POSTHARVEST PATHOLOGY OF ORGANIC APPLES FROM ROMANIA. PRELIMINARY STUDY

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Abstract

Apple is one of the most important fruit species in the Northern hemisphere. As apple production is seasonal, the disparities between production and consumption may be balanced by storing the fruits with or without controlled atmosphere. By this, locally produced apples are kept fresh, in good condition, until the next production season. Pathogens may cause considerable losses during storage, quantitative and qualitative, both by degrading the appearance and fruits taste and by producing mycotoxins, a major food safety issue that becomes increasingly important for the consumer. Qualitative depreciation is a normal process during storage, but the level of degradation depends on storage conditions. Cold stored fruits (1°C, 90% humidity) produced in two Romanian organic orchards were analyzed in 2019, four months after harvest. No postharvest treatment was applied. Our results showed that postharvest diseases were predominantly caused by fungal pathogens and that both latent infections originating from the field (Gloeosporium sp.) and wounds infections (Penicillium sp., Fusarium sp.) were damaging the fruits

Key words: Fungal pathogens, organic production, post-harvest, Romania, apple.

INTRODUCTION

Long term storage of apples faces challenges in maintaining fruit quality and reducing losses from postharvest diseases (Mari et al., 2003). Qualitative depreciation is a normal process during storage, but the level of degradation depends on storage conditions and the storage technologies used (Hulea et al., 1982; Mari et al., 2003).

Currently, the apple industry relies mainly on synthetic fungicides to control postharvest decays. However, the limitations to fungicides such as the development of resistance in pathogens, difficulties in developing new fungicides, and their effect on the environment make this practice not sustainable. In addition, growing consumer demand for fungicide-free produce and a rapidly expanding organic market necessitate development of more sustainable alternatives to synthetic fungicides. Postharvest environmental conditions, in particular temperature, have a major impact on the visual, compositional, and eating quality of

fruit and vegetables. Temperature is, in fact, the component of the postharvest environment that has the greatest impact on the quality of fresh fruits and vegetable (Brasil et al., 2018; Willi et al., 2011; Cheng et al., 2013). One of the limiting factors that influence the fruits economic value is the relatively short shelf-life period caused by pathogens attacked. It is estimated that about 20-25% of the harvested fruits are decayed by pathogens during postharvest handling even in developed countries (Droby, 2006; Zhu, 2006). Fungal fruits infection may occur during the growing season, harvesting, handling, transport and post-harvest storage and marketing conditions, or after purchasing by the consumer. Fruits contain high levels of sugars and nutrients element and their low pH values make them particularly desirable to fungal decayed (Singh and Sharma, 2007).

The pathogens enter the fruit tissues in the early stages of growth and remain hidden there during maturation, while the symptoms will only be visible after harvesting and during storage (Passey et al., 2017; Louw and Korsten, 2014). Symptoms of disease can occur in phases different phenological during vegetation, but many pathogens have affecting fruits during storage can be collected from the field or already present in the storage area (Ammar and El-Naggar, 2014; Sever et al., 2012). These damages are probably the major cause for the loss of fresh products (Köhl et al., Consequently, fungal 2015). pathogens associated with postharvest rots of pears and apples can be separated into two main groups: "latent infection" (e.g., Neofabraea spp.) and pathogens (e.g., Botrytis "wound" Penicillium spp.) (Wenneker and Köhl, 2013).

MATERIALS AND METHODS

In order to make an overview of the pathogens present on the stored fruits in Romania, all accesible public databases and printed journals have been reviewed. In addition, as the Research center for studies of food and agricultural products quality from UASVM of Bucharest has modern storrage facilities, for both cold storage and controled athmosphere, the apples stored for different post-harvest research studies have been analysed. We have verified the fungal pathogens present on the fruits stored in the research center, apples that were produced in organic conditions, in the orchard of UASVM of Bucharest.

The following steps have been taken to achieve the proposed objective:

- Determination of the rotting levels;
- Macroscopic identification of pathogens;
- Confirmation of pathogen taxonomy after the fungal fructifications were formed.

