



Antibacterial and photodynamic effects of some plant extracts for cavity disinfection

Serap Akyuz^a, Ozlem Moufti Chousein (Ntemir)^a, Ozlem Sacan^b, Refiye Yanardag^b, Sadık Kalaycı^c, Aysen Yarat^{d,*}, Fikrettin Sahin^c

^a Marmara University, Faculty of Dentistry, Department of Clinical Sciences, Istanbul, Turkey

^b Istanbul University, Faculty of Engineering, Department of Chemistry, Istanbul, Turkey

^c Yeditepe University Faculty of Engineering, Department of Genetics and Bioengineering, Istanbul, Turkey

^d Marmara University, Faculty of Dentistry, Department of Basic Medical Sciences, Biochemistry, Istanbul, Turkey

ARTICLE INFO

Keywords:

Plant extracts
Antibacterial
Photo-active
Cavity disinfection
Dentistry
Antimicrobial photodynamic therapy

ABSTRACT

Background: In current dental treatments, with the aim of a preventive approach, it is argued that removing only the infected layer of dentin is sufficient for cavity preparation. However it is impossible to be sure that the infected layer was completely removed. In addition, the cause of secondary caries and post operative sensitivities has been reported as residual bacteria in some studies. The aim of this study is to investigate the antibacterial and photo-active properties of *Cotinus coggygia* Scop., *Rumex crispatus* DC., *Beta vulgaris* L. var. *cicla* and *Eruca sativa* aqueous extracts, and to investigate their usefulness for cavity disinfection in dentistry.

Method: The aqueous solutions of plant extracts were prepared to be at a maximum concentration and the *Streptococcus mutans* solutions mixed with phosphate buffered saline to give 10⁸ cfu/mL. A 430–480 nm wavelength light source was used for the irradiation. Three different applications were made: extract + *Streptococcus mutans* mixture exposed to light; extract + *Streptococcus mutans* mixture that was not exposed to light and *S. mutans* exposed to light.

Results: No antibacterial effect was found for the second and third applications. In the first application, however, irradiation with extract + *Streptococcus mutans* mixture reduced the number of microorganisms in the beginning by 99% for only *Rumex crispatus* DC. extract (log 2).

Conclusion: *Rumex crispatus* DC. extract can be used as an alternative in photo-active disinfection of cavities in dentistry.

1. Introduction

The concept of minimal intervention dentistry has been accepted at the present time with the increased understanding of the caries process and the development of adhesive restorative materials [1]. In this concept, only superficial necrotic and demineralized dentin removal is recommended. Because it is now known that dentin, which was decalcified but has sound collagen fibers preserved, can be remineralized [2,3]. It is very important that the residual bacteria in the dentin tubules are eliminated [2,3]. This is critical for pulp vitality especially in young patients. For this purpose, it is recommended to use some cavity disinfectants before restoring the cavity [4]. Chlorhexidine digluconate is the most popular agent for cavity disinfection, there are also many other alternative agents. Unfortunately, most antibacterial agents used

for dentin disinfection can damage cells and growth factors [4–6].

Photo-active disinfection (PAD) has important advantages such as it is a localized, non-invasive treatment and does not develop microbial resistance [5,6]. The therapy involves an interaction between a photosensitizer (PS) agent and a light source, they producing reactive oxygen species (ROS) like singlet oxygen. ROS kill bacteria by damaging bacterial cell wall, membrane proteins and nucleic acids [7]. With PAD, different PSs and light sources were studied and used. Over time, increasing popularities of natural remedies encourage researchers to find natural alternatives. Most of plant extracts have been searched for this purpose [8].

Cotinus coggygia Scop. (Anacardiaceae) is a plant which is used as antiseptic, anti-inflammatory, antiulcer, antimicrobial, antifungal, antiviral, anticancer, antipyretic, antioxidant, antihemorrhagic,

* Corresponding author at: Marmara University, Faculty of Dentistry, Basic Medical Sciences, Biochemistry, Basibuyuk Yolu 9/3 34854 Basibuyuk, Maltepe, Istanbul, Turkey.

E-mail addresses: ayarat@marmara.edu.tr, sakyuz@marmara.edu.tr (A. Yarat).

