Chinese Chemical Letters 26 (2015) 215–220

Contents lists available at ScienceDirect

Chinese Chemical Letters

journal homepage: www.elsevier.com/locate/cclet

Original article

Analysis and discrimination of ten different sponges by multi-step infrared spectroscopy

Jian-Hong Gan [a,e,1], Chang-Hua Xu [a,1], Hong-Zhe Zhu [d], Fang Mao [a], Fan Yang [b,*], Qun Zhou [c], Su-Qin Sun [c,*]

[a] College of Food Science & Technology, Shanghai Ocean University, Shanghai 201306, China
[b] Key Laboratory for Marine Drugs, Department of Pharmacy, State Key Laboratory of Oncogenes and Related Genes, Ren Ji Hospital, School of Medicine, Shanghai Jiao Tong University, Shanghai 200127, China
[c] Department of Chemistry, Tsinghua University, Beijing 100084, China
[d] Department of Pharmacy, Taian Maternal and Child Health Hospital, Jiangsu 271000, China
[e] Department of Pharmacy, Changzheng Hospital, Second Military Medical University, Shanghai 200433, China

ARTICLE INFO

Article history:
Received 30 September 2014
Received in revised form 10 December 2014
Accepted 22 December 2014
Available online 21 January 2015

Keywords:
Sponge
Discrimination
Infrared spectroscopy
Second derivative infrared spectroscopy
Two-dimensional correlation infrared spectroscopy

ABSTRACT

In this study, a convenient method using multi-step infrared spectroscopy, including Fourier transform infrared spectroscopy (FT-IR), second derivative infrared spectroscopy (SD-IR) and two-dimensional correlation infrared spectroscopy (2DCOS-IR), was employed to analyze and discriminate ten marine sponges from two classes collected from the Xisha Islands in the South China Sea. Each sponge had an exclusive macroscopic fingerprint. From the IR spectra, it was noted that the main ingredient of calcareous sponges was calcium carbonate, but that of demosponges was proteins. For sponges from the same genus or having highly similar chemical profile (IR spectral profile), SD-IR and 2DCOS-IR were applied to successfully reveal the tiny differences. It was demonstrated that the multi-step infrared spectroscopy was a feasible and objective approach for marine sponge identification.

1. Introduction

Marine sponges are classified into three classes: Calcarea, Hexactinellida and Demospongiae containing 25 orders, 127 families and 790 genera. There are approximately 15,000 species reported worldwide [1–4]. Almost 1/3 marine natural products come from sponges [3,5]. In all the prevalent traditional ways to identify sponges, the most accurate and commonly used method is spicule identification, which examines sponge skeletal structure. This method routinely requires a whole piece of sponge and heavily depends on experts mastering with English, French and German and ages of experience [6]. Now the number of experts in the art of sponge identification is greatly reduced. Hence, it is necessary to establish more objective and simpler methods to discriminate marine sponges.

"Multi-step infrared spectroscopy" including FT-IR, SD-IR and 2DCOS-IR has the above advantages for sponge identification. FT-IR has been proved to be a quick, simple and effective method with good signal-to-noise ratio and excellent repeatability to investigate complicated mixtures such as herbal medicine (HM) [7,8]. To delineate the overlapped spectra in FT-IR, second derivative infrared spectroscopy (SD-IR) is used to improve the apparent resolution [9]. If the differences are still too small to discriminate, two-dimensional correlation infrared spectroscopy (2DCOS-IR) can be adopted to illustrate FT-IR spectra in a second dimension to reveal the differences more convincingly [10,11].

Ten different sponges collected from Xisha Islands in South China Sea of two classes, five orders and ten species have been studied using the multi-step infrared spectroscopy to establish a new method to discriminate sponges in a more objective, direct and quicker manner.

2. Experimental

2.1. Apparatus

A spectrum GX FT-IR spectrometer (PerkinElmer, UK) equipped with a DTGS detector and a scanning range from 400 to 4000 cm^{-1}

* Corresponding authors.
E-mail addresses: bill1985@126.com (F. Yang), sunsq@tsinghua.edu.cn (S.-Q. Sun).
[1] Author contributions: Jian-hong Gan and Chang-hua Xu had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. These authors contributed equally to this study.

http://dx.doi.org/10.1016/j.cclet.2015.01.012

Table 1
Ten China sponges collected from Xisha Islands of South China Sea.

Class	Order	Family	Genus	Specie	Abbreviation	Photo
Calcarea	Leucosolenida	Grantiidae	*Leucandra*	sp	LA	
		Leucosolenidae	*Leuconia*	sp	LO	
Demospongiae	Poecilosclerida	Mycalidae	*Mycale*	*Fibrexilis*	MY	
	Dietyoceratida	Thorectidae	*Hyrtios*	sp	HY	
		Spongiidae	*Hippospongia*	*Lachna*	HI	
			Aplysinopsis	sp	AP	
	Halichondrida	Halichondridae	*Halichondria*	sp	HA	
		Axinelliedae	*Phakellia*	*Fusca Thiele*	PA	
			Acanthella	sp1	AC1	
				sp2	AC2	

with a 4 cm^{-1} resolution was used. Spectra were calculated from a total of 32 scans at 0.2 cm/s of OPD speed. The interferences of H_2O and CO_2 were eliminated during scanning. A CKW-II programmable temperature controller (Beijing Chaoyang Automatic Instrument Co., China) was employed to perform the thermal perturbation from the range 50 °C to 120 °C. Spectra were collected at 10 °C intervals. The second derivative IR spectra were gained after 13-point smoothing of the IR spectra by Savitzky–Golay polynomial fitting. Two-dimensional IR correlation spectra were obtained after using 2D correlation analysis software (developed by IR Lab, Tsinghua University) to analyze the series of thermo perturbation dynamic spectra.

2.2. Samples and reagents

Sponges (Table 1) were obtained from Xisha Islands in South China Sea in April 2007 and 2009 (kept in Department of Pharmacy, Changzheng Hospital, Second Military Medical University) and were authenticated by Prof. Jinhe Li, Qingdao Institute of Oceanology, Chinese Academy of Sciences. KBr was bought from Sigma (St. Louis, MO, USA).

2.3. Procedure

Sponge samples were desiccated in vacuo and then were grounded into powder before IR measurement. Each sponge sample (about 1–2 mg) was mixed with KBr (100 mg), grounded into powder (200 mesh), and then pressed into a tablet.

3. Results and discussion

3.1. Analysis and discrimination of five sponges from two classes

Fig. 1 shows the IR spectra of five sponges belonging to two classes. In these five sponges, *Leucandra* sp. is a calcareous sponge and is collected for the first time in China. The main composition of