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Design and molecular docking studies of new potential PKC-δ activators based on royleanone scaffold

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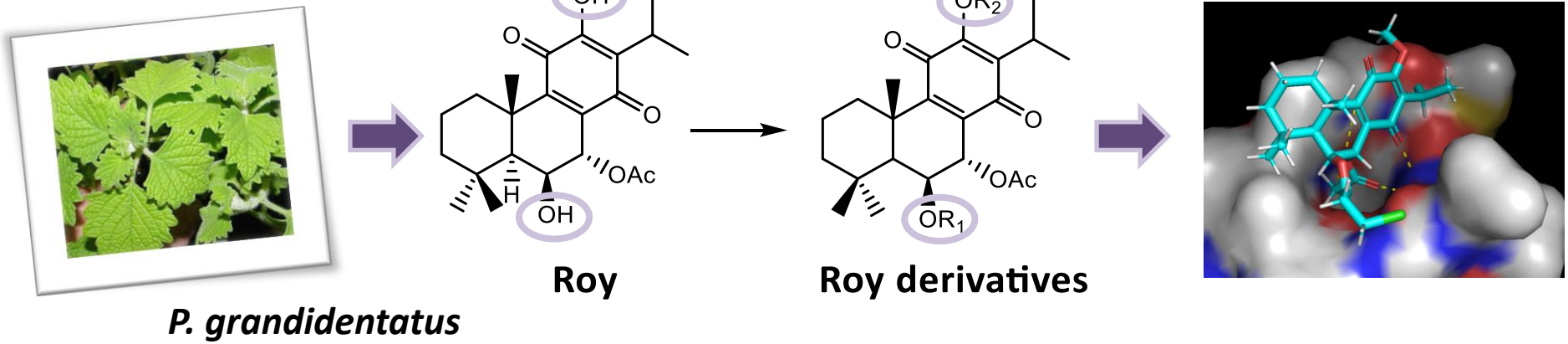
Design and molecular docking studies of new potential PKC-δ activators based on royleanone scaffold

Graphical Abstract



Abstract

The huge impact of cancer is a big concern nowadays. Protein Kinases (PKCs) are attractive anticancer targets due to their involvement in several processes of carcinogenesis. Particularly, the isoform δ (PKC-δ) acts as tumor suppressor in colon cancer, one of the most dominant cancers and cause of cancer mortality worldwide.

Promising bioactive molecules were found in *Plectranthus* genus, mainly diterpene royleanones. The 7α-acetoxy-6β-hydroxyroyleanone (Roy) is the major constituent of *P. grandidentatus*. Several biological activities of Roy were reported in the literature, including antitumoral activity. Moreover, the presence of two free hydroxyl groups in Roy structure drawn our attention to the possibility of preparing new derivatives with enhanced cytotoxic activity. In fact, in a previous work, the patented diterpene 6β-benzoyloxy-12-O-benzoylroyleanone (RoyBz) shown selective activation of PKC-δ.

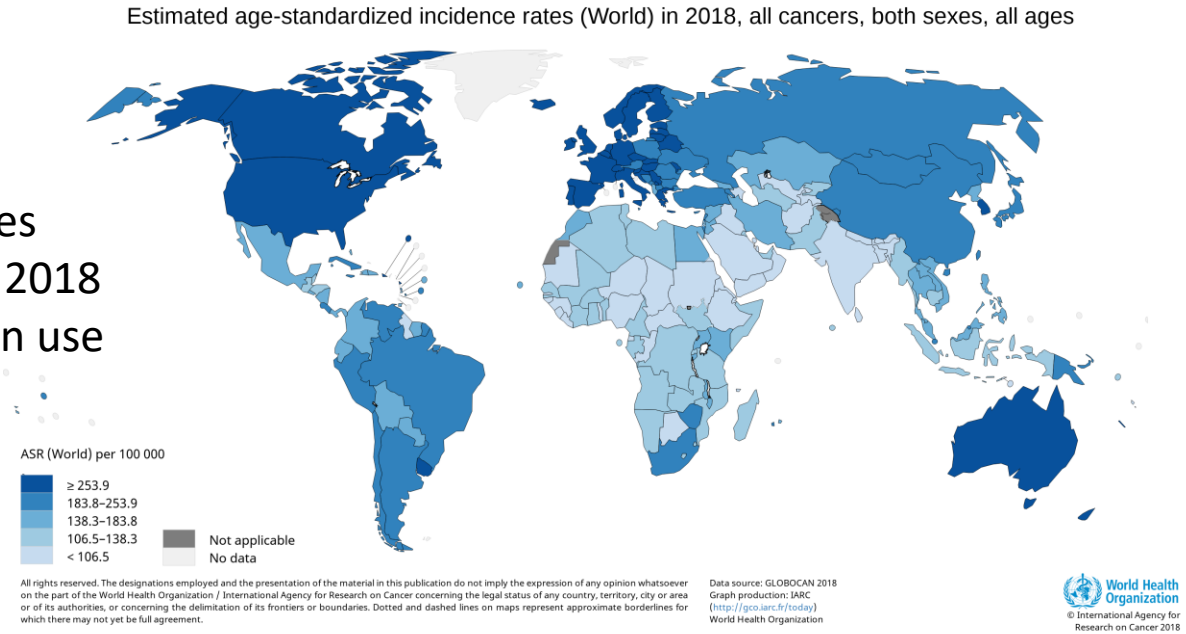
The aim of the present work is to prepare new potential PKC-δ activators from derivatization of Roy. Thus, Roy and RoyBz assisted the design of theoretical derivatives, through modification of the hydroxyl groups. Molecular docking simulations were carried out against the 3D structure of human PKC-δ regulatory domain, to identify the potential PKC-δ activators. The most promising compounds accepted by docking simulations are currently been prepared by hemi-synthesis using Roy as starting material for structure-activity relationships.