<https://doi.org/10.1016/j.pdpdt.2019.02.025>

Received 7 November 2018; Received in revised form 31 January 2019; Accepted 25 February 2019

Available online 26 February 2019

1572-1000/ © 2019 Elsevier B.V. All rights reserved.

antigenotoxic and wound healing paradontosis in folk medicine [9,10] has a large amount of flavonoids, phenols, antocyanins, terpenes, gallic acid, essential oil [11]. Yarat et al., 2013, have evaluated the *in vitro* effectiveness of *Cotinus coggygia* aqueous extract on tissue factor activity in saliva samples [12]. The leaves of *Rumex* species (Polygonaceae) plants are used in traditional medicine for the treatment of several health disorders such as infections, diarrhoea, diabetes, jaundice oedema, hypertension, diuretic, analgesic and inflammation [13]. It has various pharmacological activities such as antitumour, antibacterial, antiviral, antifungal, antioxidant, antiparasitic, antineoplastic and antiinflammatory activities [13]. Anthraquinones, flavonoids, stilbenoids, naphthalenes, tannins, triterpenoids, carotenoids, polysaccharides have been reported in the *Rumex* species [13]. Several studies have demonstrated that *Beta vulgaris* L.var.*cicla* (Chenopodiaceae) has antioxidant [14], antidiabetic [15], anticancer [16], antimicrobial [17], antiproliferative [18], antiacetylcholinesterase and hepatoprotective effects [19]. Phytochemical screenings of this plant have revealed the presence of some saponins, flavanoids, glycolipids, polyphenols, polysaccharides, folic acid, ascorbic acid, pectin betalains and phenolic amides [14]. *Eruca sativa* Mill (Brassicaceae) is used as diuretic in Turkish traditional medicine [20] and is good for alleviating coughs, giving strength, appetizing and stimulant [21]. *Eruca sativa* is considered an excellent source of antioxidants as it includes phenolic compounds, carotenoids, glucosinolate and isothiocyanates [22]. This plant has antioxidant, anticancer activity, antiplatelet, antithrombotic, antiseptic, antiinflammatory, cytoprotective and antiulcer activities [23,24].

The aim of this study is to investigate the antibacterial and photo-active properties of *Cotinus coggygia* Scop., *Rumex cristatus* DC., *Beta vulgaris* L.var.*cicla* and *Eruca sativa* aqueous extracts, and to investigate their usefulness for cavity disinfection in dentistry.

2. Material and methods

This *in vitro* study was approved by the Ethics Committee of Marmara University Health of Science with protocol no: 30.05.2016-38.

2.1. Preparation of aqueous plant extracts

Plant extracts (*Cotinus coggygia* Scop., *Rumex cristatus* DC., *Beta vulgaris* L.var.*cicla* and *Eruca sativa*) were used like a PS. Extracts were prepared at Department of Chemistry, Faculty of Engineering, Istanbul University. *Beta vulgaris* L.var.*cicla* were identified by Prof.Dr.Neriman Ozhatay and *Cotinus coggygia* Scop., *Rumex cristatus* DC., and *Eruca sativa* were identified by Prof.Dr.Kerim Alpınar Faculty of Pharmacy, Istanbul University. Plants materials were washed with water and dried at room temperature. The dried plants were stored in -20°C until used. Dried leaves (50 g) were extracted by adding 500 mL of distilled water and boiling for 30 min. The extracts were then filtered and lyophilized. Then were kept at -20°C . Extracts, used as a PS agents, was dissolved in distilled water at maximum concentration. They were 0.35 g/mL, 0.23 g/mL, 0.26 g/mL, 0.25 g/mL and 0.41 g/mL for *Cotinus coggygia* Scop., *Rumex cristatus* DC., *Beta vulgaris* L.var.*cicla* and *Eruca sativa* respectively. Solutions were stored in dark at $+4^{\circ}\text{C}$ until the experimental day.

2.2. Determination of maximum absorbances of plant extracts

Before antimicrobial study each plant extract's absorption spectrum was measured by a spectrophotometer and the light emission pattern of the light was also evaluated and the wave length to activate the PS was determined. Optical absorption spectra of all plant extracts were registered on a double-beam UV/VIS spectrophotometer Lambda 35 (Perkin Elmer, Germany) by scanning in the range of 300–900 nm at room temperature.

2.3. Preparation of microbial solution

S. mutans isolated from dental carious lesions (Yeditepe University) were cultured in Colombia blood agar and incubated at 37°C . After 48 h, microbial growth in each medium was determined by reading absorbance at 600 nm for bacteria isolates using the ELx 800 universal microplate reader (Biotek Instrument inc, Highland Park, Vermont, USA). *S. mutans* solutions mixed with phosphate buffered saline (PBS) to give 10^8 cfu/mL. Six decimal dilutions were carried out (1:10, 1:100, 1:1000, 1:10000, 1:100000, 1:1,000,000) and the last three dilutions were plated onto Colombia blood agar. This was our control group also.

