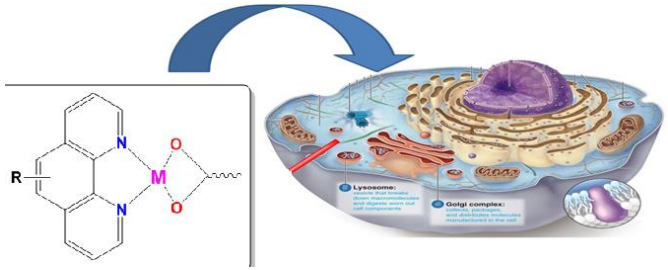


A Brazil-Ireland Platform for the Development of Metal-Based Therapeutics

Professor Michael Devereux
Dublin Institute of Technology

PURPOSE OF THE COLLABORATION

A interdisciplinary team of **chemists and biologists** with diverse and complementary expertise undertaking a comprehensive chemical and biological research programme aimed at **developing novel metal-based therapeutic agents** with potential beneficial effects for:

i) Treatment of a range of serious microbial pathogenic diseases including bacterial, protozoal and fungal infections, some of which present specific public health challenges in Brazil

(i) Age-related diseases such as Parkinson's and Cancer.

THE PRINCIPAL AND KEY COLLABORATORS

Maynooth University
National University of Ireland Maynooth

Malachy McCann
Kevin Kavanagh
Denise Rooney

UFRJ

André L.S. Santos
Marta H. Branquinha
Marcos D. Pereira
Livia Viganor

D·I·T
Dublin Institute of Technology
Institiúid Teicneolaíochta Átha Cliath

Michael Devereux
Orla Howe
Pauraic McCarron
Mary McNamara

Cátia Lacerda Sodré

UFES

Ana Paula Ferreira Nunes

ITT DUBLIN

Bernie Creaven

FIOCRUZ

Lucimar F. Kneipp
Mariangela Ziccardi de Camargo Salles
Fernanda Lopes Fonseca

DCU

Andrew Kellett

UENF

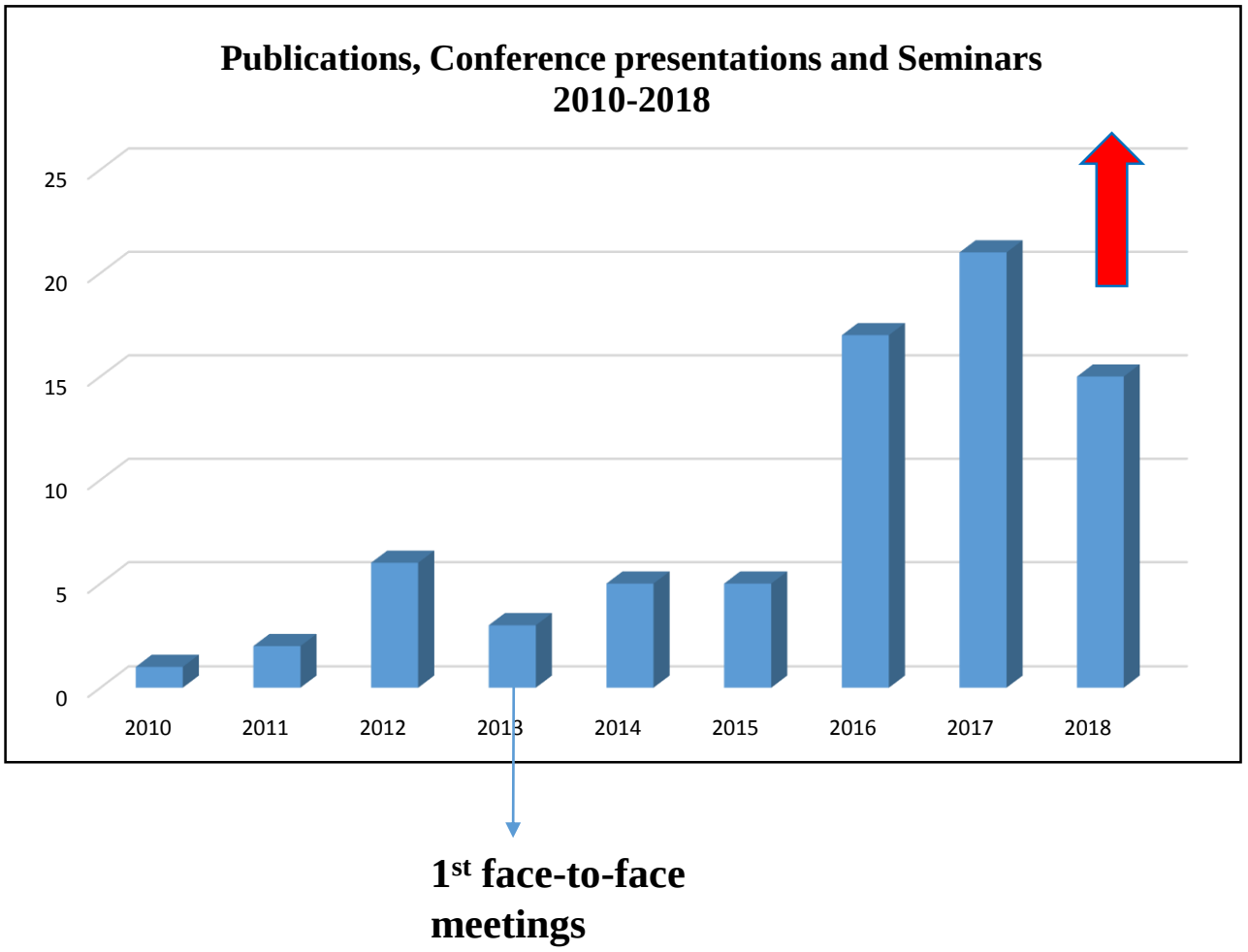
Adolfo Horn Jr.
Sarah S. Ferreira

