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JUMBO SQUID (*Dosidicus gigas*) SKIN PIGMENTS: CHEMICAL ANALYSIS AND EVALUATION OF ANTIMICROBIAL AND ANTIMUTAGENIC POTENTIAL

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ABSTRACT

There is a great potential to use seafood by-products to create new beneficial products for customers. In a continued exploration of new chemical compounds from seafood by-products, jumbo squid (*Dosidicus gigas*) skin pigmented methanolic extracts (JSSE) were evaluated for their antimicrobial and antimutagenic activities. Pigments of JSSE were extracted with a yield of 635 mg/g and oxygen radical absorbance capacity-fluorescein (ORAC) with 178 μmol TE/g JSSE using optimal conditions: 25 °C and 5 min of sonication, established by factorial analysis. The antimicrobial activity of JSSE was evaluated using the agar diffusion method. The JSSE showed more than 50% inhibition against *Haemophilys influenza*, *Listeria monocytogenes*, *Staphylococcus aureus*, and *Candida albicans*. The high antimicrobial activity of JSSE (<90%) was detected in *Salmonella enterica*. The JSSE also inhibited mutation induced by aflatoxin B_1 in the *Salmonella tryphimurium* strain TA98 (>50%), but not in the TA100 strain (<20%). Data on the solubility behaviour, the maximum absorbance (440 nm), protons observed in the ^{1}H NMR spectra, and the FT-IR spectra peak at 1742 cm^{-1} of JSSE, suggest that the compound responsible for its antimicrobial and antimutagenic activities comes from the ommochrome family. The present study suggests that squid skin ommochromes are pigments of therapeutic value in near future applications in the food or health sector.

Keywords: antimutagenic activity; antimicrobial activity, extraction optimization; FT-IR, ^{1}H NMR; squid skin ommochromes

INTRODUCTION

Due to the development of infectious diseases and degenerative processes associated with reactive oxygen species, the interest in finding natural compounds that can replace synthetic drugs, and which are safe and wholesome is fuelling one of the fastest expanding fields across several industries. In response to this trend, natural dyes and pigments from several food by-products are being used as food and cosmetic additives, among others things (**Helkar *et al.*, 2016**). Among seafood by-products, jumbo squid skin is a valuable, underutilized source of pigments (**Aubourg *et al.*, 2016**).

Marine organisms develop an extraordinary range of diverse compounds, including pigments with antioxidant, antimicrobial, and antimutagenic activities (**Aquil *et al.*, 2011**). The pigments found in marine organisms, mostly distributed in the fatty tissues of marine fish and invertebrates, are usually synthesized within the tissues of photosynthetic bacteria, algae and higher plants, being the phycobilins, melanins, and carotenoids being the most studied pigments from seafood (**Alasalvar and Taylor, 2002**). Among the compounds responsible for the colour in the cephalopods are ommochromes, which are mainly synthesized in the skin of marine molluscs (**Shamim *et al.*, 2014**). These chromatophores appear as small dots and contain red, yellow or brownish-black pigments. By controlling the size of the cells, they can vary their colour and even create changing patterns. Chromatophores are connected to the nervous system, and their size is determined by muscle contractions (**Deravi *et al.*, 2014).** The metabolic precursor of these pigments is the amino acid tryptophan, from which compounds of varied shades are derived, such as ommatins (low molecular weight, thermolabile and of a faint colour) and ommins (high molecular weight, thermostable and which are related to intense colorations) (**Sahmim *et al.*, 2014**). Ommochromes, like other pigments, produce colour in the biological system, preventing peroxidation in cellular liposomes caused by UV radiation (**Dontsov *et al.*, 1999; Sahmim *et al.*, 2014**) as well as functioning in the tryptophan detoxification process (**Figon and Casas, 2019**). The potential mechanism of action and reactivity of these molecules, established through theoretical studies, could exist by transferring electrons or transferring the hydrogen atom or both, depending on the chemical structure of the ommochrome (**Romero and Martinez, 2015**). In some cephalopod species, like *Loligo vulgaris*, *Seppia officinalis*, *Octopus vulgaris* and, *Doryteuthis pealeii* the main ommochrome identified is xanthommantin (**Bolognese and Scherillo, 1974; Willimas *et al.*, 2016**).