2.4. Photo-active disinfection procedure

This study consist of three groups; first group was extract + *S. mutans* that was exposed to light, second group was extract + *S. mutans* that was not exposed to light, and the third group was *S. mutans* only exposed to light without any extract. For the antimicrobial effectiveness colony forming (CFU) units was counted. 100 μL of each extract were mixed with 100 μL *S. mutans* solution in eppendorf tubes. The mixture in eppendorf tubes was maintained in contact for one minute with the aim of pre-irradiation time. Pre-irradiation time is important to keep the extract inside the bacteria and allowing more light absorption [25]. Then the LED device was positioned perpendicularly to tube and irradiate in one single point on the center for one minute (first group). In the second group the mixture in eppendorf tubes was maintained in contact with extract for two minutes because this group was not irradiated. In the third group 100 μL *S. mutans* solution was mixed with 100 μL of distilled water because this group was tested only with light source without any extract. Each experiment was repeated three times. And all samples transferred to Colombia blood agar for CFU counting.

2.5. Antimicrobial assay

At the end of the PAD procedure all plates were incubated at 37°C in 10% CO_2 for 24 h. After the incubation period, cfu per plate counted, so the reduction of bacteria was calculated.

3. Results

3.1. Maximum absorbances of plant extracts

Spectrophotometric analyses of plant extracts are shown in Fig. 1. In the light of this information, the plant extracts were decided to activate with a 430–480 nm wavelength (blue light) dental light source (Elipar S10[®], 3 M ESPE, USA) for one minute.

3.2. Antimicrobial effect

All extracts examined with dental light curing device, no antibacterial effect was found for the second and third applications. In the first application, however, irradiation with extract + *S. mutans* mixture reduced the number of microorganisms in the beginning by 99% for only *Rumex cristatus* DC. extract (log 2) (Fig. 2) (Tables 1–3). The log kill was obtained by the \log_{10} of the control group minus the \log_{10} of the test group according to Concannon et al [26].

Blue light irradiation alone and extract alone showed no significant (log1) antibacterial effect. Besides, in PAD group only *Rumex cristatus* DC. showed a significant antibacterial effect (log 2).

4. Discussion

Dental caries is a localized destruction of dental hard tissue under the influence of acids produced by bacteria [27]. The primary pathogenic bacteria of dental caries is considered as *S. mutans* [28]. Therefore, extracts have been tested against *S. mutans* in this study. Besides,

