

Chemical Ecological Studies Of Portuguese Invertebrates

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Chemical Ecology

Interdisciplinary

Chemistry
Biology

Ecological Mediators



Natural Products



Biosynthesis

Biological
Activity

Symbiosponge - Biology of Sponge Natural Products

1998-2001 Mas

University of Amsterdam, Rob Van Soest

INETI, Regina Tavares

Université Libre de Bruxelles, Jean-Claude Braekman

- o Chemical markers of sponges
- o Ecological Studies in sponges
- o Spatial distribution of SEC MET (sponge cells or symbionts)
- o Production of bioactive compounds by cell culture
- o Screen of antimicrobial activities from sponges

"Cellular origin and ecological role of oroidin and sceptrin in the Caribbean sponge *Agelas conifera*"

Biochem. Syst. Ecol., **2003**, 1073-1091

Are Portuguese Marine organisms a promising source of Natural Products?



Must be studied!

Comparative Study on Chemical Ecology of the Atlantic Ocean and Mediterranean Sea Invertebrates

Portugal



Italy

INETI, Lisboa
Helena Gaspar

IPM, Faro
Gonçalo Calado

ICB-CNR, Napoly
Ernesto Mollo
Margherita Gavagnin
Angelo Fontana
Adele Cutignano
Guido Cimino

Target Portuguese organisms

Phylum Mollusca

Class Gastropoda

Subclass Opisthobranchia

Phylum Porifera

ORDERS

Cephalaspidea

Acochlidea

Notaspidea

Rhodopemorpha

Gymnosomata

Thecosomata

Sacoglossa

Anaspidea

Nudibranchia

Sampling Stations

1- Ria de Aveiro



2- Reserva da Berlenga



3- Reserva da Arrábida



4- Ria Formosa



Target Sampling Station for opisthobranchs Reserva da Arrábida



Setúbal-Cabo Espichel Marine Coast
about 100 species of opisthobranchia

Target species from Arrábida

Order Nudibranchia

Family Dendroridae

Doriopsilla pelseneeri
(d' Oliveira, 1895)



Doriopsilla areolata
(Bergh, 1880)



Previous studies on *Doriopsilla*



Pacific

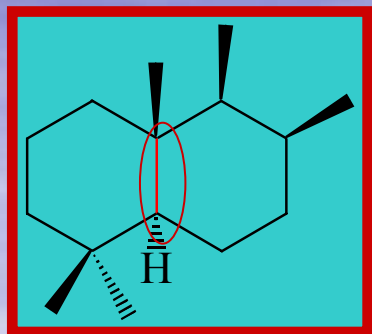


Mediterranean, Atlantic

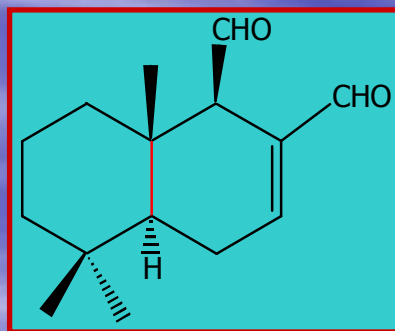


Pacific

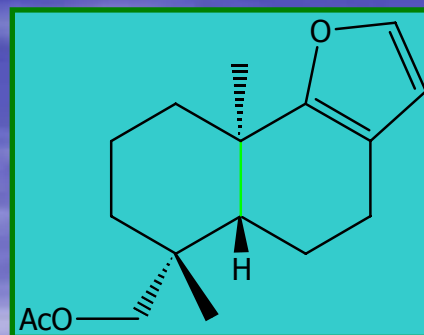
Okuda 1983 JOC 1866-1869
Spinella 1994 Tetrahedron 8665-8668
Gavagnin 2001 Chemoecology 131-136
Gavagnin 2001 Tetrahedron 8913-8916
Fontana 2003 JOC 2405-2409



