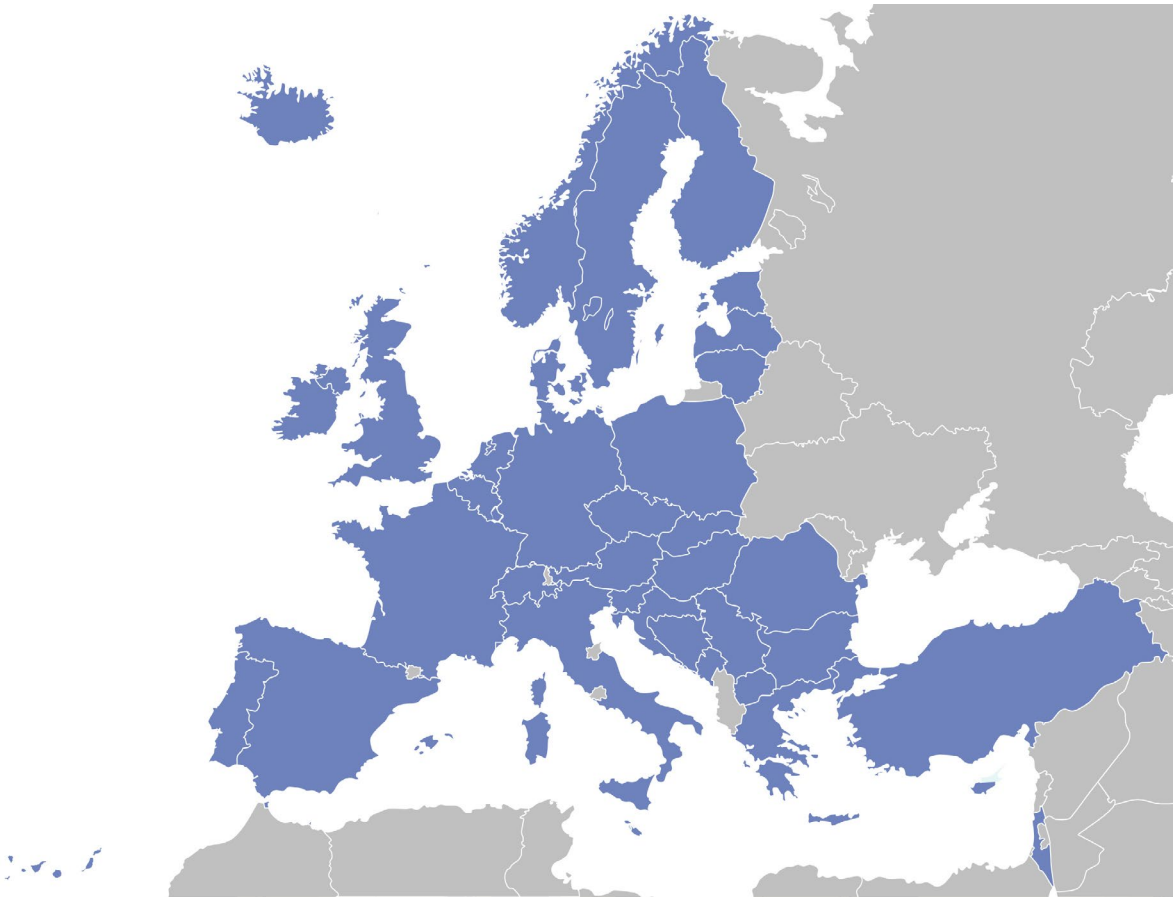
XJENZA ONLINE

VOLUME 5 SPECIAL ISSUE: COST 2017



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Xjenza
The Malta Council for Science & Technology

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Malta Chamber of Scientists

The Journal of the Malta Chamber of Scientists
Editor-in-Chief: Giuseppe Di Giovanni

Guest Editors: Janet Mifsud
David C. Magri

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Chronological List of Past and Present Editors of Xjenza The Journal of the Malta Chamber of Scientists

2013–
Editor: Giuseppe Di Giovanni
Associate Editors: *David Magri, Ian Thornton, Ian Cassar, Philip Farrugia, Sebastiano D'Amico, Nicholas Sammut, David Mifsud, Godfrey Baldacchino, Liberato Camilleri, Carmel Cefai*
Xjenza Online Vol. 4 Iss. 2 - December 2016
Xjenza Online Vol. 4 Iss. 1 - July 2016
Xjenza Online Vol. 3 Iss. 2 - December 2015
Associate Editors: *David Magri, Ian Thornton, Ian Cassar, Philip Farrugia, Sebastiano D'Amico, Nicholas Sammut, Joseph Galea, David Mifsud, Sandro Lanfranco, Mario Valentino, Godfrey Baldacchino, Liberato Camilleri*
Xjenza Online Vol. 3 Iss. 1 - August 2015
Xjenza Online Vol. 2 Iss. 2 - October 2014
Xjenza Online Vol. 2 Iss. 1 - March 2014
Xjenza Online Vol. 1 Iss. 2 - October 2013
Xjenza Online Vol. 1 Iss. 1 - March 2013

2003–2007
Editors: Joseph N. Grima and Richard Muscat
Xjenza Vol. 12 - 2007
Xjenza Vol. 11 - 2006
Xjenza Vol. 10 - 2005
Xjenza Vol. 9 - 2004
Xjenza Vol. 8 - 2003