Note: The collaboration has also involved approximately 25 postgraduate and postdoctoral researchers

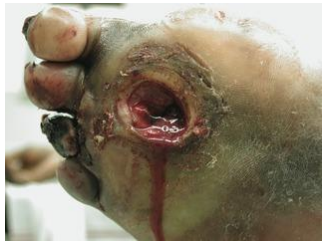
ESTABLISHMENT OF THE COLLABORATION

2005: Initial contact made by André Santos (UFRJ) – interested in published antimicrobial activity profiles for metal-phenanthroline complexes (MU and DIT).
2012: First joint publications (UFRJ, MU, DIT, DCU) – with no face-to-face engagement.
2013 – 2016: Face-to-face meetings in Rio de Janeiro and Dublin (**SFI-ISCA** and **FAPERJ**).
2014: Established the Metal-Based Therapeutics Working Group as part of the Research Brazil-Ireland (RBI) initiative.
2015: 1st Brazil-Ireland Science Week held in Dublin Castle – significant face-to-face opportunity (six Brazilian collaborators attended).
2015 – present: two-way student and staff mobility (Supported by Science Without borders, FAPERJ, SFI-ISCA, HEA GOI International Education Scholarships, DIT).

Collaboration Outputs

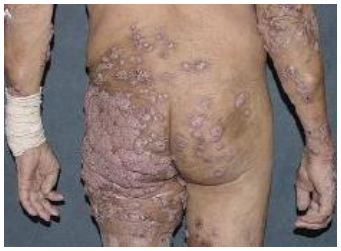


THERAPEUTIC FOCUS



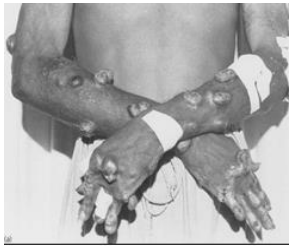
BACTERIAL

Pseudomonas aeruginosa
Klebsiela pneumonia
Acinetobacter baumannii
Mycobacterium tuberculosis



FUNGAL

Candida spp.
Scedosporium spp.
Fonsecaea pedrosoi
Phialophora verrucosa
Exophiala dermatitidis
Cladophialophora carrionii



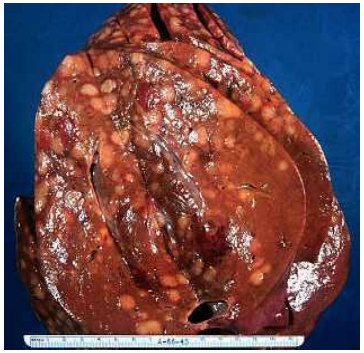
PARASITIC

Eg. Leishmania spp.

