ANTIPROLIFERATIVE AND ANTI-INFLAMMATORY ACTIVITY OF PREGNANE STEROID ISOLATED FROM THE ADRIATIC SEA CORAL, *Eunicella cavolini*

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INTRODUCTION

Gorgonian corals of the genus *Eunicella* are well-known sources of structurally diverse secondary metabolites including terpenoids, steroids as well as alkaloids to lower extent. Detailed research studies of those compounds are scarce and they have been examined mostly as anticancer agents on several cell lines [1]. On the other side, steroids (i.e. glucocorticoids) have been widely used as anti-inflammatory agents, often exerting serious side effects [2]. Therefore, there is a constant need for the discovery of new compounds that will show better and more effective activity in the treatment of inflammatory processes. Within the Adriatic Sea bioprospecting programme, we have carried out the extraction and isolation of a pregnane steroid (**1**) from the Adriatic Sea coral, Eunicella cavolini (Koch, 1887) which was evaluated for its antiproliferative and anti-inflammatory activities.

MATERIALS AND METHODS

The yellow gorgonian, *Eunicella cavolini*, was collected near the Island of Pag (Croatia) and kept at −80 °C. A dark red-orange oily organic extract (6.81 g) was obtained by maceration of the organism by using dichloromethane and methanol (3:1). Further, the extract was purified by several column chromatographies over silica gel by using cyclo-hexane and ethyl acetate. Finally, subfractions of interest were combined and purified over preparative TLC to obtain **1** (41.7 mg). Dereplication of **1** was performed by using spectroscopical dana and MarinLit database agreeing with the already published data [3]. Compound **1** was examined for antiproliferative activity applying MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide) assay. Anti-inflammatory activity was investigated by using two *in vitro* assays: inhibition of albumin denaturation and inhibition of soybean lipoxygenase.

RESULTS

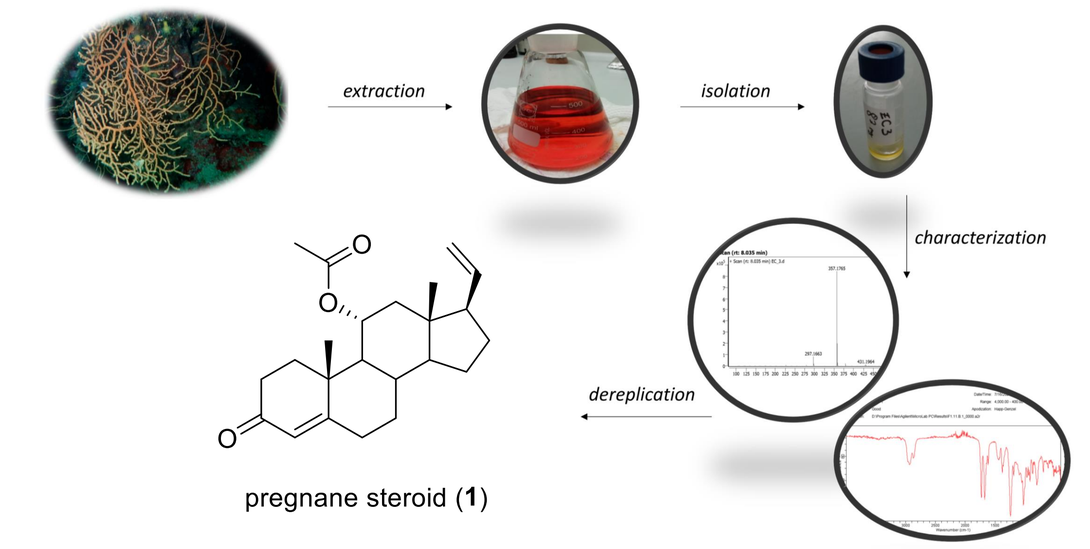

Figure 1 Workflow of pregnane steroid **(1)** isolation from the Adriatic sea octocoral *E. cavolini.*

Table 1 **Antiproliferative activity** of 1 and doxorubicin tested against four human cancer cell lines: H460, SW620, HepG2 and MCF-7 and embryonic kidney cell line (HEK293T)*.

Sample	IC_{50} (μM)				
	H460	SW620	HepG2	MCF-7	HEK293T
1	18 ± 2	18 ± 6	6 ± 1	13 ± 4	12 ± 2
Doxorubicin	0.01 ± 0.003	0.01 ± 0.003	0.03 ± 0.001	0.01 ± 0.002	0.01 ± 0.003

* *H460 (lung carcinoma, large cell lung cancer (ATCC®HTB-177™), MCF-7 (breast adenocarcinoma, ATCC® HTB-22™), SW 620 (colorectal carcinoma ATCC® CCL-227™), HepG2 (hepatocellular carcinoma ATCC® HB-8065™) and HEK293T (embryonic kidney ATCC® CRL-3216™).*