The harvested fruit was stored, while recording the frequency and severity of the rot attack and calculating the degree of attack or rotting. Frequency (F%) is the relative value of the number of attacked fruits relative to the total number of fruits analyzed.

Severity (intensity) (I%) is the percentage of attack of the fruit. The attack degree (AD%) is calculated based on the frequency and severity of the attack (Balan et al., 2010).

Laboratory investigations aimed at identifying pathogens responsible for the occurrence of diseases during the storage period, the fruits were harvested manually in perfect condition and stored in controlled atmosphere rooms with the following storage conditions: 1 C, humidity 95 %. For phytopatological determinations of pathogenic load, fruits of all varieties studied were examined. The biological material was represented by fruits from different apple varieties: 'Rubinola', 'Topaz', 'Gemini', 'Renoir'.

Observations were made at 3, 9 and 12 days.

The experiments were carried out in the Plant Protection Diagnostic Laboratory of the Research center for studies of food and agricultural products quality. PDA culture medium (potato-dextrose agar) was used and incubation was done at 22° C thermostat. followed by identification with microscope. The preparation of the PDA culture medium in the pathogen development experiment was made following the existing prescriptions in the literature (Hulea et al., 1969). For the sowing of micromycetes, the technique provided in the literature was used (Ulea et al., 2011). Disposable Petri dishes with a diameter of 90 mm were used.

RESULTS AND DISCUSSIONS

Studying the spectrum of pathogens found on the harvested fruit, it was found that the microflora present in the analyzed samples consisted of genus fungi species *Gloeosporium* spp., *Penicillium* spp., *Fusarium* spp.

These results are in concordances with those obtained by Chira et al. (2014) that noted mainly *Gloeosporium album* developed better in low temperature conditions and high relative humidity, after 140 storage days.

Table.1 Pathogens isolated on the apple the during the storage period in 2018

Variety	The pathogen			
	Gloeosporium	Penicillium	Fusarium	
	spp.	spp.	spp.	
Rubinola	-	+	+	
Topaz	+	+	+	
Gemini	+	+	+	
Renoir	-	+	+	

The analysis of the apples harvested in 2018 shows that they have been shown to be fructifications of the micromycetes

Gloeosporium spp., Penicillium spp., and Fusarium spp.

The fungus *Penicillium* spp. has been detected on all 4 apple varieties, and the micromycetes *Gloeosporium* spp. has been found on the 'Topaz' and 'Gemini' varieties. The *Fusarium* spp. fructifications have been identified on all 4 apple varieties.





Figure 1. Rubinola

Figure 2. Gemini





Figure 3. Topaz

Figure 4. Renoir

Table 2. The microflora incidence detected on apples during the storage period in 2018

Variety	The pathogen			
	Gloeosporium	Penicillium	Fusarium	
	spp.	spp.	spp.	
	(after 9 days)	(after 9 days)	(after 9 days)	
Rubinola	0	73	27	
Topaz	55	32	13	
Gemini	14	68	18	
Renoir	0	34	66	

Observations on the incidence of micromycetes detected on apples in 2018 (Table 2) show that *Penicillium* spp. and *Fusarium* spp. are present on all apple varieties studied. The highest values of *Penicillium* spp. pathogen with high values for 'Rubinola' 73%, followed by 'Gemini' with F= 68%, 'Renoir' with an incidence of 34% and 'Topaz' with 32%.

Pathogens of the *Fusarium* spp. genus showed the highest incidence rates for 'Renoir' - 66%, 'Rubinola' - 27%, and 'Gemini' - 18%. The lowest incidence rate was noted for 'Topaz' variety at 13%.

Micromycetes of the genus *Gloeosporium* spp. were present on the 'Topaz' and 'Gemini' varieties. Frequency the fungus was 55% for the 'Topaz' variety, and 14% for the 'Gemini' variety.





Fig.5 The fungus development on the apple on PDA medium, after 10 days after inoculation

CONCLUSIONS

Observations on the incidence of micromycetes detected on apples in 2018 show that *Penicillium* spp. and *Fusarium* spp. are present on all apple varieties studied.

