



In vitro screening of extracts from 38 marine animal resources for novel cosmeceutical activities

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Abstract

Marine resources have various biological activities and their constituents are more novel than those of land organisms. Several biologically active constituents have been found in marine organisms. Recently, many studies have reported that marine animals (MAs) can be used as functional ingredients in functional foods or nutraceutical due to their health benefits. However, no studies have extensively investigated the cosmeceutical activities of MAs extracts. Here, 70% ethanol extracts of 38 MAs were investigated for their activities of whitening and anti-aging properties for use as materials in novel cosmeceuticals. Anti-aging activities were determined by skin aging-related enzyme activities (anti-collagenase, anti-elastase, anti-hyaluronidase) and whitening activities (anti-tyrosinase, anti-3,4-dihydroxyl-L-phenylalanine [DOPA] oxidation) evaluated by colorimetric method. Among the 38 MAs, we found that *Urechis unicinctus* and *Petrosia corticata* extracts showed the strongest inhibitory effects against tyrosinase and DOPA oxidation, respectively. Our results additionally showed that *Protankyra bidentata* extract might provide a major source of anti-hyaluronidase and anti-elastase; meanwhile, anti-collagenase effects were similar in most MAs. Overall, these results suggest that extracts of marine animals have potential as a tyrosinase, collagenase, elastase, and hyaluronidase inhibitors. Taken together, MA resources could be considered as a novel cosmeceutical agent to be applied in cosmetic industry.

Keywords: Marine animals, Anti-aging, Skin aging-related enzyme, Cosmeceuticals

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Introduction

The skin is a voluminous organ that plays a major role in protecting the human body from environmental factors like temperature, pollutants and ultraviolet (UV) radiation. To a recent date, skin aging has received increased interest for research, and a wide number of studies about skin aging is being carried out (Jiratchayamaethasakul et al., 2020). Both intrinsic and extrinsic factors could influence skin aging. Intrinsic aging is characterized by an imbalance in hormone secretion, immunoprotein and structural protein production triggered by a decline in immunocyte and skin cell activity. The other, extrinsic aging includes freckle and age spots as well as wrinkle-formation, elasticity loss, and enhancement of asteatosis provoked by various pollutants and UV radiation-induced photoaging (Kim et al., 2008a).

The skin consists of three layers; the epidermis, dermis, and subcutaneous tissue. The outermost part is the extracellular matrix (ECM) and is organized with fibroblasts and structural proteins such as collagen and elastin (Im et al., 2019). The ECM is involved in cell growth and structural stability of the skin; and also promotes constructive remodeling of tissue in all parts of the human body. Skin aging interferes with this process based on the activities of collagenase, elastase, and hyaluronidase, the decomposers of structural proteins (Ndlovu et al., 2013).

Collagen is one of the main structural proteins most abundant in ECM and is essential for maintaining skin flexibility, by means of elasticity and integrity. Meanwhile, elastin, the key protein located in the elastic fibers of connective tissue, is significant for its unique elastic recoil properties (Bravo et al., 2016). Hyaluronic acid (HA), also called hyaluronan, contributes to maintaining the appropriate form of collagen and elastin, water-holding capability related to various physiological activities including skin moisturizing, regeneration, and consequently, anti-aging properties (Bukhari et al., 2018; Ding et al., 2018). As mentioned above, dermal enzymes indirectly activated by external stimulus destruct collagen, elastin, and HA, causing premature skin aging represented by wrinkle, freckles, saggy and chappy skin or dermatopathy like rubeosis, asteatosis (Peres et al., 2011).

Melanin is the main component of skin and hair, black or brown colored pigment synthesized from epidermal melanocytes via melanogenesis. Although melanin pigmentation plays a key role in protecting the skin against the damage of UV radiation from sunlight, its overproduction can lead to skin

melanism and aging, such as spots, lentigines, and even cutaneous carcinoma (Gam et al., 2021; Im et al., 2019). Tyrosinase is an enzyme closely related to melanogenesis that functions as a rate-limiting step. Its participation in the melanogenic pathway is progressed by three steps at large: tyrosinase hydroxylates tyrosine to 3,4-dihydroxyl-L-phenylalanine (DOPA) and oxidizes DOPA to dopaquinone. The derivatives from dopaquinone following the multilayer reactions enhance the contents of melanin in the human epidermis (Chan et al., 2011; Lin et al., 2015).

