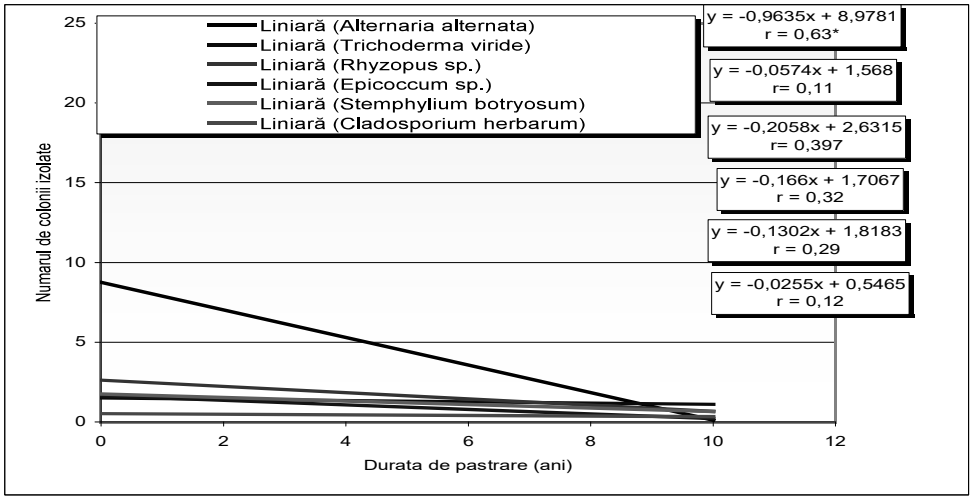

Figure 1 - Regression lines for correlation's between stored duration and isolated colonies at the identified micromicets on the analyzed bean seeds

On seeds of species *Pisum sativum* were obtained the following results (table 2):

- on the fresh seeds were identified 9 genus with 149 micromycets colonies on which: 2 parasite genus: *Ascochyta sp., Fusarium solani;* 7 saprofite genus: Cladosporium herbarum, Alternaria alternata, Stemphylium botryosum, Epicoccum sp., Rhyzopus sp, Gonatobotrys atra, Penicillium sp.

- on the stored pea seeds from year 1995 were identified: 7 saprofite genus: *Cladosporium herbarum, Alternaria alternata, Stemphylium botryosum, Epicoccum sp., Rhyzopus sp., Penicillium sp., Papularia arundinis.*

Table 2

Action mode of the pathogen and saprophyte micromicets on the pea landraces seeds studied in the two experimental conditions

The experimenthal conditions / The isolated genus of the micromycets	The fresh seeds from 2006	The stored seeds from 1992
	The number of the colonies isolated	
The parasite genus		
Ascochyta sp.	3	0
Fusarium solani	9	0
The saprofite genus		
Cladosporium herbarum	14	4
Alternaria alternata	72	13
Stemphylium botryosum	9	1
Epicoccum sp.	14	2
Rhyzopus sp.	24	5
Penicillium sp.	2	17
Gonatobotrys atra	2	0
Papularia arundinis	0	1
Total	**149**	**43**

In the wake of analyzes of the arises micromycetes on the seeds during of that two experiment periods it was observed that after 11 storage years in the environment controlled conditions the parasite mycromicetes disappeared and were reduced the number of the saprophyte colonies, such as: *Cladosporium herbarum, Alternaria alternata, Stemphylium botryosum, Epicoccum sp., Rhyzopus sp., Penicillium sp., Papularia arundinis.*

The regressions lines emphasizes that there is a negative correlation between the storage period of the seeds and the number of the isolated colonies, excepting the *Penicillium sp.* micromycete where exist a positive correlation.

It noticed, that the action of the *Alternaria alternate* (figure 2) micromycete is influenced much more by the increasing of the storage period in comparison with the action of the other four micromycets.Thus, if the storage period will increase, the infection rate with *Alternaria alternata* will be less.

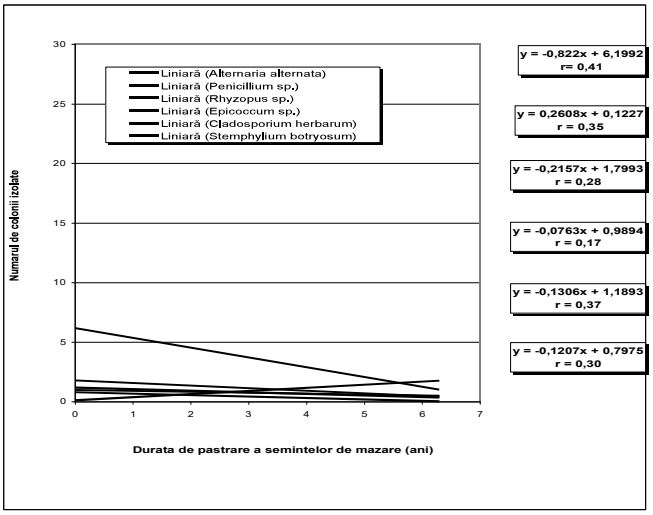

Figure 2 - Regression lines for correlation's between stored duration and isolated colonies at the identified micromicets on the analyzed bean seeds

CONCLUSIONS

The analyzes of the obtained results, emphasizes the different conduct of the pea seed samples in comparison with the bean seed samples. The number of the mycromicets genus existing both on the fresh and the stored seeds is much low at pea samples then bean samples, because of the tegument of this specie which is very hard determining the reduction of the penetration ways of the micromycets on and inside of the seeds.

At one time with increase of the storage period the viability of the some saprophyte fungous existing on the bean and the pea seeds was declined , and the viability of the parasite fungous decreased, until its total depletion.

The storage period of the seeds affected very strong the infection percent with *Alternaria alternata,* the action of the other micromycets were influenced a littler.

We recommend that the seeds destined for seed production, which do not use in the harvesting year, could use in the next years, but it is necessary to keep the seeds in the controlled atmosphere storage's and the seed germination to be over 90 % (Placinta D., 2005).

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