Keywords: *Plectranthus*; royleanones, derivatives, PKC- δ, molecular docking

Introduction

CANCER – A GLOBAL CONCERN

- 18.1 million new cancer cases
- 9.6 million cancer deaths in 2018
- Resistance to clinical drugs in use



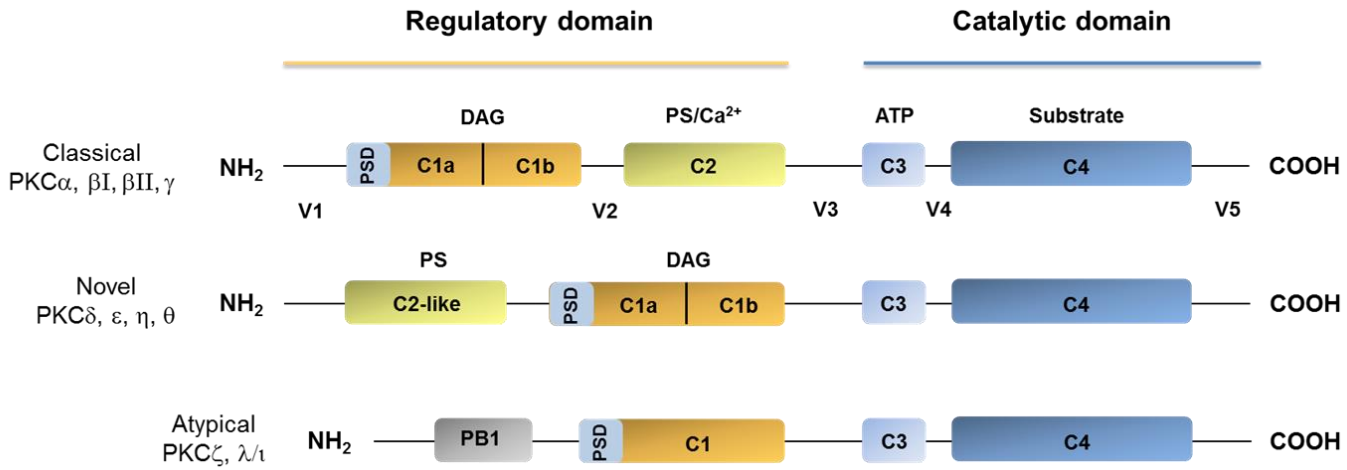
Need for new and more effective anticancer drugs

Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. CA. Cancer J. Clin., 68(6), 394–424 (2018).

Introduction

PROTEIN KINASE C (PKC) – Attractive anticancer targets

PKC Isozymes divided in Classical, Novel and Atypical



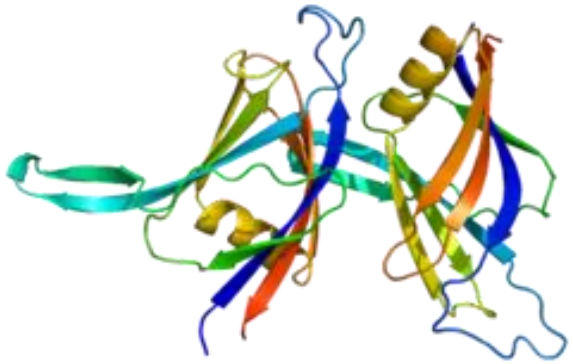
Associated with proliferation, migration, invasion, tumorigenesis, and metastasis

D. Matias, C. Bessa, M.F. Simões, C.P. Reis, L. Saraiva, P. Rijo, Natural Products as Lead Protein Kinase C Modulators for Cancer Therapy, in: Atta-ur-Rahman (Ed.), Studies in Natural Products Chemistry, 2016, pp. 45–79.