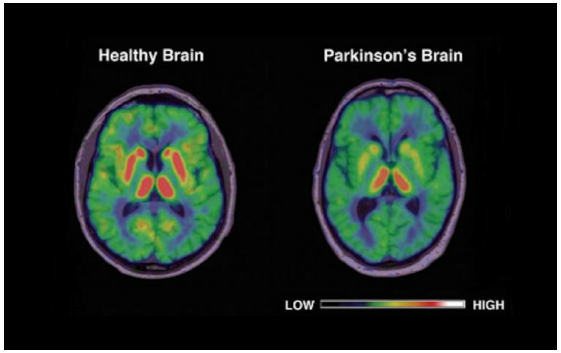
Drug resistance is a common

Present specific public health challenges in Brazil

Require cheap effective alternatives to expensive state-of-the-art drugs



ANTICANCER ACTIVITY



ANTI-NEURODEGENERATIVE ACTIVITY

THE CHEMISTRY - METAL-PHENANTHROLINE COMPLEXES

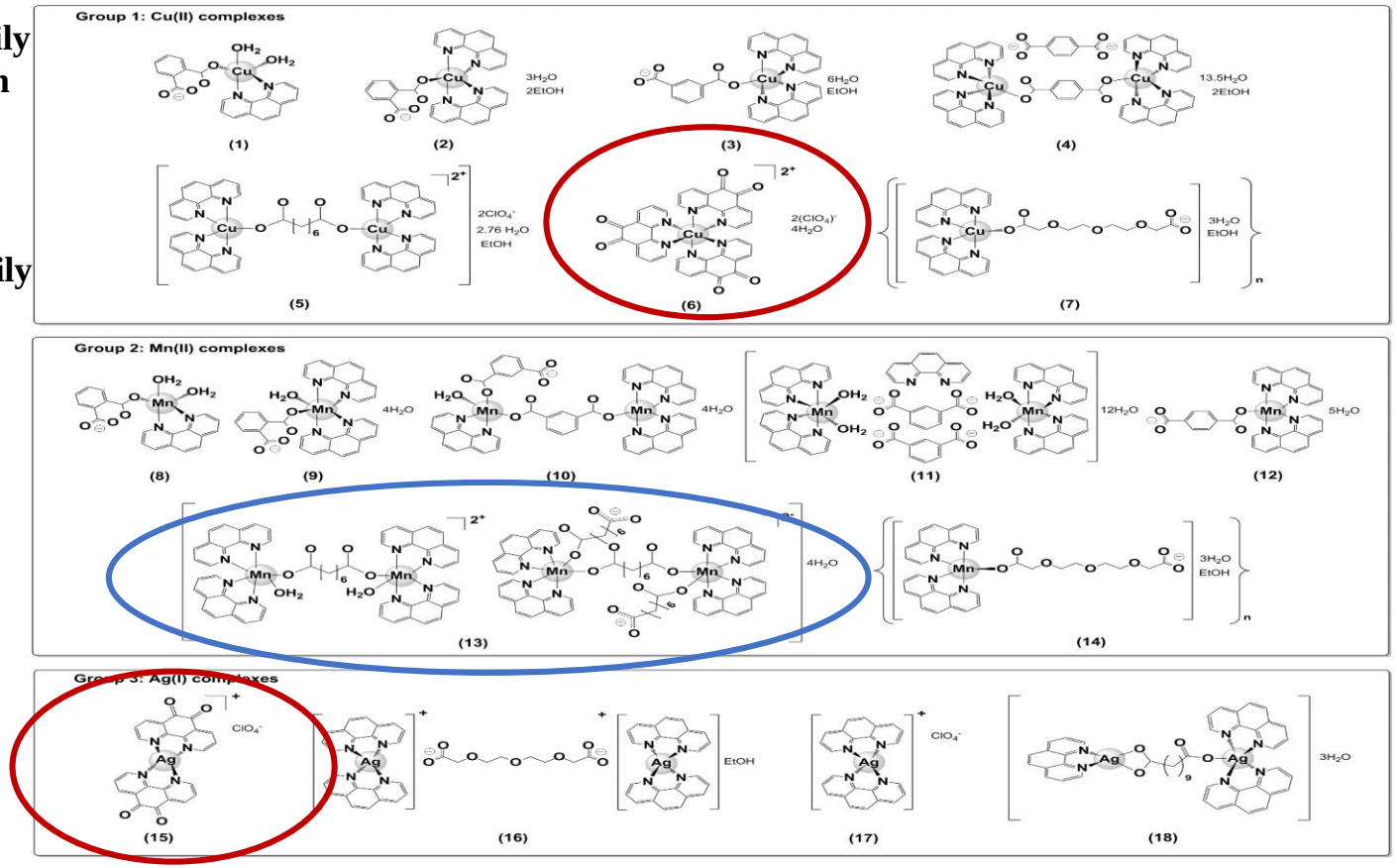
Lead: M. Devereux (DIT) and M. McCann (MU)

Cheap & easily synthesised in high yield.

Structural variation easily achieved.

The metal centre easily varied – Cu, Mn, Ag.

Stable with variable solubility and lipophilicity.



Active against a range of bacterial and fungal strains.

Exhibit biomimetic activity (SOD, CAT, nuclease, etc..).

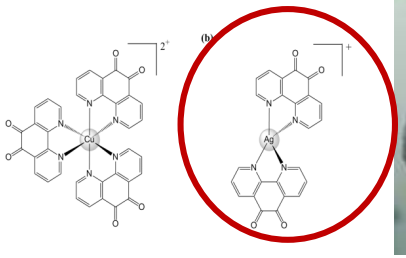
Display antioxidant & cytotoxic capabilities.

