Medicinal chemistry

Our project is organised around the following three cornerstones: (i) the training of researchers; (ii) the creation of knowledge; and (iii) the transfer of knowledge to society. In this regard, this is an integrated project, because although building on a robust chemical platform, the main goal is to identify compounds, mainly natural products or analogues, with biological/therapeutic activity (drug discovery), and to facilitate their reaching the target (drug delivery systems). Very often containing complex structural features that cover a more diverse chemical space than combinatorial chemistry libraries, natural compounds offer great opportunity to discover novel biological activities. Furthermore, a combination of natural products and combinatorial chemistry provides a potent approach for the discovery of new and safer therapeutic compounds.

Peptide synthesis is based on an appropriate combination of protecting groups and a suitable choice of coupling method. In this regard, a new protecting group for Arginine (Arg) and a novel family of coupling reagents have been developed.

We have developed 1,2-Dimethylindole-3-sulfonyl (MIS) for the side-chain of Arg. The protection of Arg side chains is a crucial issue in peptide chemistry because of the propensity of the basic guanidinium group to produce side reactions. Currently, sulfonyl-type protecting groups, such as 2,2,5,7,8-pentamethylchroman (Pmc) and 2,2,4,6,7-pentamethyldihydrobenzofuran(Pbf), are the most widely used for this purpose. Nevertheless, Arg side chain protection remains problematic as a result of the acid stability of these two compounds. This issue is even more relevant in Arg-rich sequences, acid-sensitive peptides and large-scale syntheses. The MIS group is more acid-labile than Pmc and Pbf and is therefore a better option for Arg side chain protection. In addition, MIS is compatible with tryptophan-containing peptides (Isidro-Llobet et al., 2009).

At present, almost all peptide bonds are formed in the presence of 1-hydroxybenzotriazole (HOBT) or its derivatives. After September 11, HOBT has been declared to have a high risk of explosion. Our group has proposed Oxyma Pure® as a substitute for HOBT. Oxyma Pure® displays a remarkable capacity to inhibit racemisation, together with impressive coupling efficiency in both automated and manual synthesis, superior to that shown by HOBT. Calorimetry assays showed decomposition profiles for HOBT-based additives that were consistent with their reported explosiveness and suggested a lower risk of explosion in the case of Oxyma Pure® (Subirós-Funosa et al., 2009).

Furthermore, we have introduced (El-Faham et al., 2009) a new family of uronium-type coupling reagents based on the presence of Oxyma Pure®. COMU® contains a morpholino group in conjunction with Oxyma Pure®. COMU® shows less tendency to racemise and higher coupling efficiency than HOBT derivatives.
Both Oxyma Pure® and COMU® are currently on the worldwide market (El-Faham et al, 2009; Subirós-Funosa et al, 2009).

**Synthesis of natural products. Medicinal chemistry programmes**

Aplicyanins are a new family of indole alkaloids recently isolated from the ascidian *Ap- lidium cyaneum*. These compounds are cytotoxic to the human tumour cell lines MDA-MB-231 (breast adenocarcinoma), A549 (lung carcinoma), and HT-29 (colorectal carcinoma) and also exhibit antimitotic activity. We have reported the first total synthesis of the indole alkaloids (+)-aplicyanins A, B, and E, plus 17 analogues, all in racemic form. Modifications to the parent compound included changing the number of bromine substituents on the indole, the nature of the substituents on the indole nitrogen (H, Me, or OMe), and/or the oxidation level of the heterocyclic core tetrahydropyrimidine. Each compound was screened against three human tumour cell lines, and 14 of the newly synthesised compounds showed considerable cytotoxicity. The assay results were used to establish structure-activity relationships (SARs). These results suggest that the presence of the bromine at position 5 of the indole is critical for activity, in the same way as the acetyl group on the imine nitrogen is in some compounds (Sisa et al, 2009).

Lamellarins form a family of more than forty members of marine alkaloids that show relevant bioactivity. Lamellarin D (Lam-D) is a cytotoxic agent against various tumour cells, an inhibitor of topoisomerase I, and a potent pro-apoptotic agent. We recently described the total synthesis of Lam-D, the preparation of a library of more than forty analogues, and SAR studies. We have reported the preparation of PEG-conjugates to improve the solubility of this compound, studies of cell penetration, and the apoptotic mechanism of cell death. A second generation of bio-conjugates with a nuclear localisation signal peptide and a poly(ethylene glycol)-based dendrimer has also been studied (Pla et al, 2009).