Marine organisms are reasonably promising resources from nature and ideal products for investigating and extracting biologically active substances with potential in health functional food, pharmaceuticals, and food industries (Kim et al., 2008b). Recently, marine organisms, especially marine animals (MAs) have been revealed diverse biological activities like antioxidant, anti-cancer, anti-inflammatory, antifungals, and antivirals based on the compounds contained (Gupta et al., 2014). Polyphenols, polysaccharides, peptides, growth factors, and unsaturated fatty acids are examples of functional materials that have a variety of beneficial effects on the human body (Kim, 2014). However, a less number of previous research have explored the use of MAs in cosmeceutical researches. Thus, the objective of the present study was to explore the anti-collagenase, anti-elastase, anti-hyaluronidase, anti-tyrosinase, and anti-DOPA oxidation activities of 38 MAs extract collected from the coast of South Korea to evaluate the possibility of cosmeceutical industrial application.

Materials and Methods

Chemicals and reagents

Tyrosinase from mushroom, L-tyrosine, 3,4-dihydroxyl-L-phenylalanine (L-DOPA), collagenase from *Clostridium histolyticum*, N-[3-(2-Furyl)acryloyl]-Leu-Gly-Pro-Ala (FALGPA), N-succinyl-Ala-Ala-Ala-p-nitroanilide (AAAPN), elastase from porcine pancreas, hyaluronidase from bovine testes, HA sodium salt from rooster comb, tricine, 4-(dimethylamino) benzaldehyde (DMAB) were purchased from Sigma-Aldrich Chemical (St. Louis, MO, USA) and all other chemicals and solvents used for this study were of analytical grade.

Sample preparation

70% ethanol extract of 38 MAs used in this study was kindly provided by the National Marine Biodiversity Institute of Korea (MABIK) as shown in Table 1.

Table 1. Scientific name of MAs used in this study

MAs	Scientific name (Korean name)
MA. 1	<i>Hymeniacion sinapium</i> (주황해면해면)
MA. 2	<i>Aurelia aurita</i> (보름달물해파리)
MA. 3	<i>Bullacta exarata</i> (민챙이)
MA. 4	<i>Patiria pectinifera</i> (별불가사리)
MA. 5	<i>Cliona celata</i> (호박해면)
MA. 6	<i>Protankyra bidentata</i> (가시닷해삼)
MA. 7	<i>Scaphechinus mirabilis</i> (연잎성게)
MA. 8	<i>Saxidomus purpurata</i> (개조개)
MA. 9	<i>Octopus minor</i> (낙지)
MA. 10	<i>Chicoreus asianus</i> (뿔소라)
MA. 11	<i>Octopus vulgaris</i> (참문어)
MA. 12	<i>Mizuhopecten yessoensis</i> (큰가리비)
MA. 13	<i>Atrina pectinata</i> (키조개)
MA. 14	<i>Ostrea denselamellosa</i> (토굴)
MA. 15	<i>Rapana venosa</i> (피뿔고둥)
MA. 16	<i>Uroteuthis chinensis</i> (한치꽃뚜기)
MA. 17	<i>Mytilus unguiculatus</i> (홍합)
MA. 18	<i>Styela clava</i> (미더덕)
MA. 19	<i>Petrosia corticata</i> (불똥해면)
MA. 20	<i>Callyspongia elegans</i> (예쁜이해면)
MA. 21	<i>Suberites excellens</i> (코르크해면)
MA. 22	<i>Apostichopus japonicus</i> (돌기해삼)
MA. 23	<i>Haliotis discus</i> (등근전복)
MA. 24	<i>Styela plicata</i> (주름미더덕)
MA. 25	<i>Halocynthia roretzi</i> (우렁챙이)
MA. 26	<i>Mytilus galloprovincialis</i> (지중해담치)
MA. 27	<i>Spirastella insignis</i> (굵은나선별해면)
MA. 28	<i>Aplidium pliciferum</i> (만두멍게)
MA. 29	<i>Neptunea cumingii</i> (갈색띠매물고둥)
MA. 30	<i>Aplysia kurodai</i> (둥근성게)
MA. 31	<i>Eupentacta quinquesemita</i> (오각광삼)
MA. 32	<i>Swiftopecten swiftii</i> (고랑가리비)
MA. 33	<i>Urechis unicinctus</i> (개불)
MA. 34	<i>Engraulis japonicus</i> (멸치)
MA. 35	<i>Liolophura japonica</i> (군부)
MA. 36	<i>Andara broughtonii</i> (피조개)
MA. 37	<i>Ruditapes philippinarum</i> (바지락)
MA. 38	<i>Portunus trituberculatus</i> (꽃게)