Another species who synthesizes ommochromes is jumbo squid (*Dosidicus gigas*), and they have been mainly found in its skin. Normally, this anatomical region is discarded. The information on jumbo squid skin mostly comprises collagen and its products (**Ezquerra-Brauer and Aubourg, 2019**). Based on the available scientific literature, there is little information about the functional properties of skin ommochromes. Recently, it has been discovered that ommochromes from jumbo squid skin retarded the oxidation of fish oil (**Aubourg *et al.*, 2016**) and prolonged the shelf life of two stored fish species in ice, linked to antioxidant and antimicrobial activities of these extracts (**Ezquerra-Brauer *et al.*, 2016, 2017).** These pigments showed a yellow colour and absorbance peaks in the 300-450 nm region, and had an FT-IR spectrum that showed the presence of functional groups associated with the presence of ommochromes (**Aubourg *et al.*, 2016**).

To explore a novel source of compounds with multiple potentials, the aim of this study was to document the antimicrobial and antimutagenic potential and chemical structure of pigmented compounds extracted from jumbo squid skin. This is the first study of ommochromes' antimicrobial activity against specific strains of bacteria and fungi, as well as their antimutagenic activity. The chemical characteristics of the extracted bioactive pigments was studied by analysing their physical and chemical characteristics. The results of this study provide a more information for the use of jumbo squid skin as another alternative source of bioactive pigments with biological activity.

MATERIAL AND METHODS

Materials

Ten jumbo squids (*D. gigas*) were purchased from a local establishment in Hermosillo, Mexico (29°05′56″n 110°57′15″w), and immediately skinned. The length and weight of the squid specimens ranged from 100 to 150 cm and from 40 to 60 kg, respectively. The skin (about 50 cm length) was frozen at –80 °C, freeze-dried (Labconco, Kansas City, MO, USA) for 2 days and grinded. Samples (100 mg) were put in polyethylene bags, vacuum sealed and kept at –20 °C until analyses. All chemicals used were of analytical reagent grade and purchased from Sigma-Aldrich (St. Louis, MO, USA).

Pigment Extraction

Freeze-dried skin is a mixture of mainly protein and pigments; acidified methanol does not dissolve collagenous and stromal proteins and, at the same time, it is recommended as an ommochrome remover (**Van den Branden and Decleir, 1976**). Therefore, in this work pigment extraction was prepared with acidified methanol. The extraction method consisted of treatments of different temperatures temperature (25, 35, and 45 °C) and sonication times (5, 10, and 15 min). Suitable conditions for obtaining pigmented extracts were established by factorial design in which the dependent variables were yield and antioxidant activity and the independent variables were temperature and sonication time.
Briefly, the pigment extraction process consisted of the homogenization of 20 volumes of freeze-dried skin (w/v) in acidified methanol (99:1 methanol:HCl), followed by centrifugation (Model Biofuge Stratos, Thermo Scientific, Germany) at 10,000 × *g* for 15 min. The methanol was removed using a rotary evaporator (R-100, Büchi, Switzerland) and further evaporated using nitrogen gas. The dry extracts were stored in an inert nitrogen atmosphere, at –80 °C, prior to further analysis. The dried yield was calculated, and stock solutions were prepared to assess antioxidant activity.
Extraction yield was calculated gravimetrically, using the weight of the skin sample as a reference. Pigment yield was calculated as follows:

Pigment yield (%)= [(dried pigmented extract (g))/(dried squid skin (g))]×100.

The antioxidant activity was established by the oxygen radical absorbance capacity (ORAC) method. The ORAC method was carried out according to previous methodology (**Garret *et al.*, 2010**) but with modifications. The fluorescence loss of fluorescein was monitored during 90 min at 37 °c in the presence of 2,2'-azobis(2-amidinopropane) dihydrochloride (AAPH). Each sample (0.5 mg/ml) was tested in triplicate and compared with a standard curve to express results as Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) equivalents.
Extraction conditions for measuring the antimicrobial and antimutagenic activities were selected as the better conditions (temperature and sonication time) for yield and antioxidant activity.