Dictyodendrins are a family of alkaloids isolated from the sponge *Dictyodendrilla ve- rongiformis* collected off the southern coast of Japan. These alkaloids have a common pyrrole[2,3-c]carbazole core but differ in their respective substituents at the α position of the pyrrole ring and in their degree of oxidation. We have synthesised the pyrrole[2,3-c]carbazole. The sequence is based on a Suzuki cross-coupling reaction.
Structures of Lamellarin D bioconjugates.

Figure 3. Structures of Lamellarin D bioconjugates.

Taking Thiocoraline, a potent marine anti-tumoral cyclic thiodepsipeptide, as a model, we have demonstrated that bridged N-methyl amides are isosteres for thiodepsipeptide bonds. The introduction of NMe-amides in bridges mimicked the thioester bonds without imposing steric hindrance and allowed conservation of the hydrogen bonding map of the natural product. NMe-azathiocoraline displayed nanomolar activity in the same order as the natural product, and the same mode of action. In fact, modelling of NMe-azathiocoraline bonded to a TCGA sequence showed how the methyl groups remained further away from the DNA strand, without changing the recognition pattern of thiocoraline. This synthetic approach could be used in other depsipeptides and side-chain to side-chain cyclic peptides. Taking advantage of the molecule symmetry, we have synthesised NMe-azathiocoraline by a total solid-phase convergent approach, using a complete arsenal of coupling methods and protecting groups (Tulla-Puche et al., 2009).

Siamese depsipeptides as new constrained bicyclic architectures

Taking as a model sansalvamide A (SA), which is produced by a marine fungus and shows cancer cell cytotoxicity, we constructed cyclic depsipeptide dimers connected by a CC single bond. Remarkably, these analogues showed greater activity than the natural product, thereby providing additional information on the SAR (Ruiz-Rodriguez et al., 2009).

We have reported the leishmanicidal activity of Kahalalide F (KF), which is a tumoricidal cyclic depsipeptide currently under phase II clinical trials for several types of cancer and psoriasis, and its synthetic analogues at a micromolar range of concentrations (Cruz et al., 2009). The lethality of this compound is strongly linked to the alteration of the plasma membrane (PM) of the parasite and is based on: (i) rapid depolarisation of the PM and uptake of the vital dye SYTOX Green upon addition of the dye; (ii) evidence of severe morphological damage to the membrane of the parasite, as shown by transmission electron microscopy; and (iii) a rapid drop in the intracellular ATP levels, which correlates significantly with the leishmanicidal activity for active analogues. In addition to the basic knowledge obtained, this class of lethal mechanism is considerably less prone to the induction of resistance than classical drugs. All together, these observations foster further studies for the optimisation of KF and its analogues as novel anti-Leishmania leads with a new mode of action (Cruz et al., 2009).

In addition, two other small molecule-type compounds designed and synthesised in our group are entering the pre-clinical phase at the Bellvitge and Vall d’Hebron hospitals, respectively.

Nanotechnologies. Drug delivery systems

Kahalalide F (KF) conjugated to gold nanoparticles (GNPs). Two Cys-containing analogues of the anticancer drug KF were synthesised and conjugated to 20- and 40-nm GNPs. The self-
assembly capacity of a peptide dramatically influences the final ratio number of molecules per nanoparticle, saturating the nanoparticle surface and prompting multilayered capping on the surface. In such way, the nanoparticle could act as a concentrator for the delivery of drugs, thereby increasing bioactivity. GNP size and conjugation have an influence on biological activities. KF analogues conjugated with GNPs are located subcellularly at lysosome-like bodies. This distribution may be related to the action mechanism of KF. The results suggest that the selective delivery and activity of KF analogues are improved by conjugating the peptides to GNPs (Hosta et al, 2009).