1996–2002
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Xjenza Vol. 7 - 2002
Xjenza Vol. 6 - 2001
Associate Editors: *Martin Ebejer and Richard Muscat*
Xjenza Vol. 5 - 2000
Xjenza Vol. 4 Iss. 2 - 1999
Xjenza Vol. 4 Iss. 1 - 1999
Associate Editors: *Martin Ebejer, Richard Muscat, and Christian A. Scerri*
Xjenza Vol. 3 Iss. 2 - 1998
Xjenza Vol. 3 Iss. 1 - 1998
Associate Editors: *Martin Ebejer, Richard Muscat, Christian A. Scerri and Emmanuel Sinagra*
Xjenza Vol. 2 Iss. 2 - 1997
Xjenza Vol. 2 Iss. 1 - 1997
Xjenza Vol. 1 Iss. 2 - 1996
Xjenza Vol. 1 Iss. 1 - 1996

Scope of Journal

Xjenza is the Journal of the Malta Chamber of Scientists and is published in an electronic format. Xjenza is a peer-reviewed, open access international journal. The scope of the journal encompasses research articles, original research reports, reviews, short communications and scientific commentaries in the fields of: mathematics, statistics, geology, engineering, computer science, social sciences, natural and earth sciences, technological sciences, linguistics, industrial, nanotechnology, biology, chemistry, physics, zoology, medical studies, electronics and all other applied and theoretical aspect of science.

The first issue of the journal was published in 1996 and the last (No. 12) in 2007. The new editorial board has been formed with internationally recognised scientists, we are planning to restart publication of Xjenza, with two issues being produced every year. One of the aims of Xjenza, besides highlighting the exciting research being performed nationally and internationally by Maltese scholars, is to provide insight to a wide scope of potential authors, including students and young researchers, into scientific publishing in a peer-reviewed environment.

Instructions for Authors

Xjenza is the journal of the Malta Chamber of Scientists and is published by the Chamber in electronic format on the website: http://www.mcs.org.mt/index.php/xjenza. Xjenza will consider manuscripts for publication on a wide variety of scientific topics in the following categories

1. Communications
2. Research Articles
3. Research Reports
4. Reviews
5. Notes
6. News and Views
7. Autobiography

Communications are short peer-reviewed research articles (limited to three journal pages) that describe new important results meriting urgent publication. These are often followed by a full Research Article.

Research Articles form the main category of scientific papers submitted to Xjenza. The same standards of scientific content and quality that applies to Communications also apply to Research Articles.

Research Reports are extended reports describing research of interest to a wide scientific audience characteristic of Xjenza. Please contact the editor to discuss the suitability of topics for Research Reports.

Review Articles describe work of interest to the wide readership characteristic of Xjenza. They should provide an in-depth understanding of significant topics in the sciences and a critical discussion of the existing state of knowledge on a topic based on primary literature. Review Articles should not normally exceed 6000 words.Authors are strongly advised to contact the Editorial Board before writing a Review.

Notes are fully referenced, peer-reviewed short articles limited to three journal pages that describe new theories, concepts and developments made by the authors in any branch of science and technology. Notes need not contain results from experimental or simulation work.

News and Views: The News section provides a space for articles up to three journal pages in length describing leading developments in any field of science and technology or for reporting items such as conference reports. The Editor reserves the right to modify or reject articles for consideration as 'news items'.

Commentaries: Upon Editor's invitation, commentaries discuss a paper published in a specific issue and should set the problems addressed by the paper in the wider context of the field. Proposals for Commentaries may be submitted; however, in this case authors should only send an outline of the proposed paper for initial consideration. The contents of the commentaries should follow the following set of rules: 3000 words maximum, title 20 words maximum, references 10 maximum (including the article discussed) and figures/tables 2 maximum.

Errata: Xjenza also publishes errata, in which authors correct significant errors of substance in their published manuscripts. The title should read: Erratum: "Original title" by ***, Xjenza, vol. *** (year). Errata should be short and consistent for clarity.

Invited Articles and Special Issues: Xjenza regularly publishes Invited Articles and Special Issues that consist of articles written on invitation by the Editor or member of the editorial board.

Submission of Manuscripts

Manuscripts should be sent according to the guidelines given hereafter to submissionxjenzaonline@gmail.com.

Referees All manuscripts submitted to Xjenza are peer reviewed. Authors are requested to submit with their manuscript the names and addresses of three referees, preferably from overseas. Every effort will be made to use the recommended reviewers; however the editor reserves the right to also consult other competent reviewers.

Conflict of Interest Authors are expected to disclose any commercial or other associations that could pose a conflict of interest in connection with the submitted manuscript. All funding sources supporting the work, and institutional or corporate affiliations of the authors, should be acknowledged on the title page or at the end of the article.

Policy and Ethics The work described in the submitted manuscript must have been carried out in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans (`http://www.wma.net/en/30publications/10policies/b3/index.html`); EU Directive 2010/63/EU for animal experiments (`http://ec.europa.eu/environment/chemicals/lab_animals/legislation_en.htm`); Uniform Requirements for manuscripts submitted to Biomedical journals (`http://www.icmje.org`). This must be stated at an appropriate point in the article.