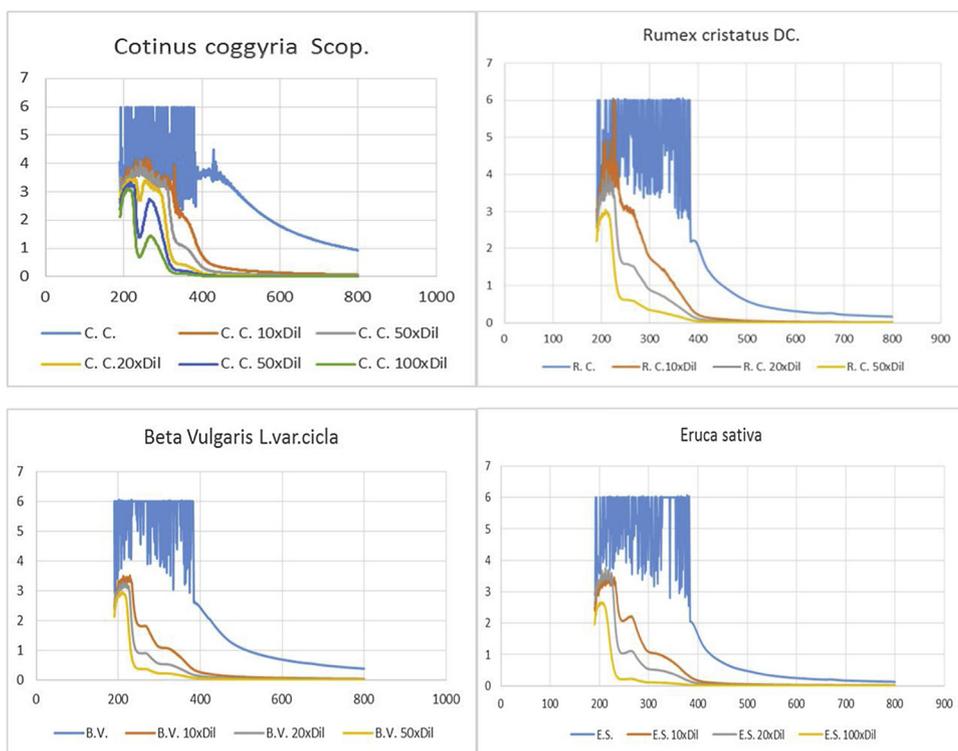


Fig. 1. Spectrophotometric analyses of plant extracts.

Each plant extract's absorption spectrum was measured between 300–900 nm and the light emission pattern of the light was evaluated. All plant extracts were activated with a 430–480 nm wavelength (blue light) dental light source (Elipar S10®, 3 M ESPE, USA) for one minute.

C.C.: *Cotinus coggyria Scop.*, R.C.: *Rumex cristatus DC.*, B.V.: *Beta vulgaris L.var.cicla*; E.S.: *Eruca sativa*; Dil : Dilution (for example 10xDil means ten times dilution).

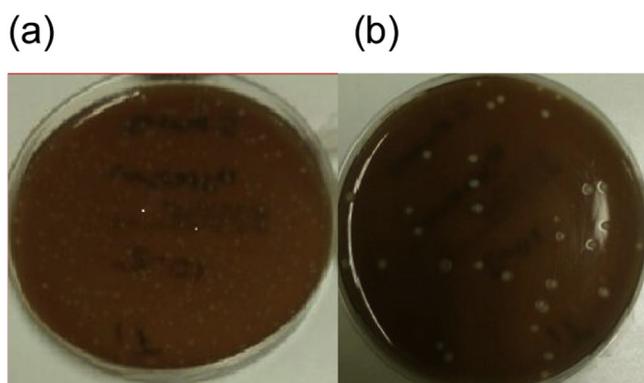


Fig. 2. Antibacterial effect of *Rumex cristatus DC.* Agar plate view of (a) Control group, and (b) *Rumex cristatus DC.* after irradiation. *Rumex cristatus DC.* showed a significant antibacterial effect (log 2). Blue light irradiation with extract + *S. mutans* mixture reduced the number of microorganisms in the beginning by 99% for only *Rumex cristatus DC.* extract.

in many other studies PSs were tested against *lactobacillus* species, which were also responsible for tooth decay [2,7,8]. A good PS should absorb light efficiently, and its excited state should be long lived in order to have time to react with neighbouring target molecules. Most compounds that form excited states that are able to produce radicals and reactive oxygen species, have a tricyclic, heterocyclic or porphyrin-like ring structures with conjugated double bonds. Small changes in the molecular structure of PS may influence its photosensitising property.

Photodynamic therapy works in two steps; application of PSs, then activation with a light source. Several studies demonstrated that different PSs and light sources can be used for PAD. In principle, any visible light can be used for light source [29] like as red light [3], halogens [30] and blue light [7]. In this way, blue-light becomes popular in dentistry, because almost all the dental clinics have these device to use for dental treatment. And most of the PSs could activate with blue light [2,31,32]. There are a lot of PS, but especially for disinfection mainly toluidine blue and methylene blue are used [2,3,7]. Moreover,

Table 1
Reduction of bacterial growth in the first group.

FIRST GROUP	Microorganism count at the beginning	Microorganism count after irradiation	Log decrease
<i>Beta vulgaris</i> L.var.cicla + <i>S. mutans</i> + LED	$2,5 \times 10^8$	1 st repeat: 1×10^8 2 nd repeat: 1×10^8 3 rd repeat: 8×10^7	< 1 log
<i>Eruca sativa</i> + <i>S. mutans</i> + LED	$2,5 \times 10^8$	1 st repeat: $1,2 \times 10^8$ 2 nd repeat: 1×10^8 3 rd repeat: 1×10^8	< 1 log
<i>Cotinus coggyria</i> Scop. + <i>S. mutans</i> + LED	$2,5 \times 10^8$	1 st repeat: 6×10^7 2 nd repeat: 8×10^7 3 rd repeat: $9,5 \times 10^7$	< 1 log
<i>Rumex cristatus DC.</i> + <i>S. mutans</i> + LED	$2,5 \times 10^8$	1 st repeat: $2,5 \times 10^7$ 2 nd repeat: $2,5 \times 10^7$ 3 rd repeat: $2,3 \times 10^7$	2 log 2 log > 2 log

Table 2
Reduction of bacterial growth in the second group.

