

1 **Biological Screening of Traditional Medicinal Plants from Villages of**  
2 **Akkuş (Ordu) in Turkey on the Effects of Tyrosinase**

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4  
5 **Running Title:** Biological Screening of the plants in Akkuş District

6  
7 **Abstract**

8 In the present study, ethnomedicinal uses of the plants naturally growing in 35  
9 villages of Akkuş District have been determined. Informations about the uses of the  
10 plants have been obtained from villagers using a questionnaire. At the end of the  
11 identification of the plants collected from villages, 58 taxa belong to 32 families have  
12 been determined. Furthermore, tyrosinase enzyme activity studies of methanolic  
13 extracts of all the species carried out spectroscopically. Eventually, the plants has  
14 been demonstrated to be used as folk medicine, food animal feed, ornamental, stuff,  
15 dyeing, odour, insecticide and fishing. According to the biological screening studies,  
16 some species have been indicated to have inhibitör effect, but the others have  
17 activator effect on tyrosinase enzyme.

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20 **Key Words:** Akkuş, Biological Screening, Ethnomedicinal, Folk Medicine, Tyrosinase  
21 Enzyme

22  
23 **1. INTRODUCTION**

24 Melanin is known to be one of the major pigments for skin and hair color of  
25 mammals. Melanin have been synthesized in differentiated cells such as  
26 melanocytes in the skin, retinal epithelium, and central nervous system in mammals  
27 (1-3).

28 Tyrosinase, being copper-containing in active site, is known to be a member of  
29 the polyphenol oxidase enzyme family, which is an key enzyme in charge of melanin

30 biosynthesis. The tyrosinase enzyme inhibitors from natural resources have been  
31 investigated by many researchers but there is not been enough studies on the  
32 tyrosinase enzyme activators (3).

33 Defect of melanin synthesis have been associated with pigmentation disorders.  
34 Decreasing of melanin level have been caused various skin diseases such as  
35 psoriasis, vitiligo, because of hypopigmentation (4). Using the agents having  
36 tyrosinase activator effect is an efficient approach for treatment of hypopigmentation  
37 disorders. Hyperpigmentation knowned increasing production of melanin have been  
38 induced some disorders like actinic damage, melasma, freckle and age-related stains  
39 The tyrosinase inhibitors can be used as a remedy for these diseases (5).

40 Also, Tyrosinase inhibitors have been believed to be used to cure ailments  
41 related to neurotoxicity like Parkinson. Tyrosinase have been give rise to  
42 accumulating of oxide-dopamine derivatizations due to neuronal damage (6).

43 Ethnobotany is described as “the use of plants by the local population” by John  
44 W. Harsberger and ethnobotanical studies are important to determine the relationship  
45 between ethnobotany and plants (7). In Turkey, the interest in ethnobotanical studies  
46 beginning in the early 19<sup>th</sup> century has been observed to be higher in recent years  
47 (8).

48 Turkey, located at the junction of three different phytogeographic regions,  
49 namely Mediterranean, Irano-Turanian, Euro-Siberian and has a rich flora (8). Turkey  
50 is a leading country in terms of medicinal and endemic plants in the World (Lamond,  
51 1978). It is known that about 10000 flowering and fern plant species has naturally  
52 grown in Turkey, and 30% of them are endemic (10-12).

53 Besides, our country which hosted many civilizations has a rich cultural heritage  
54 and a wealth of ethnobotanical datas (13). The traditional knowledge on plants and  
55 their uses has been disappearing in recent years because of urbanization, increased  
56 migration to urban areas, developments in technology, and health services easier  
57 accessibility (14-16) Therefore, any information about uses of the plant in our daily  
58 life should be recorded as soon as possible.

59 Ordu province has been located (40°18'-41°08' N, 36°52'-38°12' E) in Euro-  
60 Siberian phytogeographic region (17). However There are the plenty of studies on  
61 the folk medicine in the Black Sea region, some of which is ethnobotanical research  
62 about Ordu, any study in Akkus district has not been found (9, 14, 18-26). Also, the  
63 effects of tyrosinase enzyme of the plants naturally growing in 35 villages of Akkuş  
64 District and used as folk medicine have been explored for the first time in this study.