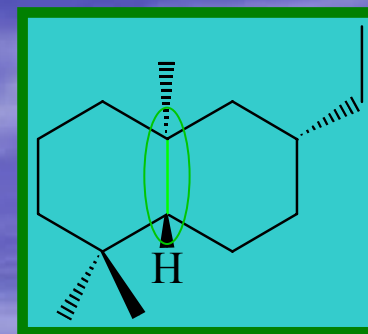
Drimane skeleton



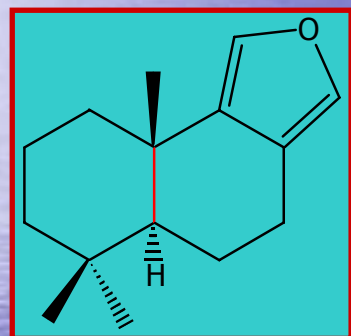
polygodial



15-acetoxy-*ent*-pallescensin-A

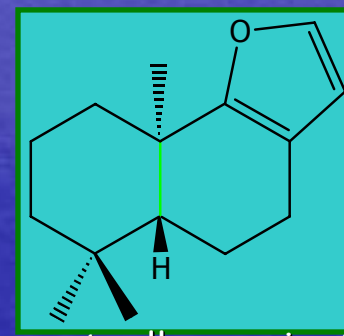


ent-pallescensin skeleton

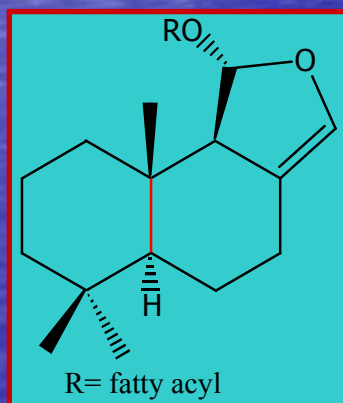


eurifurane

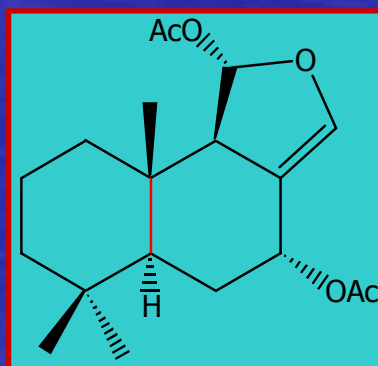
Doriopsilla



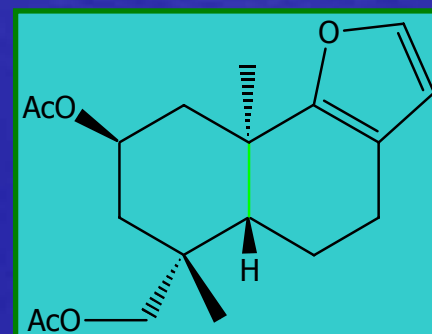
ent-pallescensin-A



Drimane esters



olepupane

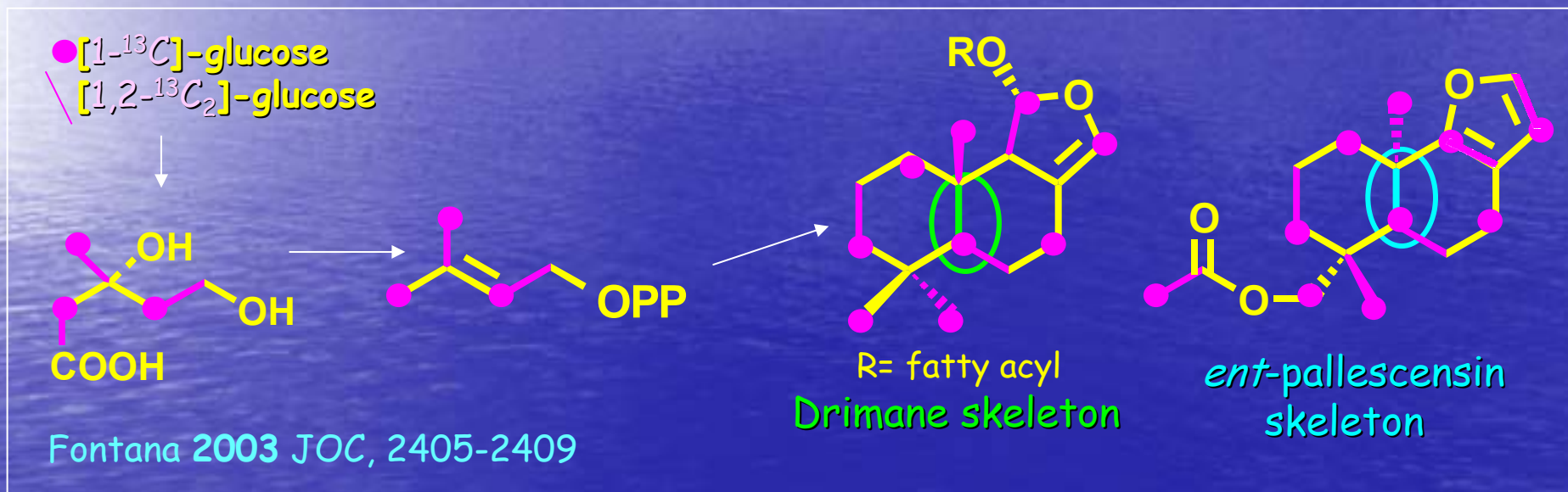


2,15-diacetoxy-*ent*-pallescensin-A

Previous Biosynthetic studies in *Doriopsilla*

Feeding experiments with (2- ^{14}C)-mevalonate proved that *D. areolata* produce *de novo* via mevalonate both ent-pallescensin-like and drimane metabolites