Antimicrobial Activity

The antimicrobial effects of the extracts (10 mg of extract) were assessed following the disc diffusion test as reported previously (**Fatrcová-Šramková *et al.*, 2016**). Antimicrobial activity of the jumbo squid skin pigmented extracts (JSSE) were tested against three Gram-negative bacteria (*Haemophilus influenza* CCM 4456, *Klebsiella pneumoniae* CCM 2318, *Salmonella enterica* subs. enterica CCM 3807), four Gram-positive bacteria (*Bacillus cereus* CCM 2010, *Clostridium perfringens* CCM 4991, *Listeria monocytogenes* CCM 4699, *Staphylococcus aureus* subs. aureus CCM 2461), six microscopic filamentous fungi (*Aspergillus clavatus*, *A. flavus*, *A. versicolor*, *Penicillium chrisogenum*, *P.* griseofulvum, *P. expansusm*) and three yeasts (*Candida albicans* CCM 8186, *C. glabrata* CCM 8270, *C. tropicalis* CCM 8223). Bacteria were collected from the czech collection of microorganisms and microscopic filamentous fungi were collected from the Department of Microbiology, Faculty of Biotechnology and Food Sciences, Slovak University of Agriculture in Nitra, Slovakia. The inhibition of microbial growth was measured around the impregnated discs. Antimicrobial activity is considered high, moderate, or trace/zero when the zone diameter is > 10 mm, 5–10 mm or 2–5 mm, respectively, and negligible effect when the value is less than 2 mm (**Boo *et al.*, 2012**).

Antimutagenic Activity

The Ames test was used to evaluate the antimutagenic activity of the squid skin extracts (**Maron and Ames, 1983**). The assay was performed using 100 µL of *Salmonella typhimurium* strains T98 and T100 grown overnight (1x109 cells/mL), 100 µL of pigment extracts (0.005, 0.05, 0.5, and 5.0 mg/mL) and the mutagenic agent (Aflatoxin B_1, AFB_1) with activation system (500 µL S9 mix) in triplicate plates. Ten precent DMSO (100 µl) without AFB_1 was used as negative control. After incubation for 48 h at 37° C, the number of revertant bacterial per plate were counted. The inhibition rate for mutagenic activity was calculated using the following equation:

Inhibition rate (%)=[((1—T))/M]× 100,

where T is the number of revertants per test sample plate in the presence of AFB1, and M is the number of revertants per plate in the positive control, after subtracting the number of spontaneous revertants from the numerator and denominator. The AFB1 mutagenesis inhibition is considered strong, moderate or weak when the values are high tan 60%, 40–60% or 20–40%, respectively, and negligible when the value is lower than 20% (**Ikke *et al.*, 1999**).

Chemical Structure Analysis

For the analysis the JSSE were freeze-dried and then evaluated.
The solubility test was performed using 5 mL of the following solvents: acetone, ethyl ether, chloroform, 77% aqueous sulfuric acid, and methanol—2% HCl. In each solvent, 5 mg of freeze-dried extracts was dissolved and stirred for 5 min at 24°C (**Van den Branden and Decleir, 1976**).
The absorbance of the extracted pigments was measured using a Cary 50 spectrophotometer (Agilent Technologists, Ciudad de México, México) over the wavelength range of 200–600 nm. The blank solution was methanol.
Fourier transform-infrared spectrum of extracted pigments was obtained from pellets, prepared with 1 mg sample and 100 mg of dry potassium bromide (KBr). The spectra were recorded using and infrared spectrophotometer, Perkin Elmer FT-IR Spectrum GX (Waltham, MA, USA). The FT-IR spectrum (16 scans) was analysed in transmittance mode between 400 and 4000 cm^{-1}.
Then, ^{1}H NMR analysis was measured at 25 °C on a Bruker Avance 400 nuclear magnetic spectrometer (Billerica, MA, USA) operating at 400 MHz. For the experiments, approximately 1 mg of freeze-dried pigments was dissolved in 0.5 ml of a 1 % (v/v) deuterated potassium hydroxide 40% solution with deuterated water. Dimethylsilapentane-5-sulfonic acid (DSS) was used as a reference. The spectral window was 20 ppm.

Statistical Analysis

A 3X3 factorial randomized complete block design was used to obtain an optimal combination of temperature and sonication time that yielded a high level of pigmented extracts with the high antioxidant activity. The selection of the levels of temperature (25, 35 and 45°C) and sonication time (5, 10, and 15 min) tested was based on preliminary studies. The experiment design and statistical analysis were carried out using JMP software (SAS, Cary, NC, USA). Differences between the means were compared using Tukey´s test ($p<0.05$).
Data of the jumbo squid extracted pigment´s physicochemical characterization, antimicrobial and antimutagenic activities were based on the average of three determinations. For spectroscopic analysis, descriptive statistics were used to analyse the data (**Glover and Mitchell, 2015**). For solubility test and antimicrobial and antimutagenic activities, the variations among replicates was <5%. The mean values of the three trials and standard deviations were calculated.