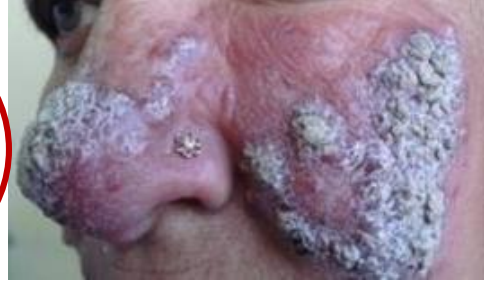
M. Devereux and McCann *et al* in:

Medicinal Chemistry Communications, 2011, 2, 579-584
Dalton Transactions, 2011, 40, 1024, 1024–1027.
Free Radical Biology and Medicine, 2012, **53**, 564-576.
Dalton Transactions. 2012, **41** (21), 6516 - 6527
International journal of clinical pharmacology and therapeutics, 2012, 50(1), 79-81
Journal of Medicinal Chemistry, 2012, **55**, 1957-1968.
Current Medicinal Chemistry, 2015, *22,* 2199-2224.
Journal of Inorganic Biochemistry, 2016, 159, 120-132.
Current Topics in Medicinal Chemistry, 2017, 17(11), 1280-1302.
Current Medicinal Chemistry, 2018, 25, 1-14.

Example 1: Studies on *Phialophora verrucosa* with $[Ag(phendione)_2]^+$

Lead: Lucimar Kneipp (Fiocruz)



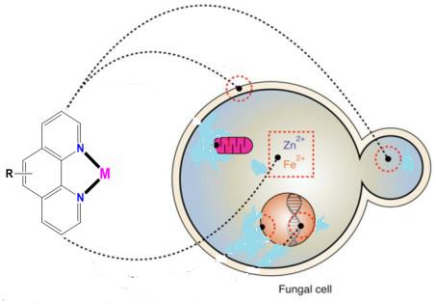


Phialophora verrucosa is an etiological agent for the chronic subcutaneous disease **Chromoblastomycosis** -

Characterised by polymorphic skin lesions – can lead to skin cancer
Affects mainly farm workers

Resistance to state-of-the-art antifungals is now a major problem - Expensive to treat

In vitro activity of **$[Ag(phendione)_2]^+$**



- Inhibits the cellular growth (MIC_{100} = 4.0 µM)
- Reduces ergosterol in the cell membrane
- Reduces metallopeptidase activity
- Induces morphological changes
- Active towards biofilm
- Reduces the viability of the fungus after interaction with human macrophages (THP-1)

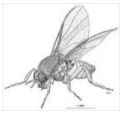
In vivo activity of **$[Ag(phendione)_2]^+$**

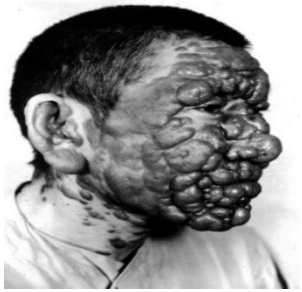
Promotes a protective effect in *Galleria mellonella* (waxmoth) larvae infected with *Phialophora verrucosa*.

1,10-phenanthroline-5,6-dione compounds are effective in blocking crucial physiological events of *Phialophora verrucosa*. M. Q. Granato, D. S. Gonçalves, S. H. Seabra, M. McCann, M. Devereux, M. C.V. Pessolani, A. L.S. Santos, L. F. Kneipp, ***Frontiers in Microbiology***, 2017, 8, article 76.
In vitro and *in vivo* studies of 1,10-phenanthroline-5,6-dione–based compounds on *Phialophora verrucosa* conidia cells. Granato, M.Q., Pereira, M., Pessolani, M.C.V., McCann, M., Devereux, M., Santos, A.L.S., Kneipp, L.F, in preparation.

Example 2:Studies on the *Leishmania braziliensis* parasite with $[Ag(phendione)_2]^+$ and $[Cu(phendione)_3]^{2+}$ Lead: André Santos (UFRJ)