Introduction

PROTEIN KINASE C-δ (PKC-δ)

- Associated with pro-apoptotic functions
- Death mediator of chemotherapeutic agents and radiotherapy
- Associated with proliferation of colon cancer cells



The selectivity through PKC isoforms limits the use of anticancer drugs

D. Matias, C. Bessa, M.F. Simões, C.P. Reis, L. Saraiva, P. Rijo, Natural Products as Lead Protein Kinase C Modulators for Cancer Therapy, in: Atta-ur-Rahman (Ed.), Studies in Natural Products Chemistry, 2016, pp. 45–79.

Introduction

Medicinal plants are a great source of new drug leads

Mint

- **Family *Lamiaceae* –** Same family of Lavender, oregano, basil, rosemary, mint and other widely used and well-known plants

Oregano

Rosemary

Basil

  - ***Plectranthus* genus –** Plants obtained from South Africa and cultured in Portugal (Instituto Superior de Agronomia de Lisboa)




***Plectranthus* spp.**

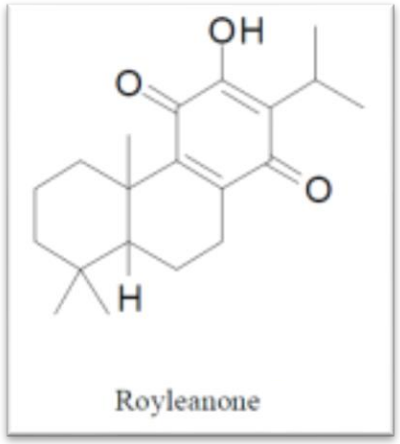
Ladeiras D, Monteiro CM, Pereira F, Reis CP, Afonso CAM , Rijo P. Curr. Pharm. Des., 22(12), 1682–1714 (2016).
Garcia C, Teodósio C, Oliveira C, Oliveira C, Díaz-Lanza A, Reis C, Duarte N, Rijo, P. Curr. Pharm. Des., 24(36), 4207-4236 (2019).

Introduction

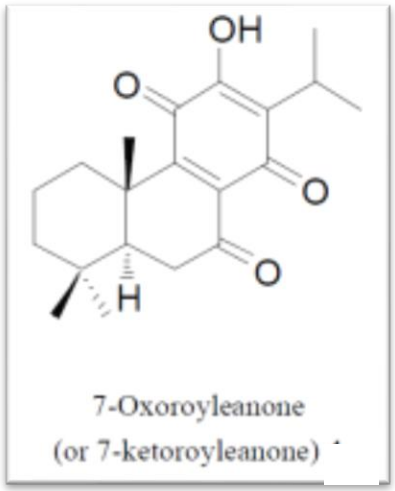
***Plectranthus* spp. widely use in traditional medicine**

Source of bioactive compounds

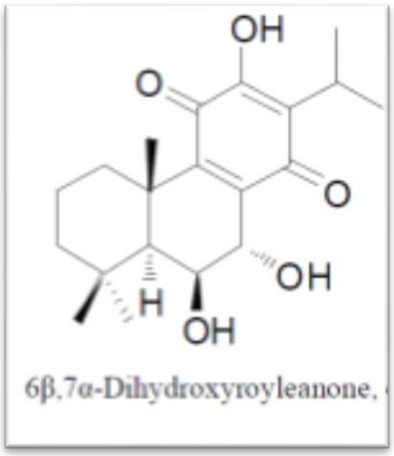
- **Diterpene quinones**
- **Royleanones**



Royleanone



7-Oxoroyleanone
(or 7-ketoroyleanone)



6β,7α-Dihydroxyroyleanone,

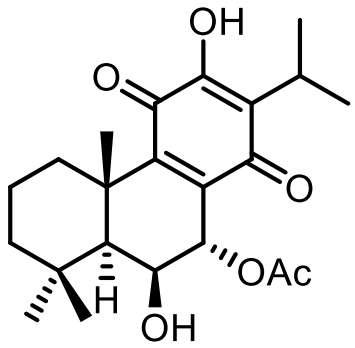
Ladeiras D, M. Monteiro C, Pereira F, P. Reis C, A. M. Afonso C, Rijo P. Curr. Pharm. Des., 22(12), 1682–1714 (2016).