SECOND GROUP	Microorganism count at the beginning	Microorganism count after contact with extract	Log decrease
<i>Beta vulgaris</i> L.var.cicla + <i>S. mutans</i>	$2,5 \times 10^8$	1 st repeat: $1,5 \times 10^8$ 2 nd repeat: $1,2 \times 10^8$ 3 rd repeat: $1,8 \times 10^8$	< 1 log
<i>Eruca sativa</i> + <i>S. mutans</i>	$2,5 \times 10^8$	1 st repeat: 2×10^8 2 nd repeat: $1,1 \times 10^8$ 3 rd repeat: 1×10^8	< 1 log
<i>Cotinus coggyria</i> Scop. + <i>S. mutans</i>	$2,5 \times 10^8$	1 st repeat: 9×10^7 2 nd repeat: $1,2 \times 10^8$ 3 rd repeat: 1×10^8	< 1 log
<i>Rumex cristatus DC.</i> + <i>S. mutans</i>	$2,5 \times 10^8$	1 st repeat: 8×10^7 2 nd repeat: 6×10^7 3 rd repeat: $7,5 \times 10^7$	< 1 log

Table 3
Reduction of bacterial growth in the third group.

THIRD GROUP	Microorganism count at the beginning	Microorganism count after irradiation	Log decrease
<i>S. mutans</i> +LED	$2,5 \times 10^8$	1 st repeat: 2×10^8 2 nd repeat: $2,3 \times 10^8$ 3 rd repeat: $2,2 \times 10^8$	< 1 log

in recent years, some plant-derived PSs have also been used to prevent possible side effects. Nakamura and friends [31] used proanthocyanidin, in addition Luthi and friends [32] used hypericin as a new disinfection technique.

For this purpose we wanted to examine *Cotinus coggygia* Scop., *Rumex cristatus* DC., *Beta vulgaris* L.var.cicla, and *Eruca sativa* extracts as a PS. The plant extract such as *Cotinus coggygia* Scop., *Rumex cristatus* DC., *Beta vulgaris* L.var.cicla, and *Eruca sativa* have antioxidant and antimicrobial activities [9,17,18]. For this reason, these plants were chosen for this study.

All extracts examined with dental light curing device, and one of them (*Rumex cristatus* DC.) was found effective against *S. mutans* for antimicrobial therapy. As far as we know, this is the first study which investigates the antibacterial efficiencies of these extracts against *S. mutans*. Rumex genus have been used in traditional medicine for constipation, blood purification, and inflammation [33]. Also, there are some studies that showed its antioxidant activities [33,34].

Other variables of these researches are pre-irradiation and irradiation time of PSs. Pre-irradiation time, is notified to be an important factor for the PSs, to diffuse into the tissue [2]. Although Bulit et al expose the biofilm with PS for 15 min due to pre-irradiation time, most *in vivo* studies apply only 5 min [2,6,30,35]. This study, left the extracts in contact with the bacterial solution for one minute. Steiner-Oliveria C. et al [3] also give 1 min pre-irradiation time as our study.

After preirradiation time, we irradiate for one minute, also like Steiner-Oliveria C. et al. [3]. The purpose of the short application time, is to be tolerable for patients, during dental treatment. Also longer irradiation time can arise pulpal temperature [36]. Studies revealed that heat threshold of pulp is 5 °C to avoid pulp damage [37,38]. For that reason, we use very limited irradiation time.

5. Conclusion

The results of our study showed that *Rumex cristatus* DC. aqueous extracts can be used as an alternative in PAD of cavities in dentistry. In the limitations of our study we could not find any antibacterial effect of *Cotinus coggygia* Scop., *Beta vulgaris* L.var.cicla and *Eruca sativa*. Nevertheless more study needs to decide the antibacterial effect of these extracts. Furthermore our study also highlights that more plant derivatives can be examined as PSs.

Conflict of interest

The authors declare that they have no conflict of interests.

Acknowledgment

This work was financially supported by the by the Marmara University Scientific Research Projects Commission [grant number SAG-C-DRP-131016-0452].