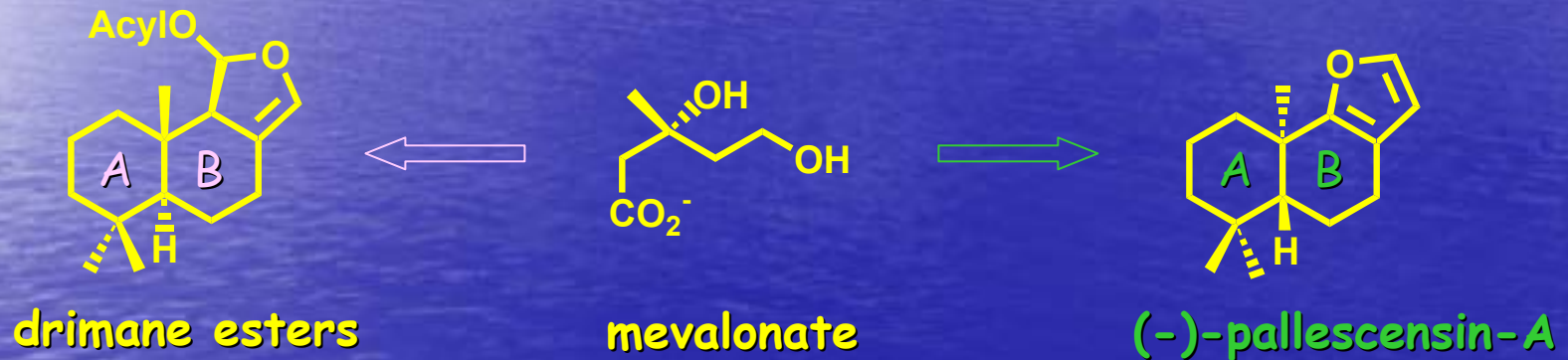
Gavagnin 2001
Tetrahedron 8913-8916



Feeding experiments with glucose (1- ^{13}C) and (1,2- $^{13}\text{C}_2$) allow to determine the sites of incorporation for both skeletons

Why *doripsilla* has acquired the ability to produce de novo the same metabolites as its preys?

biosynthesizes typical sponge sesquiterpenoids with opposite A/B ring junction



lateral gene transfer (Faulkner)

retrosynthetic mode (Ghiselin)

Why studied *Doriopsilla pelseneeri*?

Do they have drimane and *en-pallesensin*-like compounds as defensive metabolites?

If yes, do they biosynthesise them *de novo*?



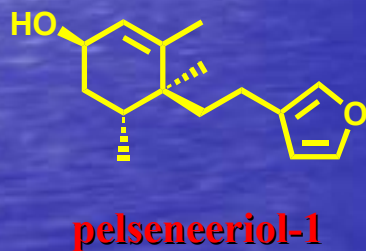
Doriopsilla pelseneeri



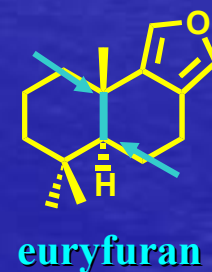
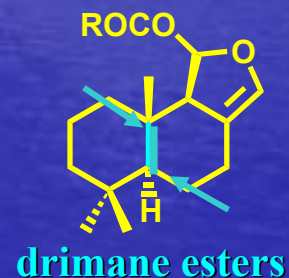
Mantle and mucus



New compounds



Internal glands



Target species from the Rias

Ria de Aveiro



Ria Formosa



Similar habitats
sandy-mud bottoms
algae and seagrasses

Class Gastropoda

Subclass Opisthobranchia

Order **Cephalaspidea**

Family Haminoeidae

Haminoea orbignyana
(Férussac, 1822)



Family Bullidae

Bulla striata
(Bruguière, 1792)

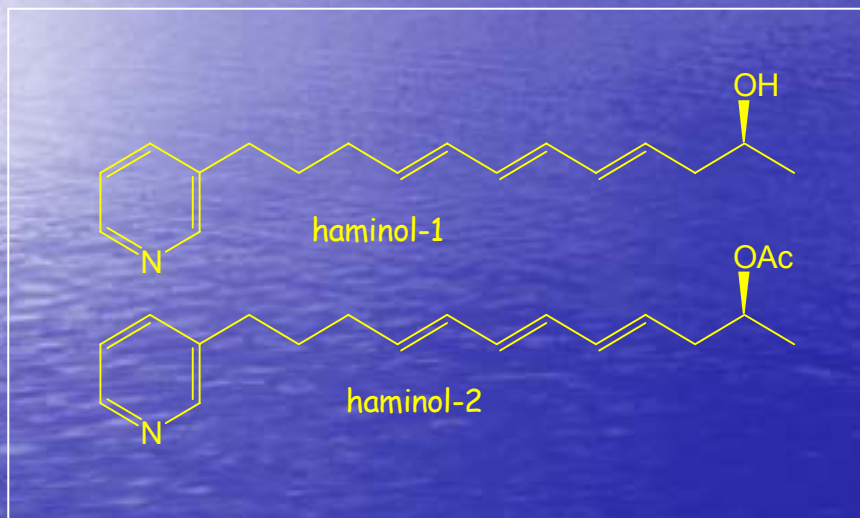


300 *Haminoea* m² !