Introduction

7α-acetoxy-6β-hydroxyroyleanone (Roy) - Cytotoxic Royleanone

P. grandidentatus

Extraction CO_2 Supercritical



Roy

- Ability to **evade** the activity of **P-gp**
- *In vitro* antiproliferative activity against several cancer cell line

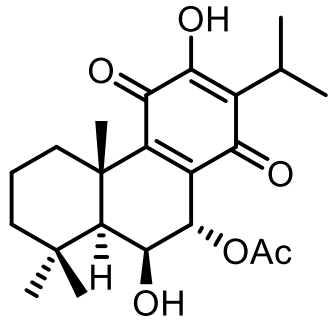
Bernardes CES, Garcia C, Pereira F, Mota J, Pereira P, Cebola MJ, Reis CP, Fátima MM, Piedade ME, Rijo p. Molecular pharmaceutics, 5(4), 1412-1419 (2018).

Introduction

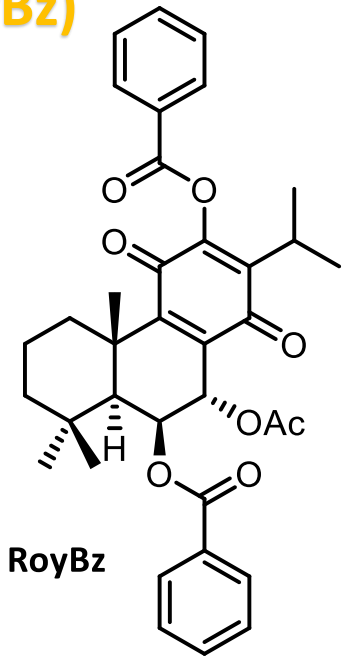
7α-acetoxy-6β,12-dibenzoylroyleanone (RoyBz)

- PKCδ-selective activator

P. grandidentatus



Roy



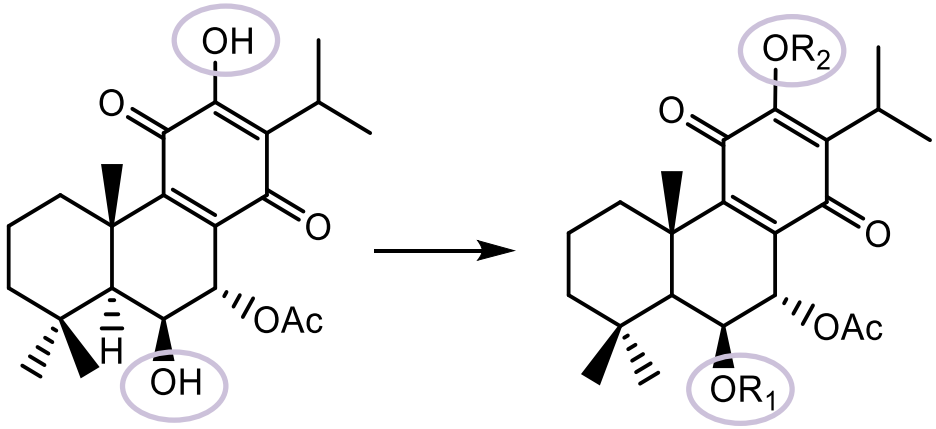
RoyBz

- First small molecule **PKCδ-selective activator**: binds to the PKCδ-C1-domain
- Potently inhibited the proliferation of colon cancer cells
- A novel **anticancer drug candidate**, particularly in colon cancer therapy

Bessa C, Soares J, Raimundo L, Loureiro, J. B., Gomes, C., Reis, F., Soares, M. L., Santos, D., Dureja, C., Chaudhuri, S. R., Lopez-Haber, C., Kazanietz, M. G., Gonçalves, J., Simões, M. F., Rijo, P., Saraiva, L. Cell Death Dis., 9(2) (2018).