RESULTS AND DISCUSSION

Pigment Extraction

The results of yield and antioxidant activity (Tab 1) indicated that a high yield and high antioxidant activity were obtained when applying a combination of 25°C and 5 min of sonication time. The optimal combination of temperature and sonication time was established by factorial analysis. When the effect of both factors was evaluated, it was observed that the levels of temperature and sonication time affected both positively and negatively the dependent variables ($p < 0.05$). Additionally, an interaction between both factors ($p < 0.05$) was found. The yield of all treatments ranged between 580 and 690 mg of pigment extract per 100 g of fresh squid skin, whereas the antioxidant activity was between 80 and 178 (µmol TE/g). The extraction yield of pigments increased because the sonication time was longer (Tab 1). However, the prolonged exposure of samples to ultrasonic sounds can render antioxidant compounds inactive (Tab 1).

Table 1 Yield and antioxidant activity of pigmented extracts with different temperature-sonication treatments[1] from jumbo squid skin.

Temperature[2] (°C)	**Sonication Time**[3] (min)					
	5		10		15	
	Yield (mg/100 g skin)	**Antioxidant Activity** (μmol TE/g)	**Yield** (mg/100 g skin)	**Antioxidant Activity** (μmol TE/g)	**Yield** (mg/100 g skin)	**Antioxidan Activity** (μmol TE/g)
25	650±5.5^{Bb}	178±2.1^{Aa}	650±5.5^{Bb}	168±3.5^{Ab}	638±6.2^{Ca}	128±2.8^{Ac}
35	659±4.1^{Bb}	150±4.0^{Ba}	659±4.1^{Bb}	135±1.1^{Bb}	685±8.9^{Ba}	115±1.7^{Bc}
45	679±6.1^{Ab}	130±5.0^{Ca}	679±6.1^{Ab}	90±7.5^{Cb}	690±4.1^{Aa}	80±5.0^{Cc}

[1]Values are the mean of three repetitions±standard deviation.
[2]Capital letters in columns denote differences by effect of the temperature ($p < 0.05$).
[3]Small letters in rows denote differences by effect of sonication time ($p < 0.05$).

The two variables used in this study have been previously reported as relevant to the extraction and antioxidant activity of several biological compounds, including pigments (**Maran *et al.*, 2015**; **Belwal *et al.*, 2016; Mokrani and Madani, 2016**). It has been reported extensively that temperatures above 30 °C help with the extraction of biologically active compounds (**Maran *et al.*, 2015**; **Belwal *et al.*, 2016; Mokrani and Madani, 2016**). As can be observed in table 1, for JSSE pigmented extracts, temperature had a significant effect on the extraction yield; however, when temperature increased above 35 °C, antioxidant activity decreased. This type of behaviour has been observed in other studies that dealt with the extraction of antioxidant compounds (**Michiels *et al.*, 2012**). Maintaining 25 °C makes the extraction both cheaper and safer, avoiding the generation of vapours and the usage of heat plates or heat sources. Another advantage is assuring the preservation of the antioxidant activity of the pigmented extract.

The use of sonication in the extraction of compounds has been extensively reported. Sonication facilitated the lysis of the cells in which the pigments are occluded. Its effectiveness in squid skin relies on the formation of vacuum bubbles in the solvent because of low-pressure and high-pressure cycles mediated by the ultrasonic waves. When the bubbles implode, the saccules that contain pigments and other compounds soluble in methanol are released. The mechanical forces eject the compounds, which are later recuperated. Similar patterns to the results obtained in this work, were observed in other foodstuffs from different origins (**Altermimi *et al.*, 2015**). The energy release from sonic waves is not completely efficient; some of it is liberated to the environment and eventually ends up generating free radicals via sonolysis in water and aqueous solutions (**Castellanos *et al.*, 2001**). Evidence has been found of the ultrasound-mediated formation of free radicals in red wine, specifically hydroxyethyl radicals (**Zhang *et al.*, 2015**). It is theorized that the antioxidants exert their function with these molecules, thus resulting in a decrease of functionality (**Zhang *et al.*, 2015**).