## 65 **2. MATERIAL and METHODS**

### 66 **2.1. Field Trips**

67 The materials of our research includes plant samples collected from 35 villages  
68 of Akkus District of Ordu province in Turkey. Plant samples were obtained by visiting  
69 the research area twice between July and September 2016. A questionnaire form  
70 (name, surname, age, telephone numbers, local names of plants, parts used,  
71 preparation methods and the purpose of using plants) was prepared for the  
72 participants. The participants were visited at their homes and asked to them to fill out  
73 the questionnaire form. After each interview, plant samples were collected with  
74 assistance from the participants.

### 75 **2.2. Study Area**

76 Akkus is located in the inner parts of the Middle Black Sea in Turkey. The height  
77 of the sea is 1340 meters and has a rough land (Fig 1). The Akkus region includes 6  
78 towns, 35 villages and 15 neighborhoods. Continental climate prevails in the region  
79 (27).

### 80 **2.3. Identification of Plants**

81 The plant samples collected from the research area were identified using  
82 references, i.e. Flora of Turkey and the East Aegean Islands (11, 28, 29).  
83 Identifications were performed by two of the authors (MB and UO). The dried plant  
84 samples are deposited at the Herbarium of the Faculty of Pharmacy of Ankara  
85 University.

86

## 87 **2.4. Studies on the Tyrosinase Enzyme**

### 88 **2.4.1. Chemical Materials Used in Experiments**

89 Methanol (CH<sub>3</sub>OH-Merck, 106009), Tyrosinase (Sigma, T3824-25KU), kojic  
90 acid (Sigma, K3125-5G), potassium dihydrogen phosphate (NaH<sub>2</sub>PO<sub>4</sub>-Sigma Aldrich,  
91 7558-80-7), disodium hydrogen phosphate anhydride (Na<sub>2</sub>HPO<sub>4</sub>-Sigma Aldrich,  
92 10039-32-4), L-DOPA (Sigma, D9628-5G) and 8-Methoxsalen (Roth, 5497.2) were  
93 used in the experiments.

### 94 **2.4.2. Preparation of the Extract**

95 Methanol extract (10 mg/mL) of each species from Akkus was prepared. Diluted  
96 samples were obtained from methanol extract with potassium phosphate buffer (pH  
97 6.8) at concentrations of 25, 50, 100 and 500 µg / mL.

### 98 **2.4.3. Tyrosinase Enzyme Inhibition**

99 Tyrosinase inhibitory activity was determined employing various concentrations  
100 of kojic acid solutions as standard (30). Tyrosinase solution (46 U/mL), methonolic  
101 plant extract (500, 100, 50 and 25 µg/mL) were prepared. 120 µL of 0.2 M phosphate  
102 buffer (pH 7.0), and 40 µL tyrosinase solution for A wells; 160 µL of 0.2 M phosphate  
103 buffer (pH 6.8) for B wells; 80 µL of 0.2 M phosphate buffer (pH 6.8), 40 µL  
104 tyrosinase solution and 40 µL sample solution for C wells; 120 µL of 0.2 M phosphate  
105 buffer (pH 7.0) and 40 µL sample solution for D wells were added and mixed in a 96-  
106 well plate and incubated for 10 min at 23 °C. Then, 2,5 mM L-DOPA solution (40 µL)  
107 was added to all wells and incubated for 10 min at 23 °C. The absorbance of the  
108 reaction mixture was determined at 490 nm using the spectrophotometric method in  
109 a microplate reader. The percentage of tyrosinase inhibitory activity was calculated  
110 using the formula follows:

$$111 \quad \% \text{ Inhibition} = \frac{[(A-B)-(C-D)]}{(A-B)} \times 100$$

112 The results were given as IC<sub>50</sub> levels.

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#### 115 2.4.4. Tyrosinase Enzyme Activation

116 Tyrosinase Enzyme Activation was determined employing various  
117 concentrations of 8-Methoxsalen (8-MOP) solutions as standard (31). Tyrosinase  
118 solution (46 U/mL), methonolic plant extract (500, 100, 50 and 25 µg/mL) were  
119 prepared. 120 µL of 0.2 M phosphate buffer (pH 7.0), and 40 µL tyrosinase solution  
120 for A wells; 160 µL of 0.2 M phosphate buffer (pH 6.8) for B wells; 80 µL of 0.2 M  
121 phosphate buffer (pH 6.8), 40 µL tyrosinase solution and 40 µL sample solution for C  
122 wells; 120 µL of 0.2 M phosphate buffer (pH 7.0) and 40 µL sample solution for D  
123 wells were added and mixed in a 96-well plate and incubated for 10 min at 23 °C.  
124 Then, 2.5 mM L-DOPA solution (40 µL) was added to all wells and incubated for 10  
125 min at 23 °C. The absorbance of the reaction mixture was determined at 490 nm  
126 using the spectrophotometric method in a microplate reader. The percentage of  
127 tyrosinase enzyme activation was calculated using the formula follows:

$$128 \quad \% \text{ Activation} = \frac{[(A-B)-(C-D)]}{(A-B)} \times 100$$

129 The results were given as AC<sub>50</sub> levels.

### 130 3. RESULTS AND DISCUSSION

#### 131 3.1. The Results of Ethnobotanical Studies

132 The plants naturally growing in 35 villages of Akkuş District has been showed to  
133 be used as folk medicine (30 taxa), food (21 taxa), animal feed (8 taxa), ornamental  
134 (5 taxa), stuff (5 taxa), dyeing (4 taxa), odour (1 taxa), insecticide (1 taxa), and  
135 fishing (4 taxa). The informations from the ethnobotanical studies have been included  
136 in Table 1.

137 **Table 1. Species used in Akkuş villages**

Family, Plant Species, Herbarium Nunmer	Local name	Plant Part(S) Used	Uses	Administration Ways
<b>Asteraceae</b>				
<i>Anthemis cotula</i> (AEF 26978)	Papatya	Aerial Parts	Asthma, Cold, and Alopecia	Boiled in water and used up as hot drink, also used as vapour
<b>A.</b> <i>tinctoria</i> var. <i>tinctoria</i> (AEF 26972)	Sarı papatya	Aerial Parts	Dyeing Food	Dyes wools to yellow for wool Consumed as tea
<i>Bellis perennis</i> (AEF 26979)	Küçük papatya	Aerial Parts	Stomach ache Sinusitis	Boiled in water and drunk Used as vapour

<i>Cirsium arvense</i> (AEF 26981)	Köygöçüren		Food	Consumed as meal
<i>Tanacetum parthenium</i> (AEF 26977)	Papatya	Aerial Parts	Alopecia	Boiled in water and drunk
<i>Tussilago farfara</i> (AEF 26884)	Öksürük otu	Flowers Leaf	Breathless Bronchitis	Boiled and used as vapour
<i>Helichrysum compactum</i> (AEF 26823)	Yayla çiçeği	Herba	Earache  Ornamental	Boiled and the water is dropped to ear Used as odour
<b>Amaranthaceae</b>				
<i>Amaranthus albus</i> (AEF 26904)	Hoşkiran, hoşberin	Herba	Food	Consumed as meal Roasted and mixed to yoghurt or consumed as soup
<b>Boraginaceae</b>				
<i>Trachystemon orientalis</i> (AEF 26966)	Kaldırık	Leaf	Food	Consumed as meal and pickle
<b>Caprifoliaceae</b>				
<i>Sambucus ebulus</i> (AEF 26890)	Yivdin, mürver	Fruit  Leaf And Fruit	Dyeing  Food Rheumatic diseases	Boiled and used to dye Eaten as fresh leaf Cooked or crushed and then applied to aching area
<b>Caryophyllaceae</b>				
<i>Dianthus carmelitarum</i> (AEF 26822)			Ornamental	Used as Ornamental plant
<i>Silene compacta</i> (AEF 26888)			Ornamental	Used as ornamental plant
<i>Silene vulgaris</i> var. <i>vulgaris</i> (AEF 26891)	Gıcırık otu, gırşılık, düdüklük otu	Leaf  Herba	food  Stuff	Consumed as meal, Roasted and mixed to yoghurt Used to make caval
<b>Chenopodiaceae</b>				
<i>Chenopodium album</i> (AEF 26902)	Küllüce, sirken	Herba	Food	Roasted and eaten
<b>Convolvulaceae</b>				
<i>Convolvulus arvensis</i> (AEF 26817)	Sarmaşık	Herba	Farming	Used as animal feed
<b>Cruciferae</b>				
<i>Capsella bursa-pastoris</i> (AEF 26895)	Kuş pancarı	Herba	Food	Roasted and eaten
<b>Cornaceae</b>				
<i>Cornus mas</i> (AEF 26897)	Kızılıcık	Fruit	Food	Boiled in water and drunk, Consumed as marmalade
<b>Euphorbiaceae</b>				
<i>Euphorbia</i> sp. (AEF 26903)	Sütotu, akkapla	Latex  Herba	Alopecia  Farming	Latex is applied to alopesic area Used as animal feed
<b>Ericaceae</b>				
<i>Vaccinium arctostaphylos</i> (AEF 26969)	Yaban gülü		Stuff	Used to make whisk
<b>Fabaceae</b>				
<i>Coronilla cretica</i> (AEF 26982)	Fiğ otu	Herba	Farming	Used as animal feed
<i>Coronilla varia</i> (AEF 26886)	Yabani korunga	Herba	Farming	Used as animal feed
<i>Lathyrus aureus</i> (AEF26819)	Karanfil		Mouth sore	