MA, marine animal.

Collagenase inhibitory assay

Collagenase inhibitory activity was determined as the method described by Thring et al. (2009) with some modifications. A

50 mM of Tricine buffer with 400 mM NaCl and 10 mM CaCl₂ at pH 7.5 was prepared and used as the buffer solution in this assay. Firstly, Tricine buffer was mixed with 20 μ L of sample in the 96-well plates and preincubated with 20 μ L of collagenase from *C. histolyticum* in Tricine buffer (0.8 collagen degrading units, CDU/mL) at room temperature (RT) for 10 min. After the preincubation, the reaction was initiated by adding 40 μ L of FALGPA collagenase substrate in Tricine buffer (2 mM) to the preincubated mixtures. The absorbance was measured immediately at 340 nm using a microplate reader (SynergyTM HTX Multi-Mode Reader, BioTek Instruments, Winooski, VT, USA).

Elastase inhibitory assay

Elastase inhibition was evaluated based on the method described by Shirzad et al. (2018) with slight modifications. The mixture of AAAPN elastase substrate (1.015 mM) in 0.1232 M Tris-HCl buffer solution (pH 8) was prepared. The substrate solution was mixed with 10 μ L of sample in the 96-well plates, and preincubated at RT for 10 min. Then, 10 μ L of elastase from the porcine pancreas (0.15 units/mL) in Tris-HCl buffer was mixed with the preincubated mixtures to initiate the reaction. Afterward, the absorbance was measured at a 410 nm by a multi plate reader (SynergyTM HTX Multi-Mode Reader).

Hyaluronidase inhibitory assay

Hyaluronidase inhibitory effect was determined by measuring the color intensity of assayed solution using the method of Ferreres et al. (2012) with few modifications. A stock solution of hyaluronidase (8 mg/mL) and HA (2.4 mg/mL) in 0.1 M of acetate buffer (pH 3.6) was prepared. A 10 μ L of hyaluronidase solution and sample were mixed in test tubes, and incubated at 37°C for 20 min. A 20 μ L of calcium chloride (12.5 mM) was added to the mixture and incubated again at 37°C for 20 min. After the incubation, 50 μ L of the HA solution was added and incubated at 37°C for 40 min. Then, the reaction was stopped by adding 2 μ L of sodium hydroxide (0.4 N) and 20 μ L of potassium tetraborate (0.4 N) and incubated in the water bath at 100°C for 3 min. Finally, the mixture solution was cooled down to RT and 600 μ L of DMAB solution containing 0.4 g of DMAB dissolved in 35 mL of 100% acetic acid and 5 mL 10 N hydrochloric acid was added into the mixture to develop the color, then, incubated 37°C for 20 min. The mixture of each test tube was transferred into 96-well plates and the absorbance was measured at a wavelength of 585 nm (SynergyTM HTX Multi-Mode Reader).