The fungus *Penicillium* spp. has been detected on all 4 apple varieties, and the micromycetes *Gloeosporium* spp. has been found on the 'Topaz' and 'Gemini' varieties. The *Fusarium* spp. fructifications have been identified on all apple varieties.

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REFERENCES

Ammar, M. I., El-Naggar, M. A (2014). Screening and characterization of fungi and their associated mycotoxins in some fruit crops. *International Journal of Advanced Research*, 2(4), 1216-1227.

Balan, V., Dobrin, I., Iacomi, B. (2010) Ghid fitopatologic. Bucharest:RO, Ed. Nivel Multimedia.
Brasil, I. M., & Siddiqui, M. W. (2018). Postharvest quality of fruits and vegetables: an overview. In M. W., Siddiqui (Ed.), Preharvest Modulation of Postharvest Fruit and Vegetable Quality (pp. 1-40). Academic Press.

Cheng, C., Gao, X., Feng, B., Sheen, J., Shan, L., and He, P. (2013). Plant immune response to pathogens differs with changing temperatures. *Nat. Commun.*, 4, 2530.

- Chira, L., Chira, A., Delian, E., Alexe, C., Marin, L. (2014), Research concerning the influence of different storage conditions on the preservation capacity of some new apple varieties. Scientific Papers. Series B, Horticulture, LVIII, 29-32.
- Crişan, A. (1973): Contribuţii la cunoaşterea ciupercilor care produc putregaiuri merelor depozitate. Studia Universitas Babeş – Bolyai, series Biologia, 1, 5-14.
- Droby, S. (2006). Improving quality and safety of fresh fruits and vegetables after harvest by the use of biocontrol agents and natural materials. *Acta Horticul.*, 709, 45–51.
- Hulea, A., Taşcă, Gh., Beratlief, C. (1982). Bolile şi dăunătorii produselor agricole şi hortiviticole după recoltare. Bucharest: RO, Ed. Ceres., 193-210.
- Hulea, A. (1969). Ghid pentru laboratoarele de micologie şi bacteriologie. Bucharest: RO, Ed. Agrosilvica.
- Köhl, J., Scheer, C., Holb, I. J., Masny, S., Molhoek, W. (2015). Toward an integrated use of biological control by *Cladosporium cladosporioides* H39 in apple scab (*Venturia inaequalis*) management. *Plant Disease*, 99(4), 535-543.
- Louw, J. P., Korsten, L. (2014). Pathogenic Penicillium spp. on apple and pear. *Plant Disease*, 98(5), 590-598
- Mari, M., Bertolini, P., Pratella, G. C. (2003). Nonconventional methods for the control of post-harvest pear diseases. *Journal of Applied Microbiology*, 94

- (5), 761-766.
- Passey, T.A.J., Robinson, J. D., Shaw, M. W., Xu, X. M. (2017). The relative importance of conidia and ascospores as primary inoculum of Venturia inaequalis in a southeast England orchard. *Plant Pathology*, 66(9), 1445-1451.
- Sever, Z., Ivic, D., Kos, T., Milicevic, T. (2012). Identification of Fusarium species isolated from stored apple fruit in Croatia. Archives of Industrial Hygiene and Toxicology, 63(4), 463-470.
- Singh, D., Sharma, RR. (2007). Postharvest diseases of fruit and vegetables and their management. In: Prasad, D. (Ed.), Sustainable Pest Management. New Delhi, India: Dava Publishing House.
- Willi, Y., Frank, A., Heinzelmann, R., Kälin, A., Spalinger, L., and Ceresini, P.C. (2011). The adaptive potential of a plant pathogenic fungus, Rhizoctonia solani AG-3, under heat and fungicide stress. *Genetica*, 139, 903.
- Wenneker, M., Köhl, J. (2013). Postharvest decay of apples and pears in the Netherlands. In II International Symposium on Discovery and Development of Innovative Strategies for Postharvest Disease Management 1053 (pp. 107-112)
- Ulea, E., Lipşa, F. D. (2011): Microbiologie. Iasi, RO: Editura Ion Ionescu de la Brad. ISBN 978-973-147-091-7.