<i>Lathyrus sp.</i> (AEF 26974)	Eşek palası	Leaf	Food	Consumed as meal
<i>Vicia sativa</i> (AEF 26973)	Yabancı fiğ	Herba	Farming	Used as animal feed
<b>Fagaceae</b>				
<i>Fagus orientalis</i> (AEF 266892)	Kayın	Seed	Anxiolytic	
<i>Quercus cerris var. cerris</i> (AEF 26879)	Meşe	Shoot	Stuff	Used to make hamper
<i>Quercus infectoria</i> (AEF 26881)	Meşe	Shoot	Stuff	Used to make hamper
<b>Hypericaceae</b>				
<i>Hypericum perforatum</i> (AEF 26820)	Kantaron, bitki otu	Herba	Sedative Antitussive	Drunk as tea Drunk as tea freshly
<b>Hypolepidaceae</b>				
<i>Pteridium aquilinum</i> (AEF 26975)	Kızılot, mayasıl, eğrelti	Herba	Antihemorrhoid	Boiled and wrapped up waist, also sit on vapour
<b>Juglandaceae</b>				
<i>Juglans regia</i> (AEF 26880)	Ceviz	Fruit	Dyeing	Used to dye rugs
<b>Lamiaceae</b>				
<i>Mentha longifolia</i> (AEF 26887)	Narpus	Herba	Cold	Consumed as tea
<i>Mentha sp.</i> (AEF 26951)	Nane, narpus	Leaf	Food Dyspepsia	Consumed as spice Consumed as tea
<i>Origanum vulgare</i> (AEF 26907)	Kekik, kaba topağı	Fruit Herba	Food Dyspepsia Food	Consumed as spice Consumed as tea
<i>Prunella orientalis</i> (AEF 26824)	Çay otu, karabaş otu	Herba	Cold Antitussive	Consumed as tea
<i>Salvia verticillata var. verticillata</i> (AEF 26900)	Karacaabla	Herba Flower	Farming Dyeing	Used as animal feed Used to dye purple
<i>Stachys byzantina</i> (AEF 26883)		Leaf	Stuff	Used as hanky
<b>Loranthaceae</b>				
<i>Viscum album var. album</i> (AEF 26906)	Çam gökçesi, gökçe otu	Leaf	Treatment of renal disorders Farming	Boiled and eaten Used as animal feed
<b>Malvaceae</b>				
<i>Malva neglecta</i> (AEF 26950)	Kömeç, ebegümeci	Herba	Anti-inflamatuar anticanser Urinary tract infection Laxative Analgesic	Roasted and eaten, also consumed as tea
<b>Papaveraceae</b>				
<i>Papaver rhoeas</i> (AEF 26967)	Gelincik		Ornamental	Used as ornamental plant
<b>Pinaceae</b>				
<i>Pinus sylvestris</i> (AEF 26901)	Çam	Shoot Fruit  Resin	Asthma, breathless, bronschitis, Food  Wound healing Stomache ache	Shoots are eaten after peeled off Fruits kept with sugar for a week and boiled in water, consumed as jam Applied to wound Consumed by chewing