Inhibition of tyrosinase activity and DOPA oxidation

Tyrosinase and DOPA oxidation inhibitory activity were modified from the assay proposed by Momtaz et al. (2008). A 0.1 M of sodium phosphate buffer (pH 6.8) was mixed with 10 μ L samples and a tyrosinase enzyme solution in phosphate buffer (1,500 units/mL). Afterwards, 20 μ L of L-tyrosine (1.5 mM) or 40 μ L of L-DOPA (10 mM) were mixed in the 96-well plates and incubated at 37°C for 12 min. The reaction was stopped by incubating on ice for 1 min and the absorbance of the mixture was measured at 490 nm using a microplate reader (Synergy™ HTX Multi-Mode Reader).

Results

Anti-collagenase and anti-elastase activities of marine animals extracts

The inhibitory effects of 38 MAs on collagenase and elastase are shown in Table 2. Firstly, the collagenase inhibitory effects were similar in all 38 MAs to a varying degrees ranging from 14 \pm 3.71% to 37 \pm 0.86%. Among 38 MAs, *Patiria pectinifera* extract showed the highest elastase inhibitory effect with 63 \pm 3.88% whereas anti-collagenase activity was relatively minor with 24 \pm 3.42%. Additionally, *Spirastella insignis* extract showed correspondingly good inhibition in both collagenase and elastase with 37 \pm 0.86% and 44 \pm 4.24% illustrated by the highest collagenase inhibitory effect.

Anti-hyaluronidase activity of marine animals extracts

The hyaluronidase inhibition effects of all MAs extracts at the final concentration of 1 mg/mL are shown in Table 2. *Pro-tankyra bidentata* extract presented the highest hyaluronidase inhibitory effect at 52 \pm 3.28% whereas no significant activities were demonstrated with the rest MAs extracts. Although 37 extracts did not show strong inhibition at 1 mg/mL, *P. bidentata* extract presented a good effect in anti-elastase activity (49 \pm 0.63%). The present results indicate that *P. bidentata* extract has anti-skin aging capacity.

Anti-tyrosinase and anti-DOPA oxidation activities of marine animals extracts

In this study, there are two methods to assess whitening activities which are tyrosinase inhibitory and DOPA-oxidation inhibitory assays. As elucidated in Table 3, the whitening activities were performed to achieve a final concentration of 1 mg/mL for both assays. Among examined 38 MAs, *Urechis uncinatus*

extract possessed the strongest tyrosinase inhibitory effect (60 \pm 4.60%) whereas of which 33 \pm 4.20% DOPA oxidation inhibitory activity was observed. Despite the next highest tyrosinase inhibition at 42 \pm 5.10% expressed by *Chicoreus asianus* extract, the anti-DOPA oxidation effect was not shown. Meanwhile, the highest DOPA-oxidation inhibitory activity was found by *Petrosia corticata* extract at 73 \pm 9.49% and the comparative inhibitory effect was found by *U. uncinatus* extract showing 33 \pm 4.20%. Furthermore, *P. corticata* extract was not shown anti-tyrosinase inhibition with 6 \pm 1.96%, *U. uncinatus* extract contrastively showed the strongest inhibitory effect against both tyrosinase and DOPA-oxidation. These findings imply that *U. uncinatus* extract inhibits melanin synthesis by inhibiting the tyrosinase and DOPA-oxidation.

Discussion

Skin aging, by the stimulus of intrinsic and extrinsic factors, is a natural biological process with the center of attention in various modern industries (Im et al., 2019). Intrinsic or chronological aging results from the natural deterioration of skin as time advanced, while on the other hand, extrinsic or premature aging is the outcome of disclosure to external environment factors like UV radiation, particulate matter, endocrine disruptor, etc. (Mukherjee et al., 2011). Recently, apart from natural aging, immoderate stimuli from the impact of industrialization and that result of proteases (collagenase, elastase, hyaluronidase, and tyrosinase) activities leading to human skin aging is a global issue (Wang et al., 2019).