Objective

Achieving a small library of compounds with enhanced cytotoxic potential

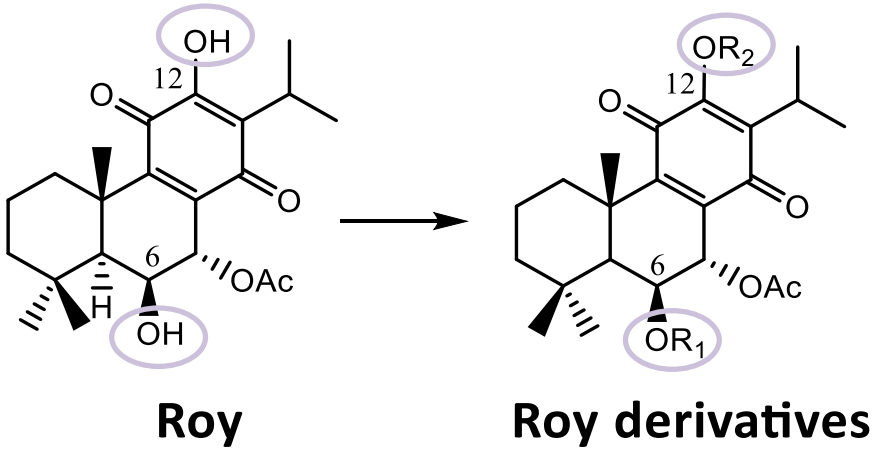


Roy **Roy derivatives**

Results and discussion

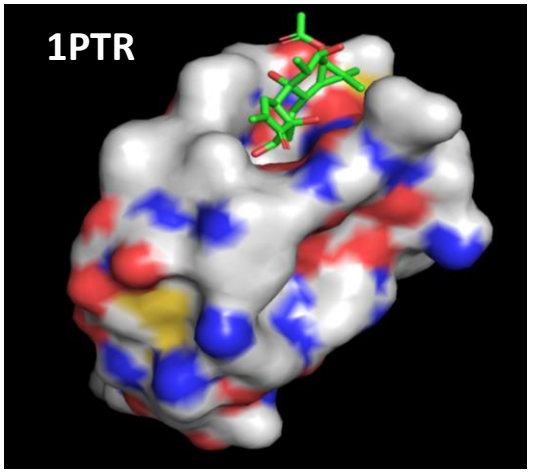
Molecular docking as tool to design new royleanone derivatives:

- Theoretical derivatives of Roy were designed through modification of the C-12 and C-6 hydroxyl groups.
- Molecular docking of theoretical derivatives in crystallographic structure of PKC-δ suggest which were the most promising compounds for hemi-synthesis.



