

Please cite the Published Version

Lee-Jones, L, Wang, Q, Bohan, S, Martin, O and Linton, PE (2016) Anti-cancer Properties of Secondary Metabolites derived from Marine Bacteria. In: 24th Biennial Congress of the European Association for Cancer Research, 09 July 2016 - 12 July 2016, Manchester, UK.

DOI: [https://doi.org/10.1016/S0959-8049\(16\)61517-4](https://doi.org/10.1016/S0959-8049(16)61517-4)

Publisher: Elsevier

Version: Accepted Version

Downloaded from: <https://e-space.mmu.ac.uk/621160/>

Usage rights: © In Copyright

Enquiries:

If you have questions about this document, contact openresearch@mmu.ac.uk. Please include the URL of the record in e-space. If you believe that your, or a third party's rights have been compromised through this document please see our Take Down policy (available from <https://www.mmu.ac.uk/library/using-the-library/policies-and-guidelines>)

a reduction of the final tumor volume ($p=0.03$) and an overexpression of VASH ($p=0.03$), but without affecting the vascular amount ($p>0.05$). When administered therapeutically, all the tested compounds were able to inhibit tumor growth ($p<0.01$), assuming that the Q3G group had decreased final tumor volume ($p=0.04$) as well increased VASH expression ($p=0.03$) and decreased vascular proliferation ($p<0.05$). It was found an inversely proportional relationship between the tumor growth of human colon adenocarcinoma/HT-29 and VASH expression ($p<0.05$).

Conclusion: Flavonoids HR and Q3G demonstrated antiangiogenic potential in colon adenocarcinoma/HT-29 when administered prophylactic and therapeutically, respectively. Q3G showed direct inhibition of the neovascular proliferation.

No conflict of interest.

[653] Anti-cancer properties of secondary metabolites derived from marine bacteria

L. Lee-Jones¹, Q. Wang¹, S. Bohan¹, O. Martin¹, P.E. Linton¹. ¹Manchester Metropolitan University, Healthcare Science, Manchester, United Kingdom

Background: Natural combinatorial chemistry has been occurring in plants and microorganisms throughout evolutionary history and consequently is far more sophisticated than that achievable in the laboratory by current combinatorial chemical procedures. This makes natural products an invaluable source of novel bioactive molecules which could lead to the development of novel pharmaceutical compounds to treat cancer.

Material and Methods: Samples were collected from Heysham, West Lancashire. Marine bacteria were isolated to pure culture from marine tidal surfaces including marine invertebrates, molluscs and marine plants using actinomycete-seawater culture media. Resultant pure cultures were grown in liquid media and cell-free crude extracts were prepared by culture centrifugation and filter-sterilisation. Bacterial species were identified by 16S rDNA PCR-sequencing methods. Crude extracts were then screened for bioactivity in the human haematological cell lines (Jurkat, U937 and K562) to determine their effects on cell proliferation, cell cycle and induction of apoptosis.

Results and Discussion: From an initial pilot study of 12 extracts derived from several strains of actinomycetes, 5/12 demonstrated anti-proliferative properties for both 24 and 48 hour timepoints. A further extract demonstrated anti-tumour activity at the 48 hour time point only.

Conclusion: The pilot study successfully investigated 12 extracts derived from marine bacteria and had a high success rate (42% for both 24 and 48 hour time points, 50% for 24 hour and/or 48 hour timepoints). One of the extracts demonstrated anti-proliferative properties that were more potent than the hydroxyurea (positive) control. Evaluation of the bioactive properties of the remaining panel of extracts ($n=82$) is now underway. Marine bacteria represent a largely untapped resource with enormous potential as a source of novel bioactive compounds.

No conflict of interest.

[654] Flavonoids Q3G and hydrolyzed rutin demonstrated antitumor activity in colon adenocarcinoma – *in vivo* study

D. Priolli¹, D.D.C. Silva¹, A.C. Duarte², G.D.C. Orfali², N.P. Martinez¹, P.D.O. Carvalho¹, A.Y.K. Grizotto². ¹Universidade São Francisco, *Stricto sensu* in Health Science, Bragança Paulista, Brazil, ²Universidade São Francisco, Medicine Course, Initiation Scientific Programme, Bragança Paulista, Brazil

Background: Flavonoids are polyphenolic compounds widely distributed in the plant kingdom, whose importance lies in its beneficial physiological effects, including antioxidant and anti-proliferative effects. Carcinogenesis is a complex process that involves metabolic and cell proliferative changes by alteration in organic circuits, including oncogenes and tumor suppressor genes such as P53 and ING2.

Method: For the experiment, 25 athymic mice were used and randomly divided into 5 groups: control, therapeutic Q3G and HR, prophylactic Q3G and HR. All animals were implanted with colon adenocarcinoma (HT-29) tumor cell line under subcutaneous tissue. Control animals had their tumor growth assessed and used as a standard curve for comparative analysis. Prophylactic groups were gavaged with their respective drugs for 7 days prior to tumor implantation, while animals from therapeutic groups were implanted and gavaged only when their tumors reached volumes equal to or greater than 100 mm³. After an evaluation of tumor growth the animals were euthanized, their tumors resected and stored for histological analysis and immunohistochemistry. The mechanism of action of the compounds was evaluated by immunohistochemical analysis of paraffin embedded tumor pieces, investigating the expression of proteins that participate in cell regulation process and progression of carcinogenesis, p53 and ING2. The analysis of the results was carried out by adopting a significance level of 5% ($p \leq 0.05$).