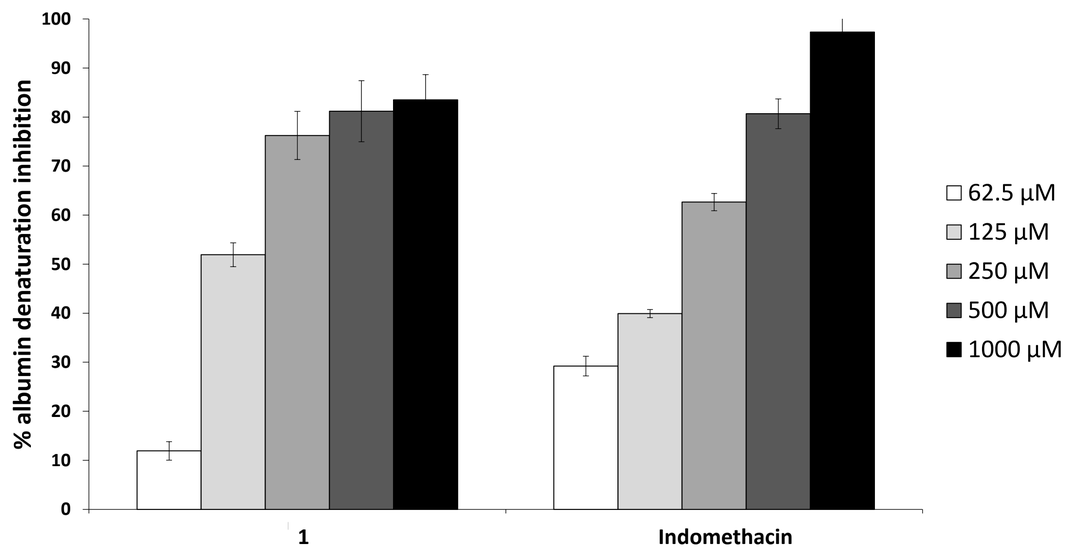

Figure 2 Effects of pregnane steroid **1** and indomethacin (positive control) on **albumin denaturation** tested in concentration range: 62.5 – 1000 μM. Data are presented as a mean ± SD of triplicate measurements.

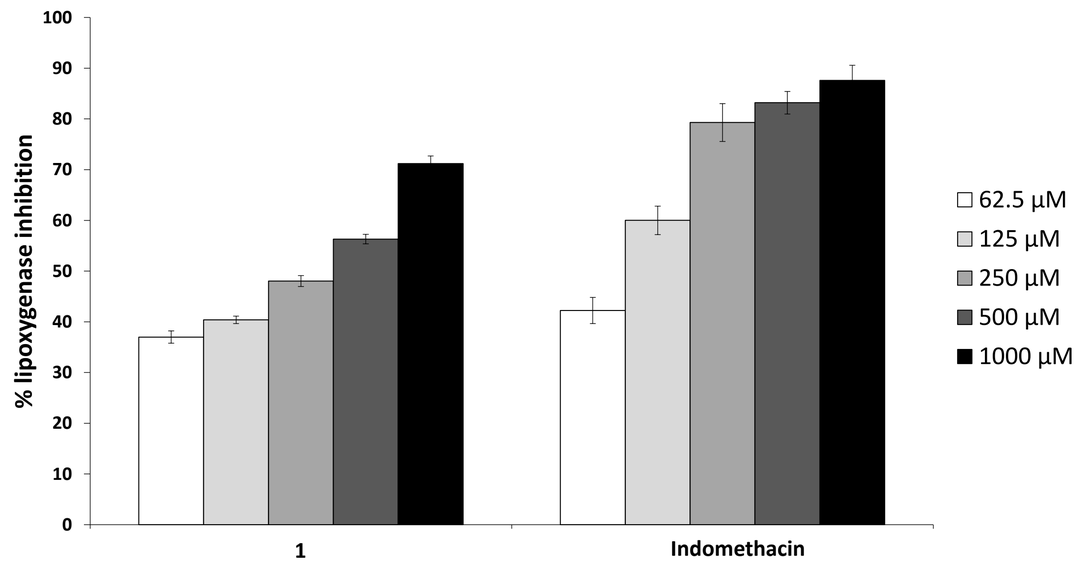

Figure 3 **Lipoxygenase inhibitory activity** of pregnane steroid **1** and indomethacin (positive control) tested in concentration range: 62.5 – 1000 μM. Data are presented as a mean ± SD of triplicate measurements.

CONCLUSIONS

Pregnane steroid **(1)** exhibited moderate, but non-selective antiproliferative effect with the highest activity against hepatocellular carcinoma cells (IC_{50} = 6 ± 1 μM).

1 showed promising anti-inflammatory activities displaying inhibtory potential against albumin denaturation (IC_{50} = 119 ± 3 μM) and lipoxygenase activity (IC_{50} = 309 ± 12 μM).

Hydrolysis of **1** and evaluation of contribution of acetate group is currently ongoing.

REFERENCES

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