The best combinations of temperature and sonication conditions yielded 635 mg/100 g JSS and 178 μmol TE/g JSSE hydrogen atom transfer capacity (ORAC test). Previously it was detected that jumbo squid pigmented extracted with ethanol-acetic acid (**Aubourg *et al.*, 2016**) measured using the ORAC assay showed a value of 15.4 μmol TE/g. Therefore, JSSE contains redox components which are ten times more active than those previously reported. Under these conditions, JSSE pigments were extracted to evaluated their potential antimicrobial and antimutagenic activities.

Antimicrobial Activity

The analysis results of antimicrobial activity of JSSE against selected microbes are shown in table 2. *Haemophilus influenza*, *Salmonella enterica* of Gram–negative bacteria, *Listeria monocytogenes*, *Satphlococcus aureus*, of Gram–positve bacteria, *Aspergillus clavatus*, *Penicillium expanssum*, of fungi, and *Candida albicans* of yeast showed a clear zone formation of growth inhibition. Antimicrobial activity in *Bacillus cereus*, *Klebsiela pneumoniae*, from microscopic fungi *Penicillium chrisogenum* and, from candida *Candida tropicalisscore*d was relative low compared to other strains. The JSSE in the case of *S. enterica* showed the high antimicrobial activity.

The antimicrobial activity detected in JSSE pigments could be due to the amphipathic nature of the ommochromes that gives them the ability to interact with cell membrane components, as well as other bacterial protection factors. At this time a widespread range of natural substances are recognized as having antimicrobial activity, but few studies related to antimicrobial efficacy of squid skin pigments have been done, and some are not made up. Some mechanisms of antibacterial activity, of similar compounds to those reported in the squid skin, are (i) the ability to form pores in cells and (ii) breaking cell walls (**Senan, 2015**). As to the antifungal activity, the main mechanisms recognized for this are attacks on the membrane, microtubules, RNA, and synthesis of ergosterol, among others. However, in the case of the compounds present in the sepia ink, the antifungal activity was related to an imbalance in the redox balance of the fungus (**Fahmy et al., 2014**).

Table 2 Antimicrobial effect of the squid skin extract on bacteria, yeasts, and fungi[1]

Microorganism	**Inhibition zone size** (mm)[2]	**Inhibition** (%)
Bacteria		
Bacillus cereus	T	39.4 ± 0.3
Clostridium perfringens	T	45.5 ± 0.8
Haemophilus influenza	M	54.5 ± 0.4
Klebsiella pneumoniae	T	39.4 ± 0.4
Listeria monocytogenes	M	60.7 ± 0.1
Staphylococcus aureus subs. Aureus	M	57.8 ± 1.3
Salmonella enterica subs. Enterica	H	93.9 ± 0.3
Fungi		
Aspergillus flavus	T	42.4 ± 2.1
Aspergillus versicolor	T	42.4 ± 1.7
Aspergillus clavatus	M	48.4 ± 0.8
Penicillium chrisogenum	T	39.4 ± 3.2
Penicillium griseofulvum	T	42.4 ± 2.4
Penicillium expansum	M	48.5 ± 1.1
Yeast		
Candida albicans	M	66.7 ± 1.5
Candida tropicalis	T	33.3 ± 2.3
Candida glabrata	M	42.4 ± 0.2

[1]**Data:** mean±standard deviation of three repetitions.
[2]**Legend**: H – > than 10 mm, M – > 5–10 mm, T– > 2–5 mm.

Antimutagenic Activity

Antimutagenic activity of squid skin pigments has not been previously reported. Although, the pigments decreased the revertants/plate in a dose-response relationship in both *S. typhimurium* TA98 and TA100 strains (Fig 1), only in TA98 was the percentage of inhibition considered effective, from strong (49–87 %) to moderate (38 %), and a very low inhibition percentage was observed in *S. typhimurium* TA 100 (<14%).