References

- [1] M.J. Tyas, K.J. Anusavice, J.E. Frenken, G.J. Mount, Minimal intervention dentistry—a review, *Int. Dent. J.* 50 (1) (2000) 1–12, <https://doi.org/10.1111/j.1875-595X.2000.tb00540.x>.
- [2] C. Guglielmi, M. Luz, M. Simonato, K. Ramalho, J. Imparato, S. Pinheiro, Clinical use of photodynamic antimicrobial chemotherapy for the treatment of deep carious lesions, *J. Biomed. Opt.* 16 (8) (2011) 088003, <https://doi.org/10.1117/1.3611009>.
- [3] C. Steiner-Oliveria, P.L. Longo, A.C.C. Aranha, K.M. Ramalho, M.P.A. Mayer, C.P. Eduardo, Randomized in vivo evaluation of photodynamic antimicrobial chemotherapy on deciduous carious dentin, *J. Biomed. Opt.* 20 (10) (2015) 108003, <https://doi.org/10.1117/1.JBO.20.10.108003>.
- [4] P.V.M. Uday Mohan, K.S. Uloopi, C. Vinay, R. Chandrasekhar Rao, In vivo comparison of cavity disinfection efficacy with APF gel, Propolis, Diode Laser, and 2% chlorhexidine in primary teeth, *Contemp. Clin. Dent.* 7 (1) (2016) 45–50, <https://doi.org/10.4103/0976-237X.177110>.
- [5] R. Fekrazad, M. Bargrizan, S. Sajadi, Evaluation of the effect of photoactivated disinfection with Radachlorin® against *Streptococcus mutans* (an in vitro study), *Photodiagn. Photodyn. Ther.* 8 (3) (2011) 249–253, <https://doi.org/10.1016/j.pdpdt.2011.03.337>.
- [6] J.P. Longo, S.C. Leal, A.R. Simioni, M. de Fatima Menezes Almeida-Santos, A.C. Tedesco, R.B. Azevedo, Photodynamic therapy disinfection of carious tissue mediated by aluminum-chloride-phthalocyanine entrapped in cationic liposomes: an in vitro and clinical study, *Lasers Med. Sci.* 27 (2012) 575–584, <https://doi.org/10.1007/s10103-011-0962-6>.
- [7] F. Bulit, I. Grad, D. Manoil, S. Simon, J.C. Wataha, A. Filieri, A. Feki, J. Schrenzel, N. Lange, S. Boullaguet, Antimicrobial activity and cytotoxicity of 3 photosensitizers activated with blue light, *JOE* 40 (3) (2014) 427–431, <https://doi.org/10.1016/j.joen.2013.12.001>.
- [8] K. Mahabala, S. Shrikrishna, S. Natarajan, A. Nayak, Ethnolic extracts of aloe vera and propolis as cavity disinfectants: an in vitro study, *Dent. Hypotheses* 7 (2) (2016) 61–66, <https://doi.org/10.4103/2155-8213.183769>.
- [9] M. Marčetić, D. Božić, M. Milenković, N. Malešević, S. Radulović, N. Kovačević, Antimicrobial, antioxidant and anti-inflammatory activity of young shoots of the smoke tree, *Cotinus coggygia* Scop., *Phytother. Res.* 27 (11) (2013) 1658–1663, <https://doi.org/10.1002/ptr.4919>.
- [10] S. Matic, S. Stanić, M. Mihailović, D. Bogojević, *Cotinus coggygia* Scop.: an overview of its chemical constituents, pharmacological and toxicological potential, *Saudi J. Biol. Sci.* 23 (4) (2016) 452–461, <https://doi.org/10.1016/j.sjbs.2015.05.012>.
- [11] S. Matic, S. Stanić, D. Bogojević, M. Vidaković, N. Grdović, J. Arambašić, S. Dinić, A. Uskoković, G. Poznanović, S. Solujić, M. Mladenović, J. Marković, M. Mihailović, Extract of the plant *Cotinus coggygia* Scop. attenuates pyrogallol-induced hepatic oxidative stress in Wistar rats, *Can. J. Physiol. Pharmacol.* 89 (7) (2011) 401–411, <https://doi.org/10.1139/y11-04>.
- [12] A. Yarat, O. Sacan, A. Akyuz, B. Alev, R. Pisiriciler, E. Ak, R. Yanardag, In vitro effect of aqueous plant extracts on antioxidant parameters in saliva samples, *J. Med. Plants Res.* 7 (3) (2013) 118–125, <https://doi.org/10.5897/JMPRO12.530>.