Leishmania braziliensis parasite carried by Sand Fly

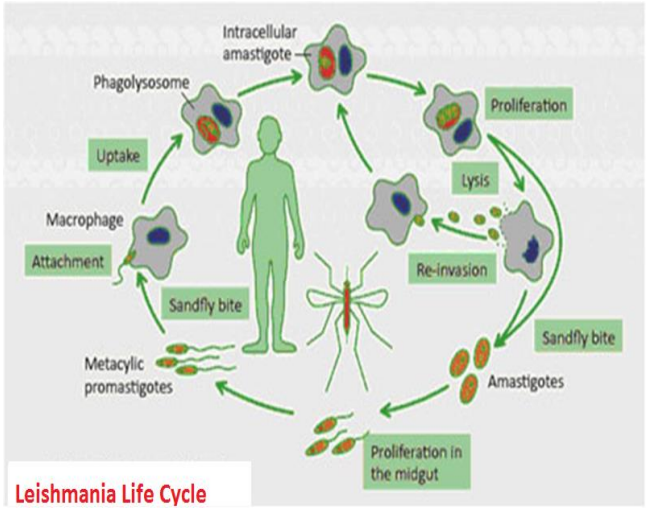


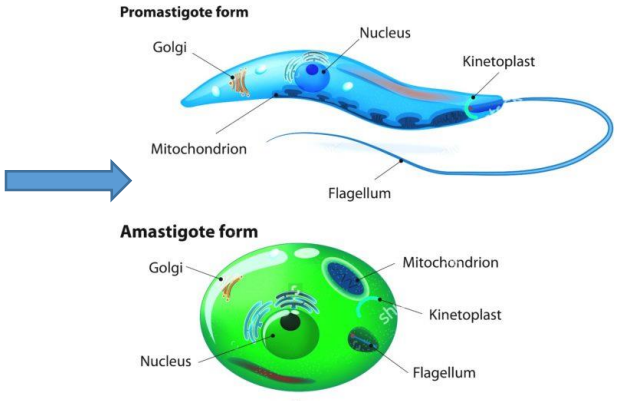


Leishmaniasis - endemic in Brazil

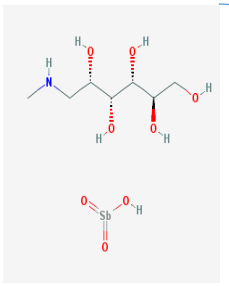
- clinical manifestations: cutaneous, mucocutaneous and visceral forms
- disfiguring with considerable morbidity and mortality

The promastigote is the infective form of the parasite



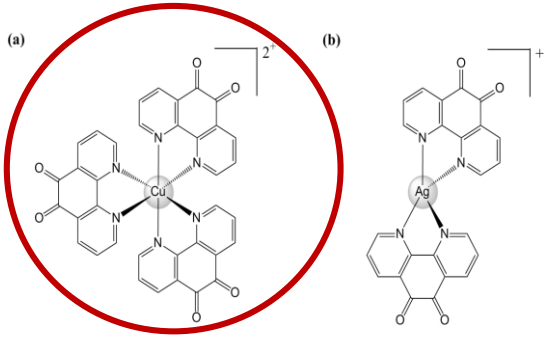


Current treatment involves antimony-based drugs such as Glucantime – but resistance now a major problem



Glucantime

Resistance



In vitro **- $[Cu(phendione)_3]^{2+}$ superior to $[Ag(phendione)_2]^+$**
(The mechanism of action has been extensively studied)