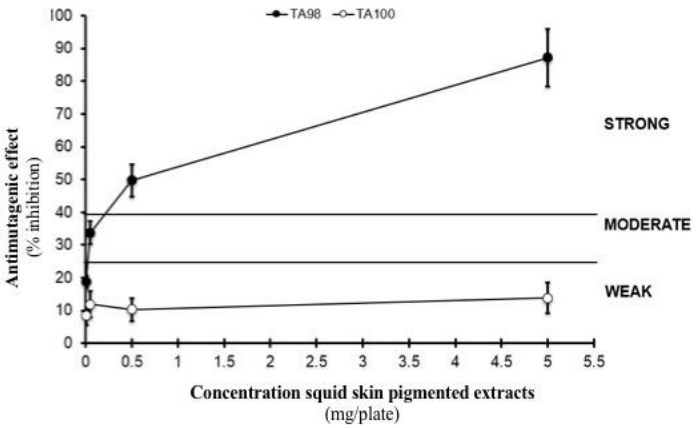

Figure 1 Effect of jumbo squid skin pigments on the mutagenicity induced by aflatoxin B_1, based on *Salmonella typhymurium* TA 98 and TA 100. All values represent mean value of triplicate determination±standard deviation.

The very low inhibition percentage observed in *S. typhimurium* could be due to the complexity of the sample. Therefore, these results suggested that the extracted pigments only protect the genetic material against only one type of mutation, a frameshift mutation detected by TA98 strain, and not a base pair

substitution, because the pigments were not capable of producing at least a moderate inhibition of mutation induced by AFB_1 in TA100 strains (**Jurado *et al.*, 1993**).

It is known that mutations induced by numerous mutagens were reduced by active oxygen scavengers (**Osuna *et al.*, 2016**). Furthermore, it was reported that some antioxidant compounds could prevent mutations because they can induce the synthesis of antioxidant enzymes (**Alasalvar and Taylor, 2002**). In the case of ommochromes, which are the main class of pigments in cephalopods, they have been reported to act as electron accepting or donating systems, as well as tryptophan detoxification products (**Shamim *et al.*, 2014**).

Chemical Structure Analysis

The reddish colour of JSSE suggests that certain types of ommochromes compounds exist in the obtained extract (**Van den Branden and Decleir, 1976**). To corroborate the nature of the components in the JSSE, solubility tests were performed (Tab 3), and the behaviour detected was similar to that expected for ommochrome (**Van den Branden and Decleir, 1976**). Therefore, the JSSE solubility behavior of the compounds present in the obtained extract can be associated with the presence of ommochromes

Table 3 Solubility tests of the squid skin extract[1].

Solvent	Squid Skin Extract[2]	Ommochromes Reported Behavior[3]
Distilled water	NS	NS
Hydrochloric acid 5 M	CS	CS
Acetone	NS	NS
Potassium hydroxide 20%	CS	CS
Acetic acid	PS	PS
Methanol	NS	NS
Acidified methanol	CS	CS
Sulfuric acid 0.25 M	CS	CS
Chloroform	NS	NS

[1] **Data**: all analyses were run in triplicate.
[2] **Legend**: NS — no solubility, PS — poor solubility, CS— complete solubility.
[3] **Reference:** Van den Branden and Decleir (1976).

To confirm whether the pigments extracted from jumbo squid skin contained ommochromes UV-Vis, FT-IR, and ^{1}H NMR spectroscopies were employed. The UV-Vis spectroscopy of extracted pigments had an absorption maximum of 440 nm (Fig 2), which is similar to those red-pigments compounds previously reported in squid *D. pealeii* (**Williams *et al.*, 2016**). Ommochromes are usually distinguished by their specific absorbance spectra; this characteristic implied that the squid pigments contain ommins, one of the two groups of ommochromes (**Shamim *et al.*, 2014**). Moreover, the ^{1}H NMR spectrum (Fig 3) was similar to those of ommins (**Kumar *et al.*, 2018**). The ^{1}H NMR spectrum indicated aromatic protons at δ 7.4 ppm (singlet) and at 7.2 ppm (singlet) and, functional group adjacent to a methyl carbon at δ 3.8 (triplet) and at 3.0 ppm (multiplet).

