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A Research on the Saprophytic Microfungi in Olive Fruit in Balikesir Province

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Abstract

The quality of two types of olive from shops in Balikesir province in respect to microfungal contamination was investigated. Altogether, 15 samples of olive were tested including black and green olive. Thirty microfungi isolates were obtained. The identification of the isolates show that there are 11 different species representing 3 genera and 18 different sterile microfungi were determined. *Penicillum* was stated as the richest taxa in terms of species numbers. We did not find any correlation between microfungi and type of olive.

Key words: *Olea europaeae*, saprophytic microfungi, isolation, identification.

INTRODUCTION

Olea is a genus of about 20 tropical and subtropical species of the Mediterranean region, Africa, southern and eastern Asia, Malesia, eastern Australia, and New Caledonia [1]. There are at least five natural subspecies distributed over a wide range: *Olea europaea* subsp. *europaea* (Europes), *Olea europaea* subsp. *cuspidata* (from Eritrea and Ethiopia south throughout East Africa, also in Iran to China), *Olea europaea* subsp. *guanchica* (Canaries), *Olea europaea* subsp. *maroccana* (Morocco) and *Olea europaea* subsp. *laperrinei* (Algeria, Sudan, Niger). [2]. Also, *Olea europaeae* is represented by 2 variety in the Flora of Turkey: *Olea europaeae* var. *europaea* Zhukovsky and *Olea europaeae* var. *sylvestris* (Miller) Lehr [3].

Considerable research has been accumulated supporting the health benefits of consuming olives, olive leaf and olive oil. The olive tree provides leaves, fruit and oil. Olive leaves are used in medicinal teas [3].

Olive constitutes a raw material source for various sectors in Turkey as it is the case in the world and contributes to economy with the processing of table olives and olive for oil production due to its natural characteristics [4]. In resent few decades, the amount of table olive and olive oil production in Turkey is given as follows [5].

Year	Table Olive (tone)	Oily olive (tone)	Olive Oil (tone)
2001	235 000	365000	65 000
2002	450 000	1 350 000	140 000
2003	350 000	500 000	79 000
2004	400 000	1 200 000	145 000
2005	400 000	800 000	115 000
2006	555 000	1 211 000	145 000

Olive fruit is major agricultural importance in the Mediterranean region as the source of olive oil and foods [4]. Olive is a high calorie food, it contains omega-3 and omega-6 fatty acids which are important for cardio vascular system. These fatty acids regulate the cholesterol level and reduce the risk of prostate cancer, bowel cancer, breast cancer and rheumatism arthritis. It has got vitamin E, A, D, K and it helps to bone upswing, loss of calcium keep from body. Olive contains cell regenerative substance which feeds on skin. Due to anti-oxidants, it presents body from noxious substance. It renews cells and reduces the aging of organs [4].

Mediterranean region have to 97% of world olive production and 87% of consumption [6]. Because of geographical location and ecological conditions Turkey is among the foremost olive and olive oil producers of the world [7]. Olive production is crowded in Aegean and Marmara regions, and is widespread in Balikesir, Aydın, Izmir, Mugla, Manisa and Canakkale provinces. According to the data of the recent years, 70% of the olive production of Turkey is suitable for using in oil production, and 30% can be used as table olives [8]. Turkey contributes 8% of world olive production and takes 2nd place after Spain in ranking of edible olive production [6].

Generally lost of studies were done about olive and olive oil were focused on phytochemicals in fruits and antioxidant and anti-inflammatory effects [2, 9]. Among these studies there is no any study on the olive inhabiting microfungi. As it is known, saprophytic microfungi has an important role in dividing dead organic materials into pieces.

MATERIALS AND METHODS

Fifteen olive examples consist of 7 black olive and 8 green olive samples used for mycological examination were obtained from local market and bazaar in 2007. These examples belong to Edremit, Akhisar and Gemlik regions.

For the qualitative study the direct inoculation method was used. These examples previously washed with 1% Benzalkonium chloride (alkyl dimethyl benzyl ammonium chloride) for surface sterilization which was diluted, and then each olive piece was cut off with a sterile thin pin. These pieces were inoculated to previously prepared peptone dextrose agar plates and then incubated in 25 ºC for 10 days [10]. In order to prevent the growth of bacteria, 30mg/l rose-bengal was added to the isolation medium [11]. This procedure is repeated form each of the 15 olive samples. The colonies grown up on petri dishes were examined under the stereomicroscope and transferred to a separate agar plate. Microfungi were identified according to micro- and macro- morphological characteristics were undertaken following the Smith's method [12]. The pure colonies of isolates were obtained in czapex dox and malt extract agar. The development of the colonies were regularly examined both macroscopically (developing degree of cultues, colour of colonies and changes in colour, colour of colony reverse, colour changes of medium, texture of colony surface, presence of odour, presence of exudates) and microscopically by using Olympus BX 51 (habit of hifa and its combination, development of fructification, colour, dimension and formation of fructification, details of structure and all details of spores) fort he final identifications. Identification of the isolates were performed using the literatures [13-23].