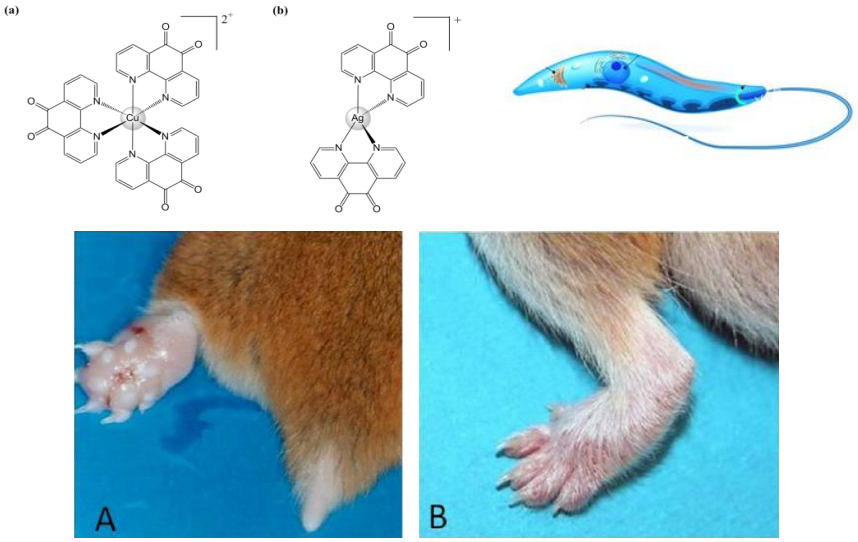
Unpublished results

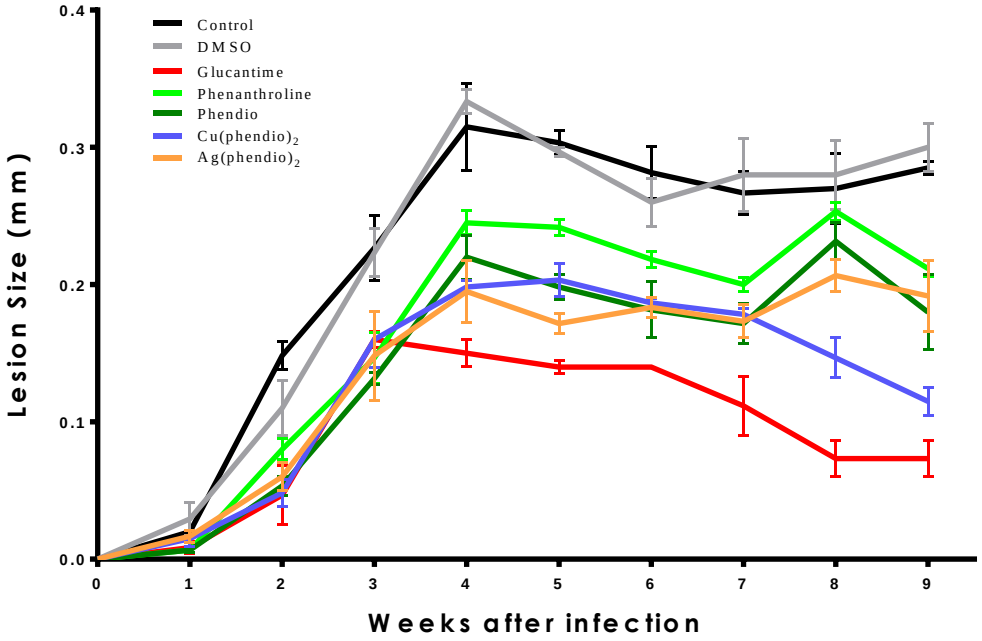
In vivo activity

- *L. braziliensis*-infected hamsters were treated with $[Ag(phendione)_2]^+$ and $[Cu(phendione)_3]^{2+}$

- Compounds intraperitoneally injected once a day for eight consecutive weeks.

- The lesion size on the foot was measured weekly over 8 weeks





Results

- Significant reduction in the size of the lesions compared to control (**Cu complex is best**)

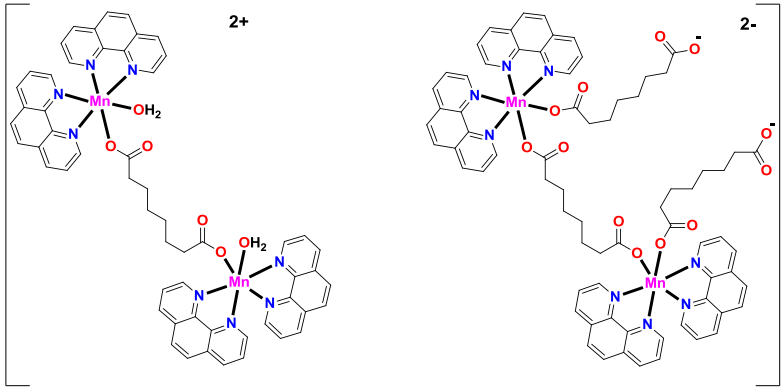
- The silver and copper compounds were well tolerated with no mortality observed during the period of treatment.

***In vivo* - $[Cu(phendione)_3]^{2+}$ displayed comparable activity to that of the clinical drug Glucantime**

Unpublished results

Example 3: Antioxidant and potential anti-neurodegenerative capability of $[Mn_2(oda)(phen)_4(H_2O)_2]^{2+}$ Lead: Marcos Pereira (UFRJ)

Solid-state structure of Mn_4 Double salt



$[Mn_2(oda)(phen)_4(H_2O)_2][Mn_2(oda)(phen)_4(oda)_2].4H_2O$

Aqueous solution

$2\ [Mn_2(oda)(phen)_4(H_2O)_2]^{2+} + 2\ oda^{2-}$

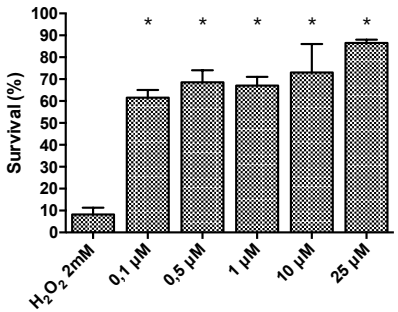
(where $odaH_2$ = octanedioic acid; phen = 1,10-phenanthroline)

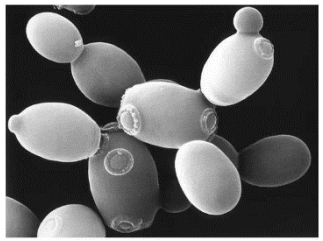
Antioxidant Activity of $[Mn_2(oda)(phen)_4(H_2O)_2]^{2+}$

- displays potent acellular superoxide dismutase (SOD) and catalase (CAT) activity.