- [13] A. Vasas, O. Orbán-Gyapai, J. Hohmann, The Genus *Rumex*: review of traditional uses, phytochemistry and pharmacology, *J. Ethnopharmacol.* 175 (2015) 198–228, <https://doi.org/10.1016/j.jep.2015.09.001>.
- [14] O. Sacan, R. Yanardag, Antioxidant and anticholinesterase activities of chard (*Beta vulgaris* L. var. cicla), *Food Chem. Toxicol.* 48 (5) (2010) 1275–1280, <https://doi.org/10.1016/j.fct.2010.02.022>.
- [15] S. Gezginci-Oktayoglu, O. Sacan, S. Bolvent, Y. Ipci, L. Kabasakal, G. Sener, R. Yanardag, Chard (*Beta vulgaris* L. var. cicla) extract ameliorates hyperglycemia by increasing GLUT2 through Akt2 and antioxidant defense in the liver of rats, *Acta Histochem.* 116 (1) (2014) 32–39, <https://doi.org/10.1016/j.acthis.2013.04.016>.
- [16] G.J.A. Kapadia, M. Azuine, G. Subba Rao, T. Arai, A. Lida, H. Tokuda, Cytotoxic effect of the red beetroot (*Beta vulgaris* L.) extract compared to doxorubicin (Adriamycin) in the human prostate (PC-3) and breast (MCF-7) cancer cell lines, *Anti-Cancer Agents Med. Chem. (Formerly Current Medicinal Chemistry-Anti-Cancer Agents)* 11 (3) (2011) 280–284, <https://doi.org/10.2174/187152011795347504>.
- [17] E. Bursal, Kinetic properties of peroxidase enzyme from chard (*Beta vulgaris* Subspecies cicla) leaves, *Int. J. Food Prop.* 16 (6) (2013) 1293–1303, <https://doi.org/10.1080/10942912.2011.585729>.
- [18] L. Gennari, M. Felletti, M. Blasa, D. Angelino, C. Celeghini, A. Corallini, F. Ninfali, Total extract of *Beta vulgaris* var. cicla seeds versus its purified phenolic components: antioxidant activities and antiproliferative effects against colon cancer cells, *Phytochem. Anal.* 22 (3) (2011) 272–279, <https://doi.org/10.1002/pca.1276>.
- [19] U.V. Ustundag, S. Tunalı, B. Alev, H. Ipekci, E. Emekli-Alturfan, T.T. Akbay, R. Yanardag, A. Yarat, Effects of Chard (*Beta Vulgaris* L. Var. Cicla) on cardiac damage in valproic acid-induced toxicity, *J. Food Biochem.* 40 (2) (2016) 132–139, <https://doi.org/10.1111/jfbc.12202>.
- [20] G.H. Mahran, H.A. Kadry, Z.G. Isaac, C.K. Thabet, M.M. Al-Azizi, M.M. El-Olemy, Investigation of diuretic drug plants. 1. Phytochemical screening and pharmacological evaluation of *Anethum graveolens* L., *Apium graveolens* L., *Daucus carota* L. and *Eruca sativa* Mill., *Phytother. Res.* 5 (4) (1991) 169–172, <https://doi.org/10.1002/ptr.2650050406>.
- [21] D. Esiyok, M.K. Bozokalfa, T.K. Aşçıoğlu, Widely utilized wild edible plants: a case study from Turkey, *Global Perspectives on Underutilized Crops*, Springer, Cham, 2018, pp. 217–257, https://doi.org/10.1007/978-3-319-77776-4_9.
- [22] L. Bell, L. Methven, A. Signore, M.J. Oruna-Concha, C. Wagstaff, Analysis of seven salad rocket (*Eruca sativa*) accessions: The relationships between sensory attributes and volatile and non-volatile compounds, *Food Chem.* 218 (2017) 181–191, <https://doi.org/10.1016/j.foodchem.2016.09.076>.
- [23] O. Sacan, H. Orak, R. Yanardag, Antioxidant activity of water extract of *Eruca sativa* Mill., *Asian J. Chem.* 20 (5) (2008) 3462.
- [24] M.F. Taviano, A. Melchini, A. Filocamo, C. Costa, S. Catania, R. Raciti, S. Saha, P. Needs, G.G. Bisignano, N. Miceli, Contribution of the glucosinolate fraction to the