Roy **Roy derivatives**

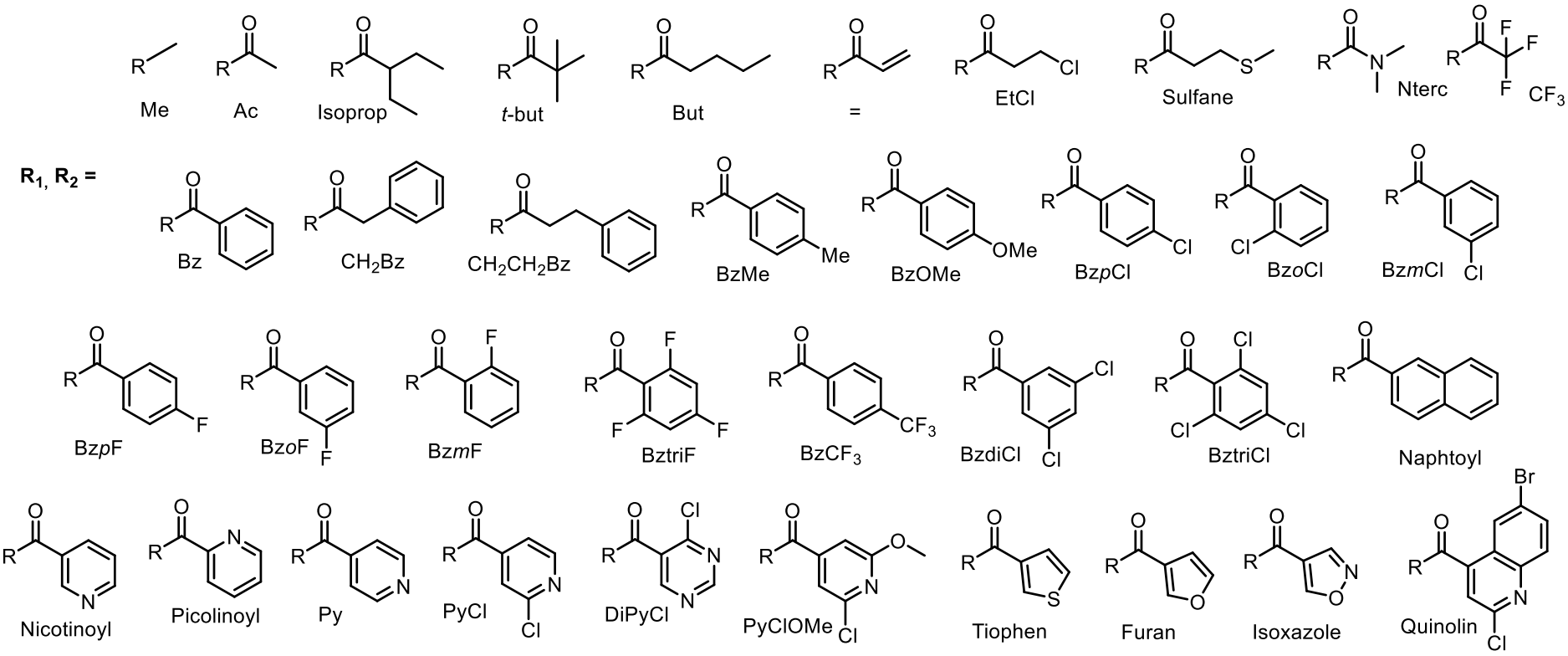
Molecular docking with Fred program



Crystallographic structure of human PKC-δ CYS2 domain complexed with natural inhibitor phorbol-13-acetate

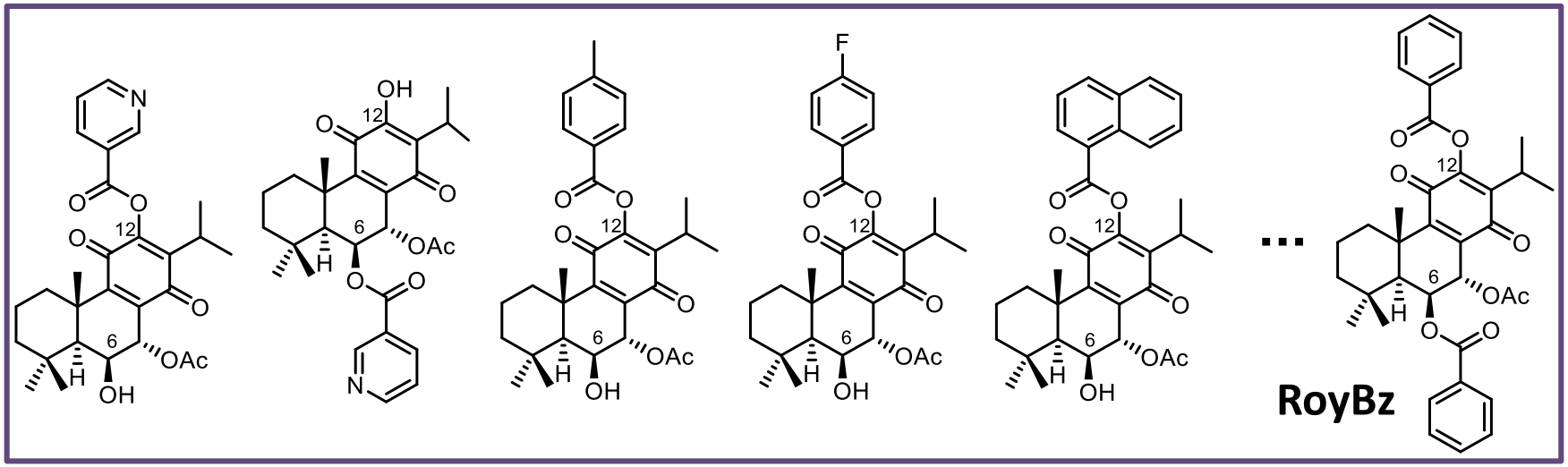
Results and discussion

Substituents tested in positions C-6 and C-12:



Results and discussion

Derivatization of both positions



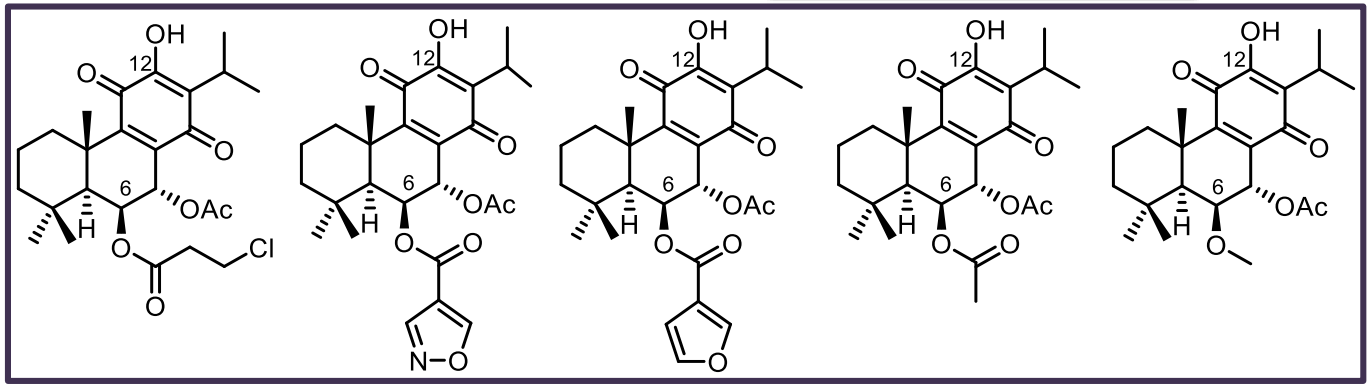
Decreased Score

- Derivatization of one position gives better scores than both
- Several molecules with better scores than RoyBz (selective PKC-δ activator)