Submission, Declaration and Verification Submission of a manuscript implies that the work described has not been published previously (except in the form of an abstract or as part of a published lecture or academic thesis), that it is not under consideration for publication elsewhere, that it has been approved for publication by all authors, and tacitly or explicitly, by the responsible authorities where the work was carried out, and that, if accepted, it will not be published elsewhere in the same form, in English or in any other language, including electronically, without the written consent of the copyright-holder.

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Preparation of Manuscripts

Xjenza accepts submissions in MS Word, Libre Office Writer and LaTeX with the latter being the preferred option. Anyone submitting in LaTeX should use the journal template, the latest version of which can be found at `http://github.com/hicklin/Xjenza-Journal-Template`. All the necessary files to run the LaTeX document should be supplied together with the rendered PDF.

If a word processor is used the styling should be kept to a minimum, only introducing bold face, italics, subscript and superscript text where the context requires it. Text should be in single-column format and the word processor options should not be used in order to justify text or hyphenate words. Together with the native format of the word processor, a pdf, generated by the word processor, must be given. Furthermore, artwork should be in accordance to the artwork guidelines give below and must be submitted separately from the word processor file. Similarly, the bibliographic data of the cited material should be submitted separately as an Endnote (*.xml), Research Information Systems (*.ris), Zotero Library (zotero.splite) or a BiBTeX (*.bib) file.

Article Structure

A manuscript for publication in Xjenza will ordinarily consist of the following: Title page with contact information, Abstract, Highlights, Keywords, Abbreviations, Introduction, Materials and Methods, Results, Discussion, Conclusion, Appendices and References.

The manuscript will be divided into clearly defined numbered sections. Each numbered subsection should be given a brief heading. Each heading should appear on its own separate line. Subsections should be used as much as possible when cross-referencing text: refer to the subsection by the section number as opposed to simply 'the text'.

Title page

- Title should be concise yet informative. Titles are often used in information-retrieval systems. Avoid abbreviations and formulae where possible.
- Author names and affiliations. Present the authors' affiliation addresses (where the actual work was done) below the names. Indicate all affiliations with a lower-case superscript number immediately after each author's name and in front of the appropriate address. Provide full postal address of each affiliation, including the country name and, if available, the e-mail address.
- Corresponding author. Clearly indicate who will handle correspondence at all stages of refereeing and publication, including post-publication. Ensure that telephone and fax numbers (with country and area code) are provided in addition to the e-mail address and complete postal address. Contact details must be kept up to date by the corresponding author.
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Abstract A concise and factual abstract is required of up to about 250 words. The abstract should state briefly the background and purpose of the research, the principal results and major conclusions. An abstract is often presented separately from the article, so it must be able to stand alone. For this reason, references and non-standard abbreviations should be avoided. If essential, these must be defined at first mention in the abstract itself.

Abbreviations Define abbreviations that are not standard in this field in a footnote to be placed on the first page of the article. Such abbreviations that are unavoidable in the abstract must be defined at their first mention as well as in the footnote and should be used consistenly throughout the text.

Introduction State the objectives of the work and provide an adequate background, avoid a detailed literature survey or a summary of the results.

Materials and Methods Provide sufficient detail to allow the work to be reproduced. Methods already published should be indicated by a reference: only relevant modifications should be described.

Results Results should be clear and concise. Numbered/tabulated information and/or figures should also be included.

Discussion This should explore the significance of the results of the work, yet not repeat them. Avoid extensive citations and discussion of published literature. A combined section of Results and Discussion is often appropriate.

Conclusion The main conclusions based on results of the study may be presented in a short Conclusions section. This may stand alone or form a subsection of a Discussion or Results and Discussion section.

Appendices Formulae and equations in appendices should be given separate numbering: Eq. (A.1), Eq. (A.2), etc.; in a subsequent appendix, Eq. (B.1) and so on. Similarly for tables and figures: Table A.1; Fig. A.1, etc.

Acknowledgements Collate acknowledgements in a separate section at the end of the article before the references and do not, therefore, include them on the title page, as a footnote to the title or otherwise. List here those individuals who provided assistance during the research (e.g., providing language help, writing assistance or proof reading the article, etc.).

Units Follow internationally accepted rules and conventions: use the international system of units (SI). If other units are mentioned, please give their equivalent in SI. Anyone using LaTeX should use the package siunitx in all cases.

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Table Footnotes Indicate each footnote in a table with a superscript lower case letter.

Artwork Electronic artwork General points:

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Tables Number tables consecutively in accordance with their appearance in the text. Place footnotes to tables below the table body and indicate them with superscript lowercase letters. Avoid vertical rules. Be sparing in the use of tables and ensure that the data presented in tables do not duplicate results described elsewhere in the article. Large tables should be submitted in CSV format.

Citations and References Reference and citation styles for manuscripts submitted to Xjenza should be in accordance to the APA v6 style.

Citation in text References to cited literature in the text should be given in the form of an author's surname and the year of publication of the paper with the addition of a letter for references to several publications of the author in the same year. For further information regarding multiple authors consult the APA v6 guidelines. Citations may be made directly

Kramer et al. (2010) have recently shown ...

or parenthetically

as demonstrated (Allan, 2000a, 2000b, 1999; Allan and Jones, 1999).

Groups of references should be listed first alphabetically, then chronologically. When writing in LaTeX use `\textcite{}` and `\parencite{}` for the respective cases mentioned.