Marine organisms have exhibited endemic viability in diverse and rough environments leading to their distinctive characteristics resulting from the bio-active substances *in vivo*. These kinds of substances have been widely utilized in traditional medicine and are still reflected as potential and novel drug candidates, cosmetic ingredients, and food supplies in pharmaceutical, cosmetology and food science, respectively (Martins et al., 2014). Among others, marine animals, especially invertebrates such as mollusks, sponges, cnidarians, echinoderms, and tunicates are deeply investigated for their secondary metabolites (including polyphenolic compounds, polysaccharides, peptides, alkaloids, etc.) beneficial to diverse physiological activities (Ebada et al., 2008; Tischler, 2020). However, it is still not deeply explored and illuminated about bioactivity and their mechanism pathway of the marine animals as promising novel cosmeceuticals. Therefore, in this study, we performed *in vitro*

Table 2. Anti-aging activities of 38 marine animal extracts

Sample	Collagenase inhibition (%)	Elastase inhibition (%)	Hyaluronidase inhibition (%)
<i>Hymeniacidon sinapium</i>	28 ± 1.04	35 ± 2.02	6 ± 1.14
<i>Aurelia aurita</i>	19 ± 1.46	29 ± 3.25	-
<i>Bullacta exarata</i>	27 ± 0.81	31 ± 2.94	12 ± 4.54
<i>Patiria pectinifera</i>	24 ± 3.42	63 ± 3.88	7 ± 3.37
<i>Cliona celata</i>	20 ± 1.52	18 ± 2.88	25 ± 11.32
<i>Protankyra bidentata</i>	28 ± 0.32	49 ± 0.63	52 ± 3.28
<i>Scaphechinus mirabilis</i>	28 ± 0.09	33 ± 0.93	22 ± 3.85
<i>Saxidomus purpurata</i>	27 ± 2.09	20 ± 0.95	17 ± 0.50
<i>Octopus minor</i>	29 ± 3.45	47 ± 3.66	7 ± 2.89
<i>Chicoreus asianus</i>	29 ± 3.45	27 ± 2.27	16 ± 3.97
<i>Octopus vulgaris</i>	19 ± 0.95	26 ± 4.97	13 ± 4.45
<i>Mizuhopecten yessoensis</i>	31 ± 0.98	30 ± 2.37	19 ± 1.79
<i>Atrina pectinate</i>	25 ± 0.10	18 ± 4.40	23 ± 1.76
<i>Ostrea denselamellosa</i>	24 ± 2.56	32 ± 2.59	18 ± 2.38
<i>Rapana venosa</i>	28 ± 0.49	34 ± 1.68	0 ± 2.49
<i>Uroteuthis chinensis</i>	23 ± 0.31	27 ± 3.49	12 ± 1.37
<i>Mytilus unguiculatus</i>	27 ± 1.68	26 ± 4.83	17 ± 3.24
<i>Styela clava</i>	31 ± 0.24	34 ± 2.04	2 ± 6.05
<i>Petrosia corticata</i>	23 ± 0.31	25 ± 1.36	8 ± 6.44
<i>Callyspongia elegans</i>	34 ± 2.85	16 ± 1.82	10 ± 3.98
<i>Suberites excellens</i>	19 ± 1.72	43 ± 4.41	6 ± 13.09
<i>Apostichopus japonicus</i>	26 ± 0.82	31 ± 1.45	27 ± 5.88
<i>Haliotis discus</i>	14 ± 3.71	25 ± 2.72	17 ± 4.02
<i>Styela plicata</i>	21 ± 0.51	27 ± 0.71	14 ± 1.83
<i>Halocynthia roretzi</i>	33 ± 1.61	27 ± 2.99	15 ± 6.48
<i>Mytilus galloprovincialis</i>	27 ± 2.07	39 ± 3.93	23 ± 2.58
<i>Spirastella insignis</i>	37 ± 0.86	44 ± 4.24	7 ± 4.18
<i>Aplidium pliciferum</i>	23 ± 0.52	41 ± 1.42	17 ± 1.13
<i>Neptunea cumingii</i>	30 ± 0.69	30 ± 2.59	23 ± 5.73
<i>Aplysia kurodai</i>	29 ± 1.22	8 ± 5.62	21 ± 4.18
<i>Eupentacta quinquesemita</i>	27 ± 1.74	32 ± 6.84	34 ± 3.43
<i>Swiftopecten swiftii</i>	26 ± 1.04	27 ± 1.77	19 ± 2.34
<i>Urechis unicinctus</i>	26 ± 3.29	33 ± 2.02	8 ± 3.77
<i>Engraulis japonicus</i>	34 ± 1.48	10 ± 1.04	11 ± 5.27
<i>Liolophura japonica</i>	21 ± 1.25	30 ± 0.75	10 ± 1.35
<i>Andara broughtonii</i>	25 ± 2.61	20 ± 0.32	18 ± 2.75
<i>Ruditapes philippinarum</i>	28 ± 0.91	14 ± 1.34	10 ± 5.24
<i>Portunus trituberculatus</i>	26 ± 0.17	20 ± 5.39	-