Results and discussion

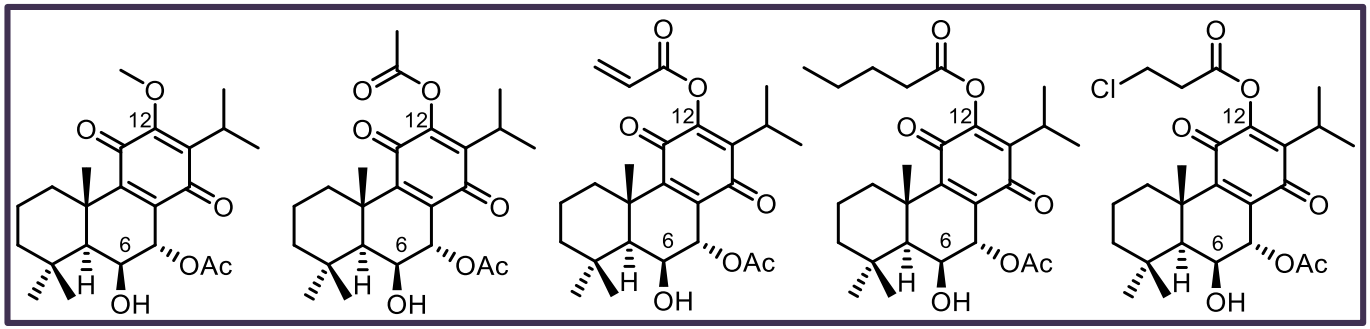
Derivatization of position C-6

Decrease Score



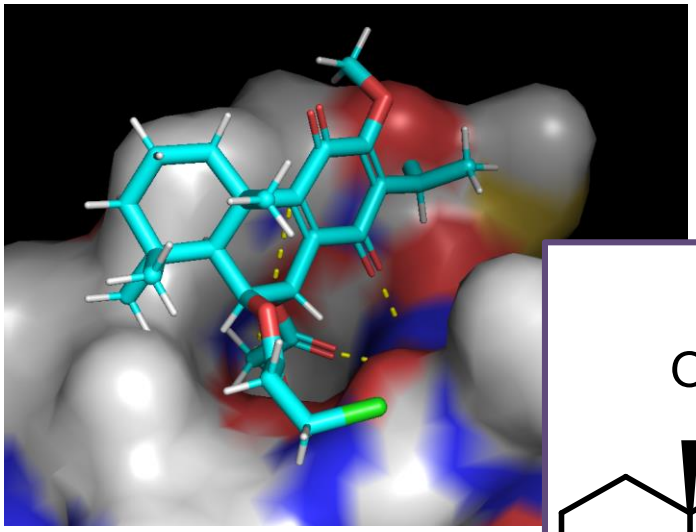
- Structural diversity

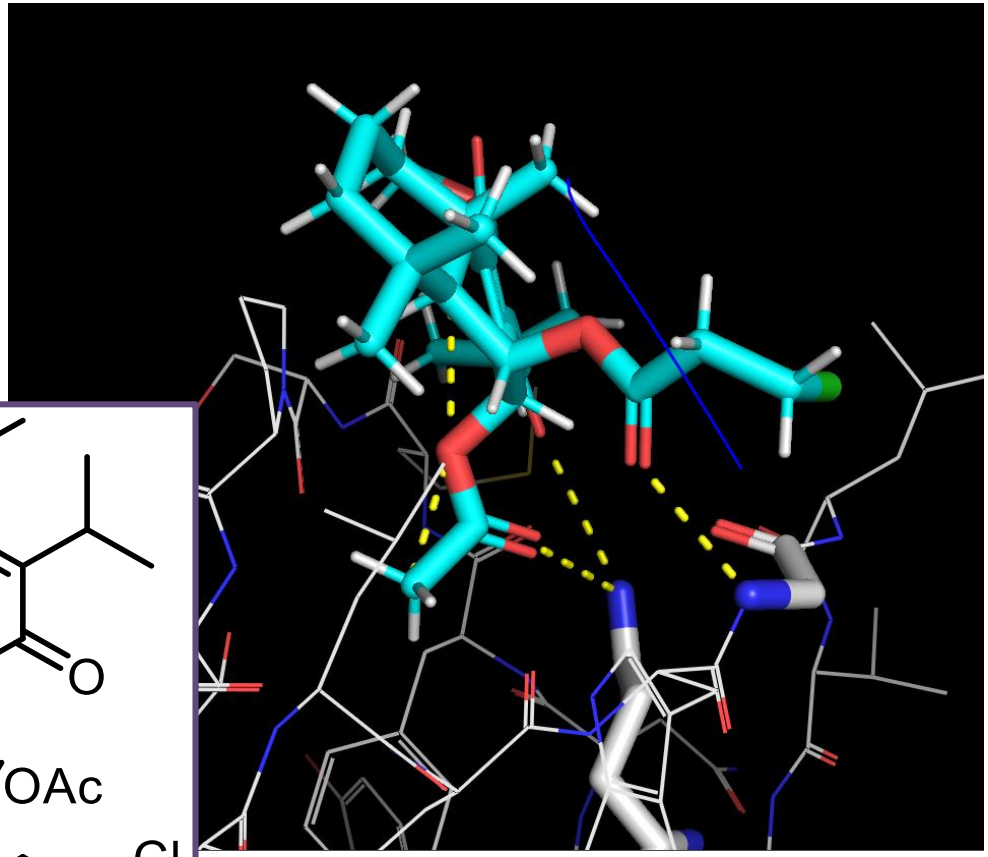
Derivation of position C-12



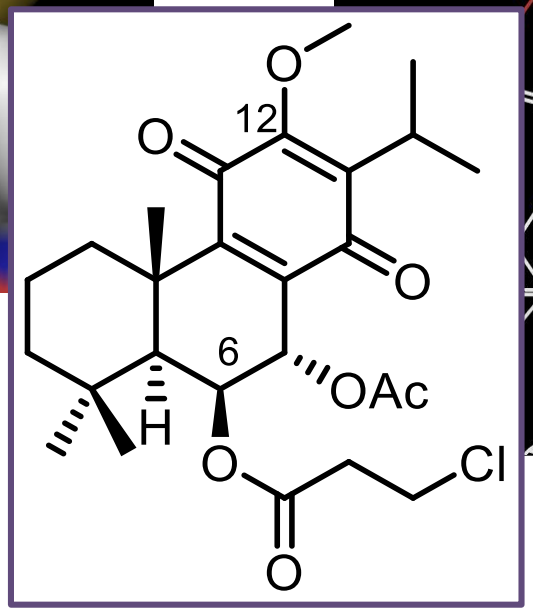
- Small linear groups

Results and discussion





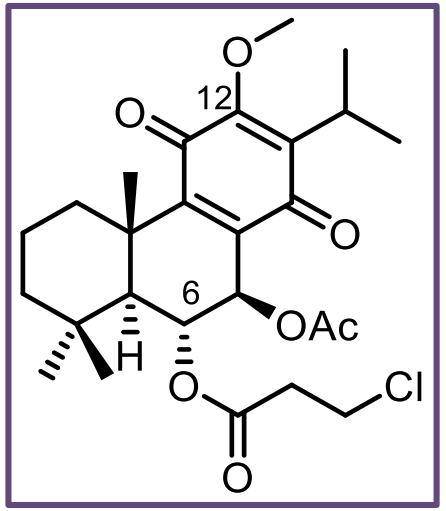
Best Hit obtained from docking screening:



3 Hydrogen bonds:

- Acetoxy group (C-17) to Glutamine 257
- C=O (C-14) to Glutamine 257
- Propionic group (C-6) to Glycine 253

Conclusions



- Several theoretical derivatives demonstrate higher scores than RoyBz.
- Derivatization of one position gives better scores than both.
- Docking Studies suggest that position 6 can bear high diversity of substituents, while position 12 requires small groups.
- Further studies regarding the effect on PKC-δ in cell lines should be conduct in the new derivatives that are currently been prepared, based on this docking results.

Acknowledgments

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Danke
Sukria
TAKK
Merci
Xie Xie!
THANK
YOU
EFHARISTO
TODA
grazi
tack
SHUKRAN
GRACIAS
KIITOS
INSTUTIY
Dankie