The reference section Every reference cited in the text should also be present in the reference list (and vice versa). The reference list should also be supplied as an Endnote (*.xml), Research Information Systems (*.ris), Zotero Library (zotero.splite) or a BiBTeX (*.bib) file. Unpublished results and personal communications are not recommended in the reference list, but may be mentioned in the text. If these references are included in the reference list they should follow the standard reference style of the journal and should include a substitution of the publication date with either 'Unpublished results' or 'Personal communication'. Citation of a reference as 'in press' implies that the item has been accepted for publication.

References should be arranged first alphabetically and then further sorted chronologically if necessary. More than one reference from the same author(s) in the same year must be identified by the letters 'a', 'b', 'c', etc., placed after the year of publication. Consult the APA v6 guidelines for multiple authors. Below are some examples of referencing different bibliographic material.

Reference to a Journal Publication:

Agree, E. M. and Freedman, V. A. (2011). A Quality-of-Life Scale for Assistive Technology: Results of a Pilot Study of Aging and Technology. *Phys. Ther.*, 91(12):1780–1788.

McCreadie, C. and Tinker, A. (2005). The acceptability of assistive technology to older people. *Ageing Soc.*, 25(1):91–110.

Reference to a Book:

Brownsell, B. (2003). *Assistive Technology and Telecare: Forging Solutions for Independent Living.* Policy Press, Bristol.

Fisk, M. J. (2003). *Social Alarms to Telecare: Older People's Services in Transition.* Policy Press, Bristol, 1st edition.

Reference to a Chapter in an Edited Book:

Brownsell, S. and Bradley, D. (2003). New Generations of Telecare Equipment. In *Assist. Technol. Telecare Forg. Solut. Indep. Living*, pages 39–50.

Web references The full URL should be given together with the date the reference was last accessed. Any further information, if known (DOI, author names, dates, reference to a source publication, etc.), should also be given. Web references can be listed separately or can be included in the reference list.

References in a Special Issue Please ensure that the words 'this issue' are added to any references in the list (and any citations in the text) to other articles in the same Special Issue.

Journal Abbreviations Journal names should be abbreviated according to:

-Index Medicus journal abbreviations: `http://www.nlm.nih.gov/tsd/serials/lji.html`;

-List of title word abbreviations: `http://www.issn.org/2-22661-LTWA-online.php`;

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Submission check list

The following list will be useful during the final checking of a manuscript prior to sending it to the journal for review. Please consult this Guide for Authors for further details of any item.

- One author has been designated as the corresponding author with contact details:
  - E-mail address.
  - Full postal address.
  - Telephone and fax numbers.
- All necessary files have been sent, and contain:
  - All figures are given separately in PDF, SVG, JPEG of PNG format.
  - Caption for figures is included at the end of the text.
  - All tables (including title, description, footnotes) are included in the text and large tables have been given separately as CSV.
  - The reference list has been given in XML, RIS, zotero.splite or BIB file format.
- Further considerations
  - Abstract does not exceed about 250 words.
  - Manuscript has been 'spell-checked' and 'grammar-checked'.

- References are in the required format.
- All references mentioned in the reference list are cited in the text, and vice versa.
- Bibliographic data for all cited material has been given.
- Permission has been obtained for use of copyrighted material from other sources (including the Web).
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Special Editorial

COST Special Issue

David C. Magri*[1], Janet Mifsud[2]
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In this special issue of *Xjenza Online*, presented are papers by scientists based in Malta, notably from the University of Malta, with collaborators from institutions in Europe that have participated in European Cooperation in Science and Technology (COST) actions. COST is Europe's longest-running intergovernmental framework for cooperation in science and technology. Founded in 1971, the mission of COST is to "enable breakthrough scientific developments leading to new concepts and products and thereby contribute to strengthen Europe's research and innovation capacities."

The COST Actions connect scientific researchers across disciplines from across Europe and the world. They provide networking opportunities for early career investigators; increase the impact of research on policy makers, regulatory bodies and national decision makers as well as the private sector. Through its inclusiveness, particularly the participation of women researchers, COST actions support integration of research communities.

This special issue of *Xjenza Online* showcases 9 articles. The first article, by Janet Mifsud, provides some history and background on COST. The paper highlights the increased participation of Malta in these actions, which has steadily grown from 47 researchers in 2011 to 149 in 2014. The other contributions are on topics associated with specific COST Actions.

Ulrich Baisch presents a paper on the Lanthanide and Actinide Chemistry in the European f-Element Network (EUFEN). As part of EUFEN (European f-Element Network), researchers are investigating crystal engineering of f-block elements. These elements are nowadays components in many advanced materials used in medical imaging, electronic superconductors, lasers, phosphors, LEDs, catalysis and energy storage.

David C. Magri, Guest editor, *Xjenza Online*
Associate Editor, Physics and Chemical Sciences

Pierre Schembri-Wismayer highlights CM1106 STEMCHEM, which aims to bring expertise together on rational drug design and medicinal chemistry in order to understand the mechanism of action of drug resistance in stem cells. Furthermore, the action aims to develop new methods for identifying drug candidates that specifically target drug-resistant cancer cells.