Citations of the authors' names presented are standardized according to the Kirk and Ansell, 1992 [24].

RESULTS AND DISCUSSION

The aim of this study was to determine the microfungi inhabiting on the olive examples. For this purpose, we examined 15 samples from different place. No microfungi could be isolated from three of these fifteen examples by using this applied isolation method under the experimental conditions but the rest thirteen examples provided 18 microfungi isolates. After the identification of the isolated 11 different species representing 3 genera were determined (Table 1). Among the samples highest microfungi density was observed in brine olive by 9 species and the most frequently encountered species was 8 isolated in *Penicillium clavigerum*. The amounts of colony forming units and so the differences among types of olive were too low to assess a significant effect of any type of olive. We did not find any effect of fermentation or origin of plant on the species composition.

It is not good enough as a food items for microbiological processes since olive and olive production contains various natural chemical compositions such as relatively low water content, having lipid substrate, lacking of oxygen...etc... However, in storage process of harvest and after harvesting it is not granted for becoming affected with mildew and production of tocsin. Therefore, in the whole production chains from field to table, modern production techniques have to be applied for preventing tocsin and microfungi improvements.

Table1. Information on collection of olive samples and isolated microfungi

Number of olive examples	Collection Place	Process Type	Substrate	Isolated microfungi
1	market	split	Black Olive	*Penicillium clavigerum* Demelius 1923
2	market	split	Black Olive	Microfungi was not isolated
3	market	split	Black Olive	*Penicillium janthinellum* Biourge 1923
4	market	split	Green Olive	*Penicillium italicum* Wehmer var. *italicum* Samson, Stolk&Hadlok 1976
5	market	split	Green Olive	*Penicillium lanosum* Westlig 1911
6	bazaar	break	Green Olive	*Aspergillus tubigensis* (Schöber) Mosseray 1934
7	bazaar	split	Green Olive	*Penicillium expansium* Link ex Gray 1821
8	market	split	Green Olive	*Penicillium brevi-compactum* Dierckx 1901 *Paecilomyces* sp.
9	market	Dry salted	Black Olive	*Aspergillus niger* van Tiegh. 1867 , *Penicillium clavigerum* Demelius 1923
10	bazaar	split	Green Olive	*Penicillium chermesinum* Biourge 1923, *Aspergillus tubingensis* (Schöber) Mosseray 1934
11	bazaar	break	Green Olive	*Penicillium clavigerum* Demelius 1923
12	bazaar	split	Black Olive	Microfungi was not isolated
13	bazar	Dry salted	Black Olive	*Penicillium clavigerum* Demelius 1923
14	bazaar	Dry salted	Black Olive	Microfungi was not isolated
15	bazaar	break	Green Olive	*Penicillium olsonii* Bainier&Sartory 1912

Microfungi play an important role in the degradation of organic debris [25]. Besides the economic loss associated with such storage pathogens, it has become clear, in recent years, that metabolic products of microfungi may represent significant health hazards. *Aspergillus niger* and three other species of the section *Nigri* now belong to ochratoxigenic fungi [26, 27]. *P. clavigerum* and *P. expansum* are ability to produce the mycotoxin patulin and cause spoilage of fruit products worldwide. The remaining patulin-producing species can be isolated from soil, are not commonly found in foods, although they could be acquired if fruits, for example, olives, are dropped on the ground and are used in processing [28]. Strains of the fungus *P. clavigerum* are known as producers of penitrem A, roquefortine A, and phenols [29] and also, *P. brevicompactum* produce brevianamide C, D, F [30], *P. olsonii* α-pyrones [31], *A. tubingensis* Ochratoxin A [32], *P. italicum* Deoxybrevianamide-E [33].

Secure and high quality olive and its productions have to be delivered without risking consumers healthy. In these days, increase of demanding of olive and its productions and importance of food processing especially not containing toxigenic microfungus in mans diet, secure food production is a highly crucial issue.

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