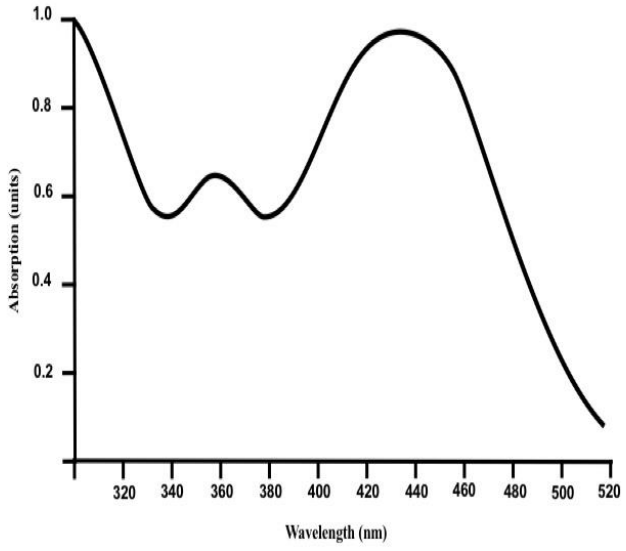

Figure 2 UV-Vis spectrophotometric spectra of jumbo squid skin pigments.

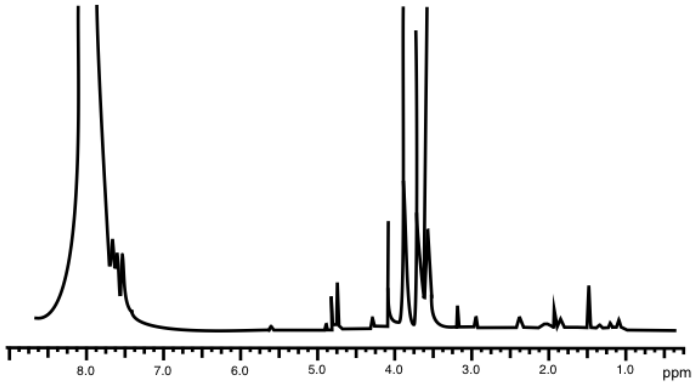

Figure 3 ^{1}H NMR spectrophotometric spectra of jumbo squid skin pigments.

Infrared spectroscopy provides more information regarding the chemical composition and conformation of the obtained pigments. The FT-IR spectra (4000–400 cm^{-1}) of the pigments (Fig 4) represented those reported for ommochromes (**Bolognese and Scherillo, 1974**). The main signals observed were at 3550–3100 cm^{-1} (N–H), 3000–2700 cm^{-1}(C–H stretching vibrations), 1500–1425 cm^{-1} (N–H and C–H bending vibrations), 1240–1050 cm^{-1} (C–O and C–N stretching vibrations) (**Dyer, 1965**). Furthermore, wave numbers for carbomethoxy C=O (1740 cm-1) and quinonic C=O (1670 cm^{-1}) indicated that squid pigments contained ommochromes compounds of the xanthommatin-type (**Bolognese and Scherillo, 1974**).

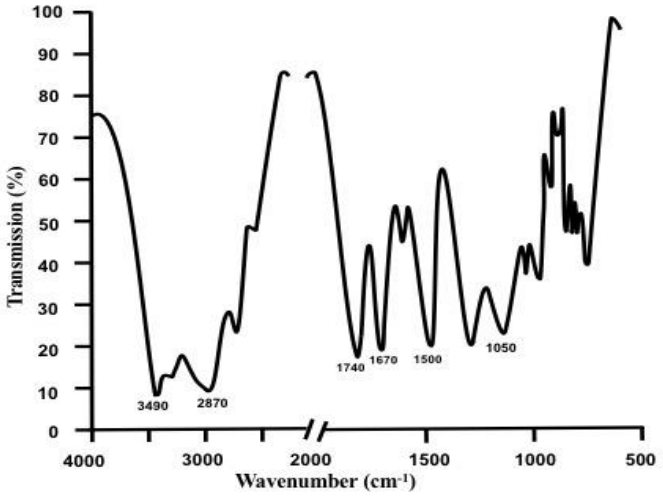

Figure 4 FTIR spectrophotometric spectra of jumbo squid skin pigments.

CONCLUSION

Jumbo squid skin pigments contain antibacterial and antimutagenic compounds, which were detected in the methanol–HCl soluble extracts. The extraction of bioactive pigments from jumbo squid skins was determined by both temperature and sonication time. Additionally, the present study suggests that one of the main compounds that exerted the biological activity in squid skin pigmented extracts were ommatins, specifically of the xanthommatin type. However, future studies need to focus on the identification of the specific antimicrobial and antimutagenic mechanisms of the compounds present in the jumbo squid skin pigmented extract.

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Conflicts of Interest: All authors declare that there are no conflicts of interest regarding the publication of this paper.

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