Thérèse Bajada examines the accessibility of public transport and the Malta bus service for the elderly while Carl James Debono is involved in an action bringing together researchers from all areas of 3D technology

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Janet Mifsud, Guest editor, *Xjenza Online*

including 3D video broadcasting and streaming. Emmanuel Buttigieg has contributed an article from COST action IS1205 on how history and social psysiology to seek ways past histories are understood by ordinary people. The action strives to advance knowledge on the role played by social representation of history from the point of view of ethnic and national groups and the role intergroup conflicts play within the European Union.

Daniel Sultana focused on an issue of a more local nature that of soil erosion in Malta. Quantitative models are used to analysis the capacity of soil erosion and recommendations are put forward to build and maintain control structures such as terraced field rubble walls while Flaminio Squazzoni is participating in the action PEERE, which aims at reviewing the various peer-review models. Representation is broad across disciplines with 31 countries represented. The aim is to increase the credibility of science by qualitative and quantitative analysis of different peer-review models.

The last two contributions are related to CMST COST Action CM1103 "Structure-based drug design for diagnosis and treatment of neurological diseases: dissecting and modulating complex function in the monoaminergic systems of the brain". The first is a Meeting Report by Rona R. Ramsay and Philippe De Deurwaerdere on the third Annual Conference hosted by the University of Bordeaux, France on 8–10 October 2014. The last contribution are the conference proceedings of the "Interdisciplinary Chemical Approaches for Neuropathology CM1103/4th Neuroscience Day University of Malta" hosted in Valletta at the Old University in October 2013. Dr Rona Ramsay, the Chair of the Action from the University of St Andrews, UK, and Prof. Di Giovanni put together an exciting program that covered a full range of interests, promoted the exchange of expertise between the various areas of structural-based drug design, synthetic chemistry and neuropathology that can all contribute information to diagnosis and treatment methods. This conference brought together international experts and Maltese scientists, fostering collaborations and dissemination of the work done in Europe.

Let us end by thanking all the contributing authors and encouraging researchers who have yet to participate in a COST Action to sign up. You will have the opportunity to attend an annual conference and meet experts with interests related to yours. You might even have an adventure, or two, and experience some serendipity along the way, which is a key element in making a new discovery.

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Mifsud, J. (2017). *Xjenza Online*, 5 Special Issue:3–7.

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Special Issue Introductory Article

Malta's participation in Europe's oldest research cooperation framework

Janet Mifsud
COST National Contact Point, COST Malta Representative on the Committee of Scientific Officers, Malta Council for Science and Technology, Kalkara

Abstract. COST (European Cooperation in Science and Technology) is the longest-running European research framework supporting networking and knowledge sharing amongst science and technology communities in Europe. It does not fund research as such but greatly facilitates the networking and the coordination between researchers working on nationally funded research across Europe. The wide range of networking activities supported by COST, such as meetings, workshops, short term scientific missions, publications and training schools and easy administration made it even very amenable to researchers based in Malta. Since 2011 Malta has participated in a constantly growing number of running COST Actions and this has resulted in better exposure opportunities for them. More importantly, early stage researchers have been given an invaluable opportunity to increase their contacts circles in their fields of interest. This has resulted in unique opportunities for participation and networking in research for researchers based in Malta on a European scale.

1 Introduction

This issue of *Xjenza* may seem somewhat eclectic, including many diverse and unrelated topics. Many may wonder what does research concerning supramolecular chemistry in water has to do with drug resistance in cancer stem cells, soil erosion and 3D video coding? Moreover, are studies on the symbiotics of history and social psychology, and research on accessibility as an indicator of transport equity considered as science?

The brief answer to these questions is **COST**. This special issue of Xjenza highlights just some of the research that has resulted from the nearly 200 COST actions, researchers based in Malta are or have participated in. You others may wonder what exactly COST is? How can I get involved? How will it benefit myself and the research landscape in Malta?

2 What is COST?

COST (European Cooperation in Science and Technology) (www.cost.eu) is in fact the longest-running European research framework supporting networking and knowledge sharing amongst science and technology communities in Europe (Halen, 2014). It does not fund research as such but greatly facilitates the networking and the coordination between researchers working on nationally funded research across Europe. Thus it contributes greatly to narrowing the gap between science, policy makers and society. **Co-operation in COST is inclusive and open, fosters new and excellent ideas through the sharing of knowledge** (see Figure 1).

Since its establishment in 1971, COST has been doing this through supporting networks (called **COST Actions**) co-ordinated by Management Committee members (MCs). A huge advantage of these **MCs** is that each COST country is allowed to nominate up to two MCs per action irrespective of size, thus ensuring inclusiveness and widening. This is a big advantage for Malta. As such, COST has always been the cornerstone of the European research funding landscape since it opens up huge networks to smaller and more peripheral COST countries, such as Malta. Every year an average of 30 000 researchers benefit from COST funding (Dietl, 2014).