- overall antioxidant potential, cytoprotection against oxidative insult and antimicrobial activity of *Eruca sativa* Mill. leaves extract, *Pharmacogn. Mag.* 13 (52) (2017) 738–743 doi: 10.4103/pm.pm.245.16.
- [25] I.M. Bevilacqua, R.A. Nicolau, S. Khouri, A. Brugnera, G.R. Teodoro, R.A. Zangaro, The impact of photodynamic therapy on the viability of *Streptococcus mutans* in a planktonic culture, *Photomed. Laser Surg.* 25 (6) (2007) 513–518, <https://doi.org/10.1089/pho.2007.2109>.
- [26] S.P. Concannon, T.D. Crowe, J.J. Abercrombie, C.M. Molina, P. Hou, D.K. Sukumaran, P.A. Raj, K.P. Leung, Susceptibility of oral bacteria to an antimicrobial decapeptide, *J. Med. Microbiol.* 52 (2003) 1083–1093, <https://doi.org/10.1099/jmm.0.05286-0>.
- [27] I. Strużycka, The oral microbiome in dental caries, *Polish J. Microbiol.* 63 (2) (2014) 127–135.
- [28] N. Hoiby, O. Gíofu, H.K. Johansen, Z. Song, C. Moser, P.O. Jensen, S. Molin, M. Givskov, T. Tolker-Nielsen, T. Bjarnsholt, The clinical impact of bacterial biofilms, *Int. J. Oral Sci.* 3 (2) (2011) 55–65, <https://doi.org/10.4248/IJOS11026>.
- [29] R. Bonnett, Photosensitizers of the porphyrin and phthalocyanine series for photodynamic therapy, *Chem. Soc. Rev.* 24 (1) (1995) 19–33.
- [30] P. Araujo, J.F. Correia-Silva, R. Gomez, M.L.A. Massara, M.A. Cortes, L.Y.A. Poletto, Antimicrobial effect of photodynamic therapy in carious lesions in vivo, using culture and real time PCR methods, *Photodiagn. Photodyn. Ther.* 12 (3) (2015) 401–407, <https://doi.org/10.1016/j.pdpdt.2015.06.003> (2015).
- [31] K. Nakamura, M. Shirato, H. Ikai, T. Kanno, K. Sasaki, M. Kohno, Y. Niwano, Photoirradiation of proanthocyanidin as a new disinfection technique via reactive oxygen species formation, *PLoS One* 8 (3) (2013) e60053, <https://doi.org/10.1371/journal.pone.0060053>.
- [32] M. Lüthi, E.B. Gyenge, M. Engström, M. Bredell, K. Gratz, H. Walt, R. Gmür, C. Maake, Hypericin-and mTHPC-mediated photodynamic therapy for the treatment of cariogenic bacteria, *Med. Laser Appl.* 24 (4) (2009) 227–236.
- [33] S. Kahraman, R. Yanardağ, (Antioxidant activity of ethanolic extract from *Rumex crispatus* Dc, *IJEMME* 2 (4) (2012) 319–326.
- [34] E. Avci, G.A. Avci, D.A. Kose, A.A. Emniyet, M. Suicmez, In vitro antimicrobial and antioxidant activities and GC/MS analysis of the essential oils of *Rumex crispus* and *Rumex crispatus*, *Hacettepe J. Biol. Chem.* 42 (2) (2014) 193–199.
- [35] P. Neves, L. Lima, F. Rodrigues, T. Leitao, C. Ribeiro, Clinical effect of photodynamic therapy on primary carious dentin after partial caries removal, *Braz. Oral Res.* 30 (1) (2016), <https://doi.org/10.1590/1807-3107BOR-2016.vol30.0047>.
- [36] G. Reggiani-Mello, R.R. Krueger, Comparison of commercially available femtosecond lasers in refractive surgery, *Expert Rev. Ophthalmol.* 6 (1) (2011) 55–65, <https://doi.org/10.1586/eop.10.80>.
- [37] M. Mirzaie, E. Yassini, S. Ashnagar, A. Hadadi, N. Chiniforush, Evaluation of temperature change during antimicrobial photodynamic therapy with two different photosensitizers in dental caries, *Photodiagn. Photodyn. Ther.* 14 (2016) 115–118.
- [38] H. El Yazami, T. Zeinoun, S. Bou Saba, L. Lamard, A. Peremans, M. Limme, S. Geerts, M. Lamy, S. Nammour, Pulp temperature increase during photo-activated disinfection (PAD) of periodontal pockets: an in vitro study, *Lasers Med. Sci.* 25 (5) (2010) 655–659, <https://doi.org/10.1007/s10103-009-0686-z>.