Results and Discussion: Histological analysis showed poorly differentiated adenocarcinoma with the presence of signet ring cell. There was a reduction

of tumor growth in all groups tested in the control group ($p=0.00$). Immunohistochemical analysis revealed a reduction in the expression of p53 ($p=0.00$) in all groups compared to control group. There was no difference in the expression of ING2 ($p=0.97$). The drugs tested exhibited tumor growth reduction and involvement in antiproliferative pathways mediated by the p53 protein, corroborating literature data showing activation of apoptotic mitochondrial pathway of flavonoid.

Conclusions: Flavonoids Q3G and HR demonstrated antitumor activity. Reduction of tumor growth was observed in all groups, moreover it was directly proportional to the reduction in mutated p53 expression in the therapeutic Q3G group ($p=0.03$), indicating a potential pro-apoptotic action and cell cycle arrest (G1 / S phase) through p53 signaling pathways.

No conflict of interest.

[655] New biocompound for the treatment of glioma: Is rutin hydrolyzed by hesperinase an alternative?

C. Pacheco¹, P.M. Moro¹, R. Colenci¹, A.C. Duarte¹, M.C.F. Messias², G. Mamprim³, C.T. Parisi², D.G. Priolli², T.W. Santos². ¹São Francisco University, Medicine Course, Initiation Scientific Programme, Bragança Paulista, Brazil, ²São Francisco University, *Stricto sensu* in Health Science, Bragança Paulista, Brazil, ³São Francisco University, Medicine Course, Bragança Paulista, Brazil

Flavonoids have shown biological effects, including antitumor activity by cell cycle arrest, DNA repair and apoptosis induction. Quercetin-3-O-glucoside (Q3G) has been the aim of several studies; however, it is scarce in nature. Q3G can be obtained with enzymatic de-glycosylation of quercetin-3-rutinoside (Rutin), resulting in a mixture addressed as Hydrolyzed Rutin (HR), with significant increase in bioavailability and antitumor potential. This study sets out to evaluate HR's cytotoxic activity *in vitro*, and its antiproliferative, pro-oxidative and genotoxic potential in animal model of glioma/U-251.

Method: Tumor cell lines panel were evaluated for HR's antiproliferative activity using Sulforhodamine B assay. HR's action mechanisms were also investigated in cells apoptosis and cell cycle by flow cytometry. 15 nude mice randomly were divided in 3 groups: control ($n=7$), HR treatment ($n=4$) and HR prophylaxis ($n=4$). All animals were grafted with human Glioma/U-251. The control group didn't receive any treatment. The HR prophylaxis group received the biocompound for 5 days prior to tumor graft, while the HR treatment group first received the graft, initiating compound administration only when tumors reached volumes ≥ 100 mm³. All animals had tumor growth daily monitored. The control group was used as a standard for comparative analysis. The animals were euthanized and the tumors resected. The histopathological analysis used to confirm diagnosis, morphological characteristics, number of mitosis and the presence of necrosis. Lipid peroxidation was done by TBARS method and genotoxicity tests by COMET assay. The results were carried out adopting a significance level of 5%.

Results: Among the tumor cell lines tested, HR obtained the best antiproliferative effect on Glioma/U251, able to reach significant results of total growth inhibition (TGI) with 3.6 µg/mL of drug concentration. No results were found in apoptosis flow cytometry. However, HR showed action of cell cycle arrest in the G1 phase, demonstrating a cytostatic effect. There was tumor growth inhibition on treated ($p=0.03$) and prophylaxis group ($p=0.01$). There was a decrease in mitoses ($p<0.04$) in the treated groups. There was an increase in lipid peroxidation of both prophylactic and therapeutic HR groups. There was no significant variation in oxidative DNA damage level.

Conclusion: HR exhibited moderate antiproliferative action in glioma. Cytotoxicity was proven by TBARS assay; although, no genotoxic effect was shown. The evaluation of possible action mechanisms of HR in glioblastoma demonstrated cytostatic effect, but no apoptosis induction. Therefore, HR presents antiproliferative effect in high grade glioma/U-251. Results suggest that mitosis reduction and cellular morphology changes are due to mechanisms related to drug action through a process of apparent oxidative damage on the cell membrane.

No conflict of interest.

[656] Hydrolyzed rutin in an animal model of human glioma

P. Prado¹, C. Pacheco¹, C.T.P. De Oliveira², P.D.O. Carvalho², G. Mamprim³, D. Priolli², A.Y.K. Grizotto¹. ¹São Francisco University, Medicine Course, Initiation Scientific Programme, Bragança Paulista, Brazil, ²São Francisco University, *Stricto sensu* in Health Science, Bragança Paulista, Brazil, ³São Francisco University, Medicine Course, Bragança Paulista, Brazil

Among the various types of cancers, glioma is configured as a neoplasm of high power of invasion, proliferation and recurrence, being among the most lethal human cancers. The average survival time is usually less than one year after diagnosis. Thus, the need to search for new drug to combat this tumor improves survival of patients. Flavonoids are phenolic compounds of natural origin that are present in most foods of the human diet and have antitumor activity.