Haminoea orbignyana



Portuguese populations



Haminoea synthesis *de novo*
through a polyketide pathway using
nicotinic acid as the starter unit

Cutignano 2003, Angew. Chem. Int. Ed, 2633
Cutignano 2004, TL 2627-2629

Mediterranean Population
Spinella 1993, Tetrahedron, 1307-1314

Haminols work as Alarm pheromones

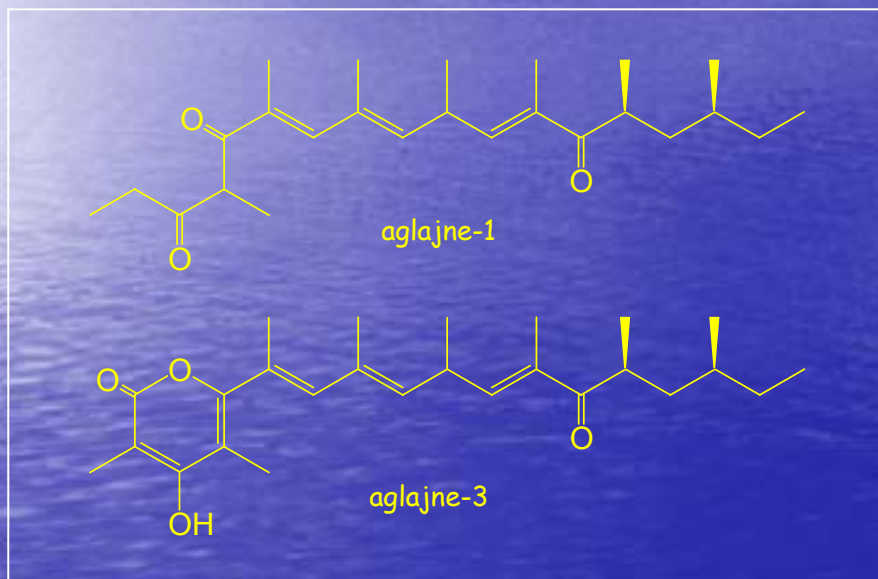
Cimino *et al.* 1991, *Experientia*, 47, 61



Bulla striata



Portuguese population



Mediterranean Population
Cimino 1987, JOC, 5326-5331

Bulla striata synthesised
de novo polypropionate

Fontana 2004, TL 6847-6850

Conclusion

- This was the first chemical study of the nudibranch *Doriopsilla pelseneeri*
- Simillary with other *Doriposilla*, *D. pelseneeri* contain sesquiterpenes related to typical sponges metabolites.
- *D. pelseneeri* is able to produce *de novo* the drimane skeleton
- The skeleton of the new pelseneriols also agrees with the reported proposed biogenesis for *D. areolata* => generic for *doriopsilla* genus
- *Haminoea orbignyana* from different portuguese populations contains the same alarm pheromones, haminol-1 and haminol-2, than the mediterranean population
- *Bulla striata* from a portuguese population contains the same polypropionates, *aglajne-1* and *aglajne-3* than the mediterranean population

Further studies

Perform biosynthetic studies
in *Bulla striata* and *Haminoea orbignyana*

Perform experiences with *Doriopsilla* in order to determine the
biosynthetic pathway for both skeletons

Isolation of the terpene cyclase from *Doriopsilla* since it could
be a promising tool for metabolic engineering

Test pure compounds for several biological activities

in vitro Biological Activities

For pure compounds or extracts

- **Anticancer**

University Porto (Prof M. Nascimento) Portugal

- **Antiprotozoal**

Univesyty Madrid (Prof R. Martinez-Diaz) Spain

- **Antifeedant/Allelopathic/Cytotoxicity**

CSIC Madrid (Dra. A. González-Coloma) Spain

- **Antituberculosis**

Refree Center of Tuberculosis (Dra. M. C. Lourenço) Brasil

- **Antimicrobial**

INETI-LMI (Dra. S. Feio) Portugal

University Algarve (Prof. M. L. Faleiro) Portugal

GRICES

GABINETE DE RELAÇÕES INTERNACIONAIS DA CIÊNCIA E DO ENSINO SUPERIOR

Ministério da Ciência, Tecnologia e Ensino Superior



FUNDAÇÃO CALOUSTE GULBENKIAN



Istituto di
Chimica
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