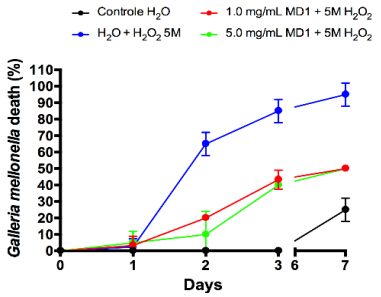
- protects *S. cerevisiae* and *G. mellonella* from H_2O_2-induced oxidative stress;

S. cerevisiae



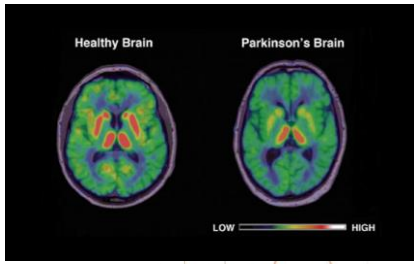


G. mellonella



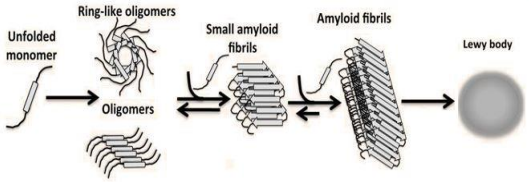
Evaluation of antioxidant activity of a Mn^{2+} coordination compound and its potential therapeutic use against alpha-synuclein aggregation. T. P. Ribeiro, J. Freitas, S. Frases, A. S. Pinheiro, M. McCann, M. Devereux, T. F. Outeiro and M. D. Pereira, in preparation,

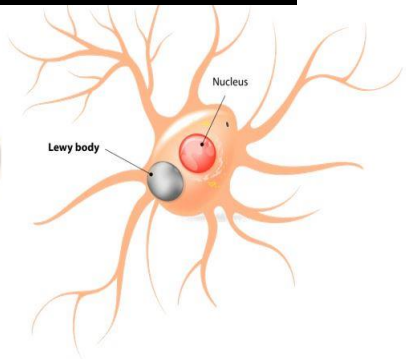
Anti-neurodegenerative capability



- α-Synuclein is a pre-synaptic protein, highly expressed in the central nervous system.

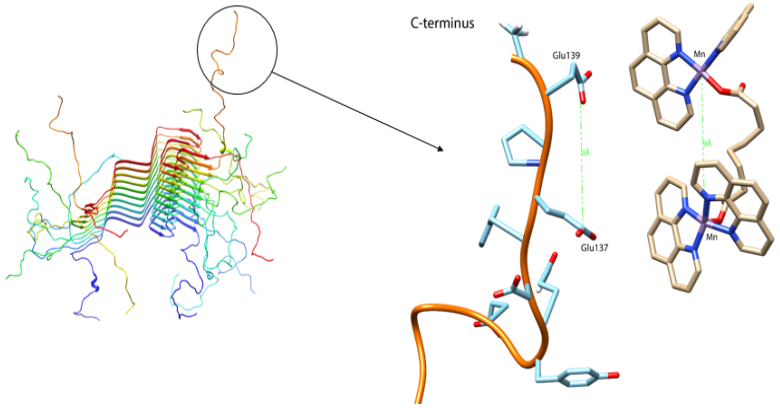
- Its oligomerisation, aggregation and accumulation leads to the formation of **Lewy Bodies**, the pathological hallmark of Parkinson's Disease.





- Lewy bodies develop in nerve cells in regions of brain involved in motor control

$[Mn_2(oda)(phen)_4(H_2O)_2]^{2+}$:



- reduces α-Synuclein toxicities in a yeast model of Parkinson's Disease.
- mitigates oligomerization and aggregation of α-Synuclein in mammalian neuroglioma H4 cells.
- NMR spectroscopy indicates that it binds to the C-terminal of α-Synuclein through interactions with Aspartic acid residues which are critical components for oligomerisation.

$[Mn_2(oda)(phen)_4(H_2O)_2]^{2+}$ offers potential as a prototype for Parkinson's Disease therapeutics

Evaluation of antioxidant activity of a Mn^{2+} coordination compound and its potential therapeutic use against alpha-synuclein aggregation. T. P. Ribeiro, J. Freitas, S. Frases, A. S. Pinheiro, M. McCann, M. Devereux, T. F. Outeiro and M. D. Pereira, in preparation,

THE INTRACELLULAR AND IN-VIVO ANTIOXIDANT ACTIVITY OF A MANGANESE(II) COMPLEX SALT AND ITS POTENTIAL THERAPEUTIC USE AGAINST α-SYNUCLEIN AGGREGATION

Thales P. Ribeiro[1], Júlia Freitas[1], Susana Frases[1], Anderson S. Pinheiro[1], Malachy McCann[2], Andrew J.S. Knox[3], Michael Devereux[3], Tiago F. Outeiro[4] and Marcos D. Pereira[1].

Chemotherapeutic potential of novel water-soluble and photo-stable silver dicarboxylate complexes containing 1,10-phenanthroline ligands

Orla Howe[1], Livia Viganor[1,2], Megan O' Shaughnessy[1], Pauraic McCarron[1], Leticia O.N. Assad[3], Marcos D. Pereira[3], Malachy McCann[4] and Michael Devereux[1].