Over the course of its 40 year history, COST has also been able to constantly adapt itself to the changing environment defining research policy in Europe. It has a unique bottom up approach, using open calls with no pre-defined priorities for research. It also promotes

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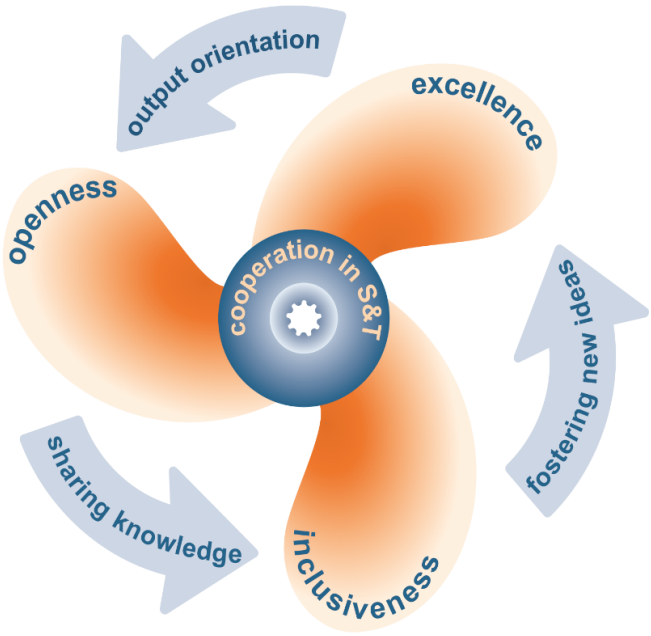

Figure 1: Co-operation in COST is inclusive and open, fostering new and excellent ideas through the sharing of knowledge.

interdisciplinary approaches, knowledge sharing, talent development and assists research communities in finding their own ways to tackle important societal challenges.

Since its inception, COST has provided a unique approach that enables the sharing of concepts and scientific developments across 35 European countries and has even attracted the participation of many non EU countries.

There are three main features that make it a unique tool:

Capacity COST acts to connect high-quality scientific minds both across Europe and internationally and enables the creation of research communities in various fields.

Networking As a networking platform, it supports researchers to connect and build consortia that can lead to the submission of transnational project proposals for funding from the EU's Framework Programme. This networking is interdisciplinary, facilitating the participation from researchers outside the academic community, such as SMEs, public entities and NGOs.

Impact COST enables the formulation of publications and promotes the dissemination of information that increases impact on policy-makers and decision-making bodies (Armeni and Mifsud, 2015).

COST has also always been a frontrunner in bridging the gap between policy, research and end users, by promoting connections between COST Actions and other European S&T policy concepts with relevant stakeholder communities. It has provided the basis for increased societal impact of research and S&T innovation in Europe and beyond.

3 Malta in COST

Malta first joined COST in 1996 before it became a full EU member in 2004. The Malta Council for Science and Technology, as the managing authority for COST in Malta, then recognised the key role COST participation could have in facilitating the entry of researchers based in Malta to wider European Research networks and funds such as in the Framework Programmes. Moreover the wide range of networking activities (tools) supported by COST, such as meetings, workshops, short term scientific missions, publications, and training schools, made it even more amenable to researchers based in Malta.

Since 2011 Malta has participated in a constantly growing number of running COST Actions. In 2014, the Maltese research community not only participated in the Management Committee (MC) of 149 Actions (Table 1) but had access to all their networking activities (Halen, 2015).

The number of Maltese participations to networking activities (meetings, workshops, STSM, Training Schools) has also been growing since 2011 (Table 1). In 2014, Maltese researchers participated in 200 COST Action activities.

COST also promotes the participation of Early Career Investigators - ECI (less than PhD + 8 years) in all of its networking activities (meetings, workshops, STSM, Training Schools). In 2014, almost half of the Maltese participations in networking activities (as shown in the previous Table 1) were from Early Career Investigators-ECI (Table 1).

The Maltese research and innovation system benefits more and more from COST Actions' budget. In fact in 2014, around EUR 188,000 benefited Maltese researchers (Table 1). This includes participations in meetings, workshops, STSM, Training Schools, Local Organiser Support, but also Maltese Grant Holder institutions' administrative support – FSAC.

In 2014, Maltese researchers participated in 15 Short Term Scientific Missions (STSMs). Thirteen Maltese trainees and one Maltese trainer were also involved in training schools (Table 1).

Maltese institutions also gained increasing visibility thanks to meetings, Training Schools and STSMs being hosted in their premises. In 2014, 4 COST meetings, 1 Training School and 5 STSMs were held in Malta helping to strengthen the existing networks and foster collaboration links between researchers, institutions and ultimately countries. They also helped to increase the

Year	2011	2012	2013	2014
COST actions with participation from Malta	47	70	116	149
Maltese participation in COST networking activities	46	75	161	200
Maltese early career investigators participation in COST networking activities	18	25	60	82
COST networking budget transferred to Malta (in euros)	42 233	72 674	141 883	188 015
COST Short Term Scientific Missions (STSM) and training school participation from Malta				
STSM participants	4	3	4	15
Trainees in Training Schools		3	13	13
Trainers in Training Schools		1	1	1
COST meetings, training schools, STSMs and Annual Progress Conference (APCs) organised in Malta				
Meetings	1	1	3	4
Training Schools	–	1	2	1
STSM	2	1	7	5
APC	–	–	1	4

Table 1: COST action statistics (Halen, 2015).

visibility of hosting institutions. Maltese institutions seem to be particularly attractive to researchers coming on STSMs (Table 1). In addition, Malta also had the honour to host five Annual Progress Conferences (APCs) from five of the ten COST domains in 2013 and 2014.

4 COST impact in Malta

In 2014, MCST carried out a survey among Malta- based COST MC members in order to gauge the success, failure and impact of COST since its initiation in Malta, aiming to identify the resulting impact of this participation in COST on the local research arena and innovation initiatives (Armeni & Mifsud, 2014).