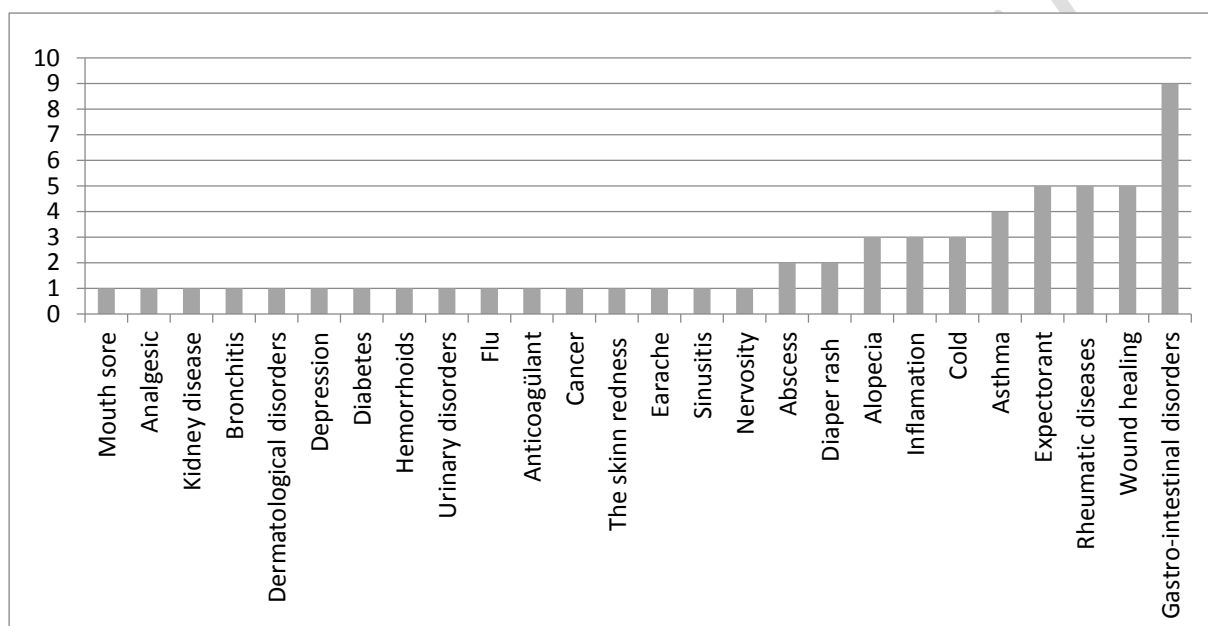
<b>Plantaginaceae</b>				
<i>Plantago major</i> var. <i>major</i> (AEF 26885)	Sinir otu, siğil otu, kesik otu, sinirli yaprak, yara otu	Leaf	Antihemorrhagic  Antitussive Antidiabetic Antirheumatic  Anti-inflamatuar	Dried, powdered and applied to wound and incision by wrapping Consumed as tea  Haeted slightly and wrapped Boiled in water and consumed as tea
<b>Platanaceae</b>				
<i>Platanus orientalis</i> (AEF 26894)	Çınar	Leaf	antirheumatic	Consumed as tea
<b>Polygonaceae</b>				
<i>Polygonum convolvulus</i> (AEF 26976)	Perzi	Leaf	Food	Consumed as meal
<i>Polygonum persicaria</i> (AEF 26984)	Biber otu	Leaf Herba	Fishing Farming	Used to fishing Used as animal feed
<i>Rumex acetosella</i> (AEF 26968)	Acumuk, kuzukulağı	Leaf Herba	Food Insecticide	Eaten freshly Used for fleas
<i>Rumex patientia</i> (AEF 26971)	Efelik	Leaf  Stem	Analgesic for aching of knee, to cure itching, and skin disorders  Food Food	Boiled and wrapped on knee, Kaynatılarak dize sarılır, treated for itching because of nettle Consumed as meal Eaten after peeling off
<b>Primulaceae</b>				
<i>Lysimachia verticillaris</i> (AEF 26821)			Ornamental	Used as ornamental plant
<b>Rhamnaceae</b>				
<i>Frangula alnus</i> (AEF 26965)	Çeti ağacı	Shoot	To treatment rash, wound, and burn Antiinflammatory	Consumed as tea, applied to wound, infected area
<b>Rosaceae</b>				
<i>Agrimonia eupatoria</i> (AEF 26905)	Mikbaşı	Leaf	Dyspepsia	Roasted and eaten
<i>Crataegus stevenii</i> (AEF 26897)	Kuş dikenı	Fruit	Food	Consumed as jam
<i>Mespilus germanica</i> (AEF 26889)	Töngel, muşmula	Stem Shoot And Stem	Dyspesia and cold  Antitussive	Boiled and consumed as tea Boiled and consumed as tea
<i>Pyrus sp.</i> (AEF 26899)	Armut ağacı	Herba	Stuff	Used to make spoon
<i>Rubus ideaus</i> (AEF 26983)	Böğürtlen	Stem  Fruit	To treatment stomachaches and antiinflammatory Food	Consumed as jam
<b>Scrophulariaceae</b>				
<i>Verbascum pyramidatum</i> (AEF 26980)	Siğır kuyruğu, küçük kabalak	Herba	Food	Eaten after peelinf off
<b>Tiliaceae</b>				
<i>Tilia platyphillos</i> (AEF 26896)	İhlamur	Flowers	Antitussive and to treatment stomachache	Consumed as tea
<b>Umbelliferae</b>				
<i>Chaerophyllum byzantinum</i> (AEF 26970)	Baldıran	Herba	Food	Boiled and the water is drunk