The values are expressed as the mean ± SD in triplicate experiments. The final concentration of tested samples was 1 mg/mL.

cosmeceutical activity of 38 MAs collected from the coast of South Korea to evaluate the possibility of cosmeceutical industrial application.

Collagen and elastin, the main component of the ECM, critical for maintaining skin lubricity and elasticity are disassembled by collagenase, and elastase that play a central role in

Table 3. Whitening activities of 38 marine animal extracts

Sample	Tyrosinase inhibition (%)	DOPA inhibition (%)
<i>Hymeniacion sinapium</i>	19 ± 1.08	13 ± 2.59
<i>Aurelia aurita</i>	17 ± 2.84	3 ± 6.60
<i>Bullacta exarata</i>	9 ± 0.24	2 ± 3.92
<i>Patiria pectinifera</i>	14 ± 0.44	3 ± 6.94
<i>Cliona celata</i>	16 ± 3.68	-
<i>Protankyra bidentata</i>	24 ± 5.60	-
<i>Scaphechinus mirabilis</i>	10 ± 3.51	-
<i>Saxidomus purpurata</i>	17 ± 2.32	-
<i>Octopus minor</i>	13 ± 2.39	5 ± 1.81
<i>Chicoreus asianus</i>	42 ± 5.10	-
<i>Octopus vulgaris</i>	-	13 ± 1.31
<i>Mizuhopecten yessoensis</i>	21 ± 1.40	7 ± 8.83
<i>Atrina pectinata</i>	16 ± 2.72	11 ± 9.25
<i>Ostrea denselamellosa</i>	26 ± 3.99	13 ± 1.97
<i>Rapana venosa</i>	11 ± 8.38	-
<i>Uroteuthis chinensis</i>	2 ± 3.68	10 ± 2.88
<i>Mytilus unguiculatus</i>	9 ± 2.39	7 ± 0.65
<i>Styela clava</i>	21 ± 1.29	-
<i>Petrosia corticata</i>	6 ± 1.96	73 ± 9.49
<i>Callyspongia elegans</i>	18 ± 2.36	-
<i>Suberites excellens</i>	10 ± 2.03	8 ± 12.59
<i>Apostichopus japonicus</i>	24 ± 3.29	-
<i>Haliotis discus</i>	10 ± 2.07	12 ± 3.95
<i>Styela plicata</i>	25 ± 1.48	6 ± 8.20
<i>Halocynthia roretzi</i>	17 ± 2.05	20 ± 0.73
<i>Mytilus galloprovincialis</i>	17 ± 4.55	7 ± 7.31
<i>Spirastella insignis</i>	25 ± 1.71	29 ± 6.58
<i>Aplidium pliciferum</i>	15 ± 4.16	-
<i>Neptunea cumingii</i>	25 ± 6.51	-
<i>Aplysia kurodai</i>	3 ± 5.96	11 ± 8.32
<i>Eupentacta quinquesemita</i>	30 ± 4.26	-
<i>Swiftopecten swiftii</i>	40 ± 1.55	0 ± 17.77
<i>Urechis unicinctus</i>	60 ± 4.60	33 ± 4.20
<i>Engraulis japonicus</i>	11 ± 0.25	13 ± 6.81
<i>Liolophura japonica</i>	11 ± 2.75	4 ± 4.03
<i>Andara broughtonii</i>	17 ± 2.43	-
<i>Ruditapes philippinarum</i>	17 ± 2.90	11 ± 6.35
<i>Portunus trituberculatus</i>	3 ± 2.40	-