5 Who is the Maltese COST researcher?

The survey was carried out by means of an online questionnaire using Survey Monkey®. There were a total number of 100 responses, from 193 potential respondents that were participating in COST actions across Malta and Gozo at the time the survey was taken. As also confirmed by the data provided in Table 1, the majority of respondents, 37.10 %, were between 30 and 39 years of age, followed by 25.81 % between 40 and 49 years of age, 16.94 % between 50 and 59 years of age and 13.71 % between 21 and 29 years of age. Only 3.23 % noted that they were 60 years of age or older (Table 1). In this respect COST has reached out to and involved comparatively high numbers of young researchers below the age of 40 in the majority of its domains. Such capacity building and opportunities for early stage researchers, who are key human resources, will thus contribute to a continued growth of innovation, product development and commercialisation in Malta.

In terms of career status, most respondents noted that they hold the position of senior lecturers (27.42 %), whereas 19.35 % noted that they are lecturers, 9.68 % noted that they are associate professors and 3.23 % noted that they are professors. There was a positive uptake from PhD students (13.71 %) and post-doctorate researchers (7.26 %) indicating an increasing interest by the younger population of researchers across the domains and the involvement and integration of a more juvenile cohort of academics and scientists in the various research agendas of the different COST actions.

Significantly lower were percentages for participation from industry (1.61 %) and NGO's (4.03 %). In addition, there is very limited involvement reported from the public entities and the public sector suggesting that more efforts could be focused to generate awareness and interest there. Academia continues to enjoy a strong hold in the programme. The lack of participation from industry could imply that COST is not yet understood by Maltese industry as potentially being a significant contributor towards their participation in transnational consortia or in research arenas. Increased efforts, directed at this particular sector, could result in increased participation when relevant domains/actions arise.

6 What are the outcomes of participation in COST?

Most respondents stated that their COST action members intended to continue to co-operate once the COST

action was completed (80 %). Nonetheless, some commented that any speculation of potential future action, at present, would be premature (29 %). An equal amount simply noted 'future collaborations', such as through access to infrastructures and joint research activities, as possible sources of action (29 %) Others highlighted future research contributions or submissions under Horizon 2020 as a possible way forward.

7 What can be done to improve Malta COST participation?

Participation in COST actions has had its benefits and difficulties. The main benefits noted by the respondents were: networking, opportunities for cooperation and collaboration with European counterparts, increased exposure at a European level, opportunities for training and staff/student exchange and access to research infrastructures. Furthermore, the majority of respondents who claimed prior participation in, or organisation of, COST events, both locally and abroad, noted that the output and impact was well worth the return on investment, particularly in terms of time and effort.

The main obstacles were: financial and administrative burdens, lack of time and resources, bureaucracy, language barriers, travel requirements and other work. In this respect, it was noted that the provision of more frequent information sessions and assistance in sourcing and joining existing consortia, coupled with more hands on guidance in the compilation and submission of research proposals for Horizon 2020 would be beneficial. This, as well as, funding to conduct further research remains a key concern for most respondents. Notably, lack of financial support implies lack of advancements in research and analytics in each domain which could place Malta at a disadvantage when compared to other countries. For this purpose, some respondents suggested a potential allocation of some resources that may act as an incentive for local researchers to construe research groups in areas that are deemed relevant to work upon. The areas may not have direct relevance to Malta, but Malta can act as a cost effective base where research can be carried out, generating income and knowledge. In addition, continued efforts towards increasing opportunities for the mobility of researchers across Europe, to foster scientific excellence, is recommended together with an alignment of the selection of local participation in specific COST actions with the national scientific priorities' as outlined in the National Research and Innovation Strategy 2020 (Malta Council for Science and Technology, 2014).

In addition, the majority cohort of academic senior lecturers as MC members and the lack of participation from industry, raises concerns, particularly, in view of the need to bridge the gap between research in academia and industry. Lack of easily accessible information, easier access to academics, lack of available time and resources and lack of tangible and/or immediate return on investment could be precluding players from industry from actively participating in COST. To this end, one-to-one meetings, networking events, information sessions and the CNC's or MCST staff participation in events organized by other bodies representing industry and young researchers, could be beneficial. In addition, in line with efforts to mediate gender equality and balance in scientific spheres, it is advised that more emphasis is made on mediating the current gender balance in COST actions

8 COST: the next steps

COST has been proud of its ability of renewal. The renewal started in 2010, in response to the COST FP7 Mid-Term Evaluation (Horvat et al., 2010) and through the COST Strategy (COST CSO, 2011) aims to achieve best performance, output orientation, better cooperation, and good governance. The new phase has been characterised by reviewing the institutional, operational and administrative levels of the COST Framework and the creation of a new independent legal entity, the '**COST Association**', tasked with becoming COST's implementing agent in 2014 (COST CSO, 2013).

Over the last few years, COST has also adapted and changed to respond to the requirements and challenges in Horizon 2020 ensuring that COST actions are focussed on Spreading excellence and widening participation as well as ensuring Europe in a changing world – inclusive, innovative and reflective societies (COST CSO, 2014). Adaptations have always been made keeping in mind the above principles that govern the COST Framework from both a strategic and operational point of view: **supporting excellence** and being **open and inclusive**. The driving forces (deriving from the principles described above) of **fostering new ideas, sharing knowledge and output orientation** have been enabling COST to make significant contributions to the competitiveness and overall development of the European research landscape and assisting European research communities in overcoming the many challenges they face and thereby contributing to Europe's Innovation Union goals.