<b>Urticaceae</b>				
<i>Urtica dioica</i> (AEF 26882)	Sırgan otu, ısrırgan	Herba	Food To treatment knee ache	Consumed as meal and soup Heated and wrapped up aching area
<b>Vitaceae</b>				
<i>Vitis vinifera</i> (AEF 26893)	Asma yapağı, üzüm yapağı	Leaf	Food	Consumed as meal

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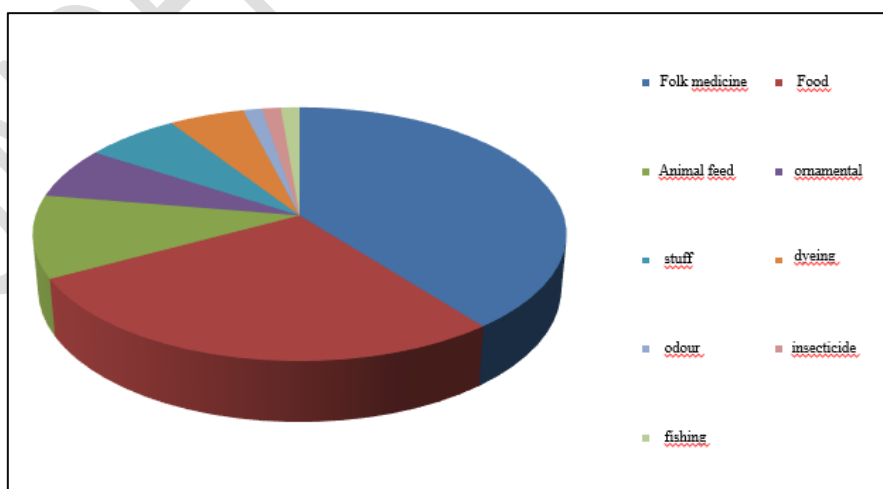
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143 **Fig 1. Ethnomedicinal usages of the plants in Akkuş**

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145

146 **Fig 2. Classification of plants collected from Akkuş according to their usage**

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### 148 3.2. The Results of Biological Screening Studies

149 According to the biological screening studies, some species collected Akkuş  
150 district have been evidenced to have inhibitör effect, and that all the species don't  
151 have activator effect on tyrosinase enzyme. The results of the studies have been  
152 sum up in Table 2. In the tyrosinase enzyme inhibition and activation studies, the  
153 results of the methanol extracts of the species have been compared with kojic acid  
154 for tyrosinase inhibition and 8-MOP for tyrosinase activation used as positive  
155 control. Among the species, *Chaerophyllum byzantinum* have been showed the  
156 highest tyrosinase inhibitor effect.

157 **Table 2. The effects on tyrosinase enzyme of species used in Akkuş villages**

Tyrosinase Inhibitor Effective species	
IC <sub>50</sub> (µg/mL)	
<i>Chaerophyllum byzantinum</i>	25.60
<i>Vaccinium arctostaphylos</i>	103.75
<i>Trachystemon orientalis</i>	133.98
<i>Hypericum perforatum</i>	148.70
<i>Rumex acetosella</i>	270.62
<i>Capsella bursa-pastoris</i>	425.37
$\alpha$ -Kojic acid	3.48
Tyrosinase Activator Effective Species	
AC <sub>50</sub> (µg/mL)	
8-MOP	17.16

158 There are the plenty of studies on the folk medicine in the Black Sea region,  
159 some of which is ethnobotanical research about Ordu (18-26), but any study in Akkus  
160 district has not been found. Also, the effects of tyrosinase enzyme of the plants  
161 naturally growing in 35 villages of Akkuş District and used as folk medicine have  
162 been explored for the first time in this study.