Using optical tweezers (OT) and atomic force microscopy (AFM), we have studied the topoisomerase activity of Lam-D. OT exhibited a large increase in hysteresis of the force cycles as a result of the initial nicking activity of Topo I. The presence of Lam-D prevents the religation step and blocks enzyme turnover, as evidenced by the absence of a higher force plateau and by large, non-vanishing hysteresis between the stretching and relaxing paths of the force cycle. AFM showed that Lam-D blocks Topo I cleaving activity, as indicated by the prevalence of supercoiled topoisomers. Taken together, the data obtained indicate that, upon Lam-D inhibition, Topo I keeps a non-covalent interaction with the 50 end of the cleaved DNA strand. This interaction is strong enough to prevent supercoil relaxation in solution (Pla et al, 2009).

Scientific output

Publications


**Research projects and networks**

Advancing the field of drug delivery - Combined targeted treatments against human breast cancer

Spanish Ministry of Science and Innovation, EUI2008-00174 (2009-2011)

Principal investigator: Fernando Albericio

Cooperación transnacional para la innovación tecnológica en el desarrollo de moléculas para el tratamiento de la obesidad y de la diabetes

SUDOE - Interreg IV, DIOMED - SOE1/P1/E178 (2009-2012)

Principal investigator: Fernando Albericio

Creación de un laboratorio de nanobiotecnología para el desarrollo de nuevas herramientas para el diagnóstico y terapia de enfermedades de interés regional con la Universidad de Santiago de Chile

Spanish Agency for International Cooperation (AECI), D/021016/08 (2009)

Principal investigator: Fernando Albericio

Identificación de moléculas antimicrobianas miméticas obtenidas a partir de mapeo de péptidos activos con la Universidad Católica de Valparaíso

Spanish Agency for International Cooperation (AECI), A/016856/08 (2009)

Principal investigator: Fernando Albericio

Plataforma comibiquimica basada en productos naturales: descubrimiento y administración de fármacos


Principal investigator: Fernando Albericio

Quimica combinatória per al desenvolupament de nous composts Generalitat de Catalunya. 2009SGR1024 (2009-2013)

Principal investigator: Fernando Albericio

Synthesis and evaluation of antimycobacterial peptides targeting MDR and XDR strains

Spanish Ministry of Science and Innovation, HS2008-0009 (2009-2010)

Principal investigator: Fernando Albericio

**Collaborations**

**Antiinflammatory compounds**

Enrique Pérez-Payá, Instituto Príncipe Felipe (Valencia, Spain)

Anti-leishmanias compounds

Luis Rivas, CSIC (Madrid, Spain)

**Antimicrobial peptides**

Sergio Marshall and Fanny Guzmán, Catholic University of Valparaiso (Valparaiso, Chile)

Antimycobacterial peptides targeting MDR and XDR strains

Thavi Govender, University of KwaZulu Natal (KwaZulu Natal, South Africa)

**Antitumoral compounds**

Rosa Aligué, Faculty of Medicine, University of Barcelona (Barcelona, Spain)

**Biological evaluation of molecules and ChemBioBank**

Mabel Loza, University of Santiago de Compostela (Santiago, Spain)

**Combinatorial chemistry for purification of proteins**

Osvaldo Cascone, University of Buenos Aires (Buenos Aires, Argentina)

**Delivery systems for siRNA**

Ramon Eritja, IRB Barcelona (Barcelona, Spain)

Dendrimers as drug delivery systems

Simó Schwartz Jr, Institut de Recerca Hospital Universitari Vall d’Hebron (Barcelona, Spain)

**Development of nanoparticles as vehicles for the treatment of metastatic colorectal cancer**

Ramón Manguès, Institut de Recerca Hospital de Sant Pau (Barcelona, Spain)

**Multiple agonists for D1 and D2 dopamine receptors**

Rafael Franco, University of Barcelona (Barcelona, Spain)

**Nanoparticles for therapy**

Marcelo Kogan, University of Chile (Santiago, Chile)
Production of libraries and medicinal chemistry program
Almirall (Barcelona, Spain)

Synthesis and conformational analysis of cyclodepsipeptides from marine origin
Ernest Giralt, IRB Barcelona (Barcelona, Spain)

Synthesis of natural products of marine origin
Instituto Biomar (Leon, Spain); PharmaMar (Madrid, Spain)

Synthesis of peptides
Lonza AG (Visp, Switzerland); Luxembourg Biotech (Rehovot, Israel)

Therapeutic compounds
Antonio Zorzano, IRB Barcelona (Barcelona, Spain)

Therapeutic polymers
Maria Jesus Vicent, Instituto Príncipe Felipe (Valencia, Spain)

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