[1]The Centre for Biomimetic & Therapeutic Research, Focas Research Institute, Dublin Institute of Technology, Camden Row, Dublin 8, Ireland
[2]Laboratory of Microbiology, Federal University of Rio de Janeiro (UFRJ), Brazil.
[3]Laboratory of Cytotoxicity and Genotoxicity, Department of Biochemistry – Institute of Chemistry, Federal University of Rio de Janeiro (UFRJ), Brazil.
[4]Chemistry Department, Maynooth University, National University of Ireland, Maynooth, Co. Kildare, Ireland

Development of Raman Spectroscopic model of drug-DNA interactions

Ferreira, S. S[1,2], Cullen, D[3], Horn Jr, A[4], Fernandes, C[4], Byrne, H.J.[3], Devereux, M[1,3] and Howe, O[1,3]

[1]The Centre for Biomimetic & Therapeutic Research, FOCAS Research Institute, Dublin Institute of Technology, Dublin 8, Ireland
[2] Instituto Federal Fluminense, IFF, Campos dos Goytacazes, Rio de Janeiro, Brazil
[3] FOCAS Research Institute, Dublin Institute of Technology, Dublin 8, Ireland
[4] Universidade Estadual do Norte Fluminense, UENF, Campos dos Goytacazes, Rio de Janeiro, Brazil

EFFECTS OF 1,10-PHENANTHROLINE AND ITS DERIVATIVES ON THE ELASTASE B ACTIVITY AND EXPRESSION OF *Pseudomonas aeruginosa*

L. Viganor[1,2]; A. C. M. Galdino[2,3]; M. McCann[4]; M. Hunt[5]; S. Sundkvist[6]; T. C. Ramalho[7]; A. A. Castro[7]; A. L. S. Santos[2,3] and M. Devereux[1]

UFRJ

Antifungal Potential of Copper(II), Manganese(II) and Silver(I) 1,10-Phenanthroline Chelates Against Multidrug-Resistant Fungal Species Forming the *Candida haemulonii* Complex

Rafael M. Gandra[1,2,3], Pauraic Mc Carron[3,4], Mariana F. Fernandes[1], Lívia S. Ramos[1], Thaís P. Mello[1], Ana Carolina Aor[1], Livia Viganor[3], Orla Howe[3], Marta H. Branquinha[1], Malachy McCann[4], Michael Devereux[3], André L. S. Santos[1,2]

[1]Laboratório de Investigação de Peptidases, Departamento de Microbiologia Geral, Instituto de Microbiologia Paulo de Góes, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil,
[2]Programa de Pós-Graduação em Bioquímica, Instituto de Química, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil,
[3]The Centre for Biomimetic & Therapeutic Research, Focas Research Institute, Dublin Institute of Technology, Dublin, Ireland,
[4]Chemistry Department, Maynooth University, National University of Ireland, Maynooth, Ireland

METAL-BASED DRUGS BIOLOGICAL ACTIVITY AGAINST *Phialophora verrucosa*

Marcela Queiroz Granato[1], Marcos Dias Pereira[2], Maria Cristina Vidal Pessolani[3], Malachy McCann[4], Michael Devereux[5], André Luís Souza dos Santos[6] Lucimar Ferreira Kneipp[1]

FIOCRUZ

UFRJ

Novel Dual SOD & CAT Mimics: A New Therapeutic Approach?

UFRJ

Deciphering the mechanisms of antimicrobial action of 1,10-phenantroline- 5,6-dione-based metallocompounds on *Pseudomonas aeruginosa*

Anna Clara M. Galdino, Lívia, V. Silva, Malachy McCann, Michael Devereux, Marta H. Branquinha, André L.S. Santos

OBSERVATIONS/LESSONS FROM THE COLLABORATION

- The collaboration effectively aligned the research question(s) to priority agendas for both countries (eg. *Leishmania* for Brazil v's *Pseudomonas aeruginosa.* as it relates cystic fibrosis in Ireland).

- Emphasis on use-inspired research – focusing on practical affordable solutions to real problems for society and enterprise.

- Mutual collateral gains through:
  - Experimental quality improvement
  - Expertise sharing (knowledge transfer)
  - Complementarity of scientific knowledge (eg. different point of views from chemist and biologists), helping to understand the complex biochemical events.
  - Human capital development - promoting the improvement of skillsets.

THE KEY: Mobility and face-to-face meetings have been critical to developing the relationship/collaboration (Funded through SFI-ISCA, HEA GOI International Education Scholarships FAPERJ, CAPES and CNPq)

THE CONCERN: sustainability of the collaboration (Funding challenges - Brazil and Ireland)?

Our first face-to-face meeting in UFRJ in November 2013

Transition Metals in Human Biology and Medicine. M. Devereux and M. McCann, ***a mini course (10 hrs) delivered to post-graduate biology and biochemistry students from the Institutes of Microbiology and Biochemistry, the Federal University of Rio de Janeiro (UFRJ)***, November 2013.