163 200 plants have been collected from Akkuş and its villages for the present  
164 study. At the end of the identification of the plants collected from villages, 58 taxa  
165 belong to 32 families have been determined [Asteraceae (7 taxa), Amaranthaceae (1  
166 taxa), Boraginaceae (1 taxa), Caprifoliaceae (1 taxa), Caryophyllaceae (3),  
167 Chenopodiaceae (1), Convolvulaceae (1), Cruciferae (1 taxa), Cornaceae (1 taxa),  
168 Euphorbiaceae (1 taxa), Ericaceae (1 taxa), Fabaceae (5 taxa), Fagaceae (3 taxa),  
169 Hypericaceae (1 taxa), Hypolepidaceae (1 taxa), Juglandaceae (1 taxa), Lamiaceae  
170 (6 taxa), Loranthaceae (1 taxa), Malvaceae (1 taxa), Papaveraceae (1 taxa),

171 Pinaceae (1 taxa), Plantaginaceae (1 taxa), Platanaceae (1 taxa), Polygonaceae (4  
172 taxa), Primulaceae (1 taxa), Rhamnaceae (1 taxa), Rosaceae (5 taxa),  
173 Scrophulariaceae (1 taxa), Tiliceae (1 taxa), Umbelliferae (1 taxa), Urticaceae (1  
174 taxa), Vitaceae (1 taxa)].

175 The most popular species in the district for medical uses are *Plantago major*  
176 var. *major*, *Pinus sylvestris*, *Malva neglecta*. The species from Akkuş have been  
177 used mostly for treatment of cough, rheumatic diseases and wounds, traditionally.

178 *C. cretica*, *V. pyramidatum*, *P. convolvulus*, *C. album*, *T. farfara*, *A. albus*, *S.*  
179 *verticillata* var. *verticillata*, *P. major* var. *major*, *P. orientalis*, *P. persicaria*, *M.*  
180 *longifolia*, *S. byzantina*, *A. eupatoria*, *O. vulgare*, *D. carmelitarum*, *F. Alnus*, *C. varia*,  
181 *P. orientalis*, *Q. infectoria*, *U. dioica*, *H. compactum*, *M. neglecta*, *T. parthenium*, *S.*  
182 *ebulus*, *R. ideaus*, *V. album* var. *album*, *B. perennis*, *A. tinctoria* var. *tinctoria*, and *A.*  
183 *cotula* from the collected species have been showed to have tyrosinase inhibitory  
184 effect; but the results were not significant. Because the IC<sub>50</sub> values of them haven't  
185 been our detection range.

186 As shown in Table 2; *T. orientalis*, *C. bursa-pastoris*, *V. arctostaphylos*, *R.*  
187 *acetosella*, *C. byzantinum*, and *H. perforatum* have been caused to inhibition on  
188 tyrosinase enzyme. In view of values of IC<sub>50</sub> of the methanolic extracts of the plants,  
189 *C. byzantinum* have been indicated to have the best effect on the enzyme. Besides,  
190 *P. aquilinum*, *C. arvensis*, *S. compacta*, *F. orientalis*, *T. platyphyllos*, *Mentha* sp., *S.*  
191 *vulgaris* var. *vulgaris*, *C. arvensis*, *P. rhoeas*, *R. patientia*, *P. Sylvestris* and *C. mas*  
192 have been detected to have tyrosinase activator effect; however the values of IC<sub>50</sub> of  
193 them were greater than 1000 µg/mL, so the results were not significant.

#### 194 **4. CONCLUSION**

195 Compared with previous studies carried in neighborhood in terms of the folk  
196 medicine, the similar datas have been obtained in terms of ethnomedicinal studies.  
197 Through the study, it has been determined the cultural interactions between the  
198 human and plant in Akkuş district by identifying the plants used by villagers, the  
199 ethnomedicinal properties, their usages and local names for the first time. In addition,  
200 the effects of tyrosinase enzyme of collected plant have been carried out a

201 preliminary study in terms of potential usages of the species for skin disorders and  
202 the neurodegenerative damages.

203 The present study is important for being transferred of the ethnobotanical and  
204 ethnomedicinal features of the area to next generation. The obtained results will shed  
205 light on future research based on this pilot.

206

### 207 **Conflict of Interest**

208 The authors declare that there are no conflicts of interest

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