The values are expressed as the mean ± SD in triplicate experiments. The final concentration of tested samples was 1 mg/mL.

hydrolyzing structural proteins and wrinkle formation following skin aging (Ndlovu et al., 2013; Wahab et al., 2014). HA is a glycosaminoglycan in ECM, vital for various physiological

activities including anti-inflammatory, anticancer, and anti-diabetic as well as cosmetic properties like skin moisturizing via its water-retentive capacity. Hyaluronidase is responsible for the breakdown of HA, leading to skin dehydration, wrinkle formation, and eventually skin aging (Bravo et al., 2016; Bukhari et al., 2018). Tyrosinase plays a key role in melanin synthesis, and contributes to hyperpigmentation and skin aging, through a series of oxidative polymerization following DOPA and DOPA quinone (Pak et al., 2016). Thus, the repression of skin aging-related enzymes (collagenase, elastase, hyaluronidase, and tyrosinase) that involved in the destruction of ECM components, and melanogenesis, is regarded as a core strategy against dermal senility and dermatopathy. Consequently, the inhibitory effects of collagenase, elastase, hyaluronidase, tyrosinase, and DOPA oxidation of 38 MAs were investigated at the final concentration of 1 mg/mL as summarized in Tables 2 and 3.

Our screening results showed that *P. bidentata* extract indicated the significant anti-aging activities with anti-hyaluronidase (52 ± 3.28%) and anti-elastase (49 ± 0.63%). These results suggest that *P. bidentata* extract could inhibit hyaluronidase and elastase activity and that such inhibitory effect might be attributed to increased HA production and decreased skin-wrinkle generation. Additionally, a study by Shen et al. (2013) reported the purification and potential of antitumor activity both *in vitro* and *in vivo*. Taken together, our study investigated the anti-aging effect of *P. bidentata* extract firstly and it is worth that more experiments would be performed.

According to the whitening activities from 38 MAs extracts, *U. unicinctus* showed the highest tyrosinase inhibition activity (60 ± 4.60%) with minor DOPA oxidation inhibition (33 ± 4.20%). There were numerous studies on its physiological properties such as anti-microbial, anticoagulant, and anti-thrombosis and bioactive substances like collagen, glycosaminoglycan, neuropeptides, and even lysozyme (Bi et al., 2013; Oh et al., 2018; Sung et al., 2008). Moreover, a recent study showed the enrichment method of bio-peptides extracted from *U. unicinctus* protein hydrolysate to enhance its economic value (Li et al., 2021). Results of this study suggested that *U. unicinctus* could be potential for utilization as a whitening agent candidate for the first time.

Overall, the above results suggest that MAs extract could act as potent inhibitors of skin aging-related key enzyme. In addition, our current findings provide preliminary confirmation of anti-aging and whitening activities of MAs extracts. Furthermore, further studies are needed to identify 50% inhibitory concentration and skin cell-based activity.

Conclusion

In this study, we demonstrated both the anti-aging and whitening activities of 70% ethanol extracts of 38 MAs collected from the coast of South Korea. This study examined anti-collagenase, anti-elastase, anti-hyaluronidase, anti-tyrosinase, and anti-DOPA oxidation activities to find novel cosmeceutical candidate from MAs. From the results, the *P. bidentata* extract exhibited highest inhibitory effects against elastase and hyaluronidase. Meanwhile, The *U. unicinctus* extract exhibited profound inhibitory effect against both tyrosinase and DOPA oxidation, suggesting that the *U. unicinctus* extract might be a potential whitening agent. In summary, the skin aging-related key enzyme inhibitory activity of the marine animal extracts suggests the development potential as promising cosmeceuticals with anti-aging and whitening abilities. Furthermore, studies will be needed to further experiments of cell-based activity, side effects, and identification of the bio-active substances in the extract.

Competing interests

No potential conflict of interest relevant to this article was reported.

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Availability of data and materials

Upon reasonable request, the datasets of this study can be available from the corresponding author.

Ethics approval and consent to participate

This article does not require IRB/IACUC approval because there are no human and animal participants.

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