Researchers based in Malta are now integrated in each part of the COST process, from participation in the high level policy Committee of Scientific Officers (CSO), having a representative on the strategic Committee of Scientific Experts, a full cohort of eighteen Review Panel Experts and also close to 300 COST experts. Malta's success in COST has often been highlighted by the COST Association and Malta has even been invited to be Vice-Chair for the COST working

group on Inclusiveness and Widening.

Malta's participation and investment in COST has resulted in better exposure opportunities for various parties. More importantly, the younger cohort of researchers have been given an invaluable opportunity to increase their contacts circles in their fields of interest. This, coupled with opportunities for participation in research on a European scale, opportunities for specialisation through joint collaborations and opportunities to showcase our local potential, continue to support and advocate Malta's necessity to participate in this programme and acts as an incentive for local participation to continue to be supported and encouraged.

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Review Article

Connecting Frontier Research with Industrial Development - Lanthanide and Actinide Chemistry in the European f-Element Network (EUFEN)

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Abstract. The chemistry of f-elements, even though often not known to the general public, forms part of many industrial processes, manufacturing and medical applications, such as medical imaging (e.g. MRI scans), strong magnets, data storage media, superconducters, LEDs, catalysis, as well as energy, and metal extraction. It plays a crucial role in the scientific and industrial landscape of the European Union (EU) in fields of energy, security, training, sustainability, and society. The use of these elements widens the scope of synthetic possibilities in chemistry, and materials with outstanding electromagnetic properties have already been realised. The synthesis of lanthanide containing supramolecular materials with exceptional materials properties has already been reported, e.g. the ability to bind and release gases, high-temperature superconductivity, and all-white light emitting diodes. The EUFEN (European f-Element Network) COST action provides cooperative mobility mechanisms for nationally funded f-element chemists pursuing fundamental frontier research to initiate collaborations, training, networking, and dissemination among each other. Novel developments and results in terms of f-element crystal engineering carried out at the University of Malta are therefore part of EUFEN.

1 Introduction

EUFEN, the European f-Element Network is a COST action which provides cooperative mobility mechanisms for f-element chemists from all over Europe for pursuing fundamental frontier research to initiate collaborations, training, networking, and dissemination with each other (COST, 2014).

Two goals for the action have been identified: (i) to tackle unsolved problems in f-element chemistry and (ii) to supply industry with trained researchers from universities, restructuring the EU science-base, and thus result in a research output that is greater than the sum of its parts.

Although already more than 200 years old, the chemistry of f-elements (a block of 28 elements at the bottom of the periodic table) can still be considered as one the of areas of chemistry that are least known by the general public. Hardly anybody outside the field of inorganic chemistry will have read or heard of "f-elements" as well as groups of elements called lanthanides, actinides or rare earths; a good example are the actinide elements uranium or plutonium: even though one might know these elements very well in the context of radioactivity or nuclear energy, only a small group of people would know that uranium and plutonium are part of the group of elements in the periodic table called f-elements and that there is a surprisingly rich amount of synthetic chemistry of e.g. uranium and thorium complexes published every year (Bünzli, 2006); a search with the program SciFinder using the key words thorium or uranium complex resulted in over 5000 publications since 2000.

The innocence of the existence of a whole group of 28 elements is somewhat surprising as f-elements (in particular lanthanides) are indispensable components in many materials everybody uses day by day. f-Element chemistry contributes to medical imaging (MRI contrast agents), magnetic (strong magnets), electronic (superconductors), and photonic devices (lasers, phosphors, displays, LEDs), catalysis, energy, and metal extraction and is therefore strategically crucial to science, energy, security, training, sustainability, and society (Blake et al., 1999; Bünzli, 2006; Bünzli & Piguet, 2002; Edelmann, 2009). This wide range of applications is the

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result of the particular electromagnetic properties of f-elements. Often just a very small percentage of f-element (usually between 0.01 and 10 %) needs to be present in a material to change its optical, electrical or magnetic properties completely.

The synthesis of lanthanide containing supramolecular materials with exceptional materials properties have been discovered recently, e.g. the ability to bind and release gases, superconductivity, and luminescence (Blake et al., 1999).

In addition to the general lack of knowledge about the existence of these elements in public, there are also a number of ways to name certain groups of f-elements and other related elements in the periodic table. Elements called lanthanides and actinides can either i) be the row of elements from lanthanum (La) to ytterbium (Yb) and actinium (Ac) to nobelium (No) or ii) include also the elements lutetium (Lu) and (Lr) lawrencium, respectively. However, according to the conventions set by the International Union of Pure and Applied Chemistry (IUPAC), the latter (ii) would refer to these groups as lanthan**oi**ds and actin**oi**ds rather than lanthanides and actinides. Some sources also state that the group of lanthanides consists of the group of elements from cerium (Ce) to lutetium (Lu) (Bünzli, 2006). Rare earths is also an expression which is often used for the group of 4f and 5f-elements. This group consists not only of 4f and 5f elements, but it includes also other elements from Group 3 of the periodic system, namely yttrium (Y) and scandium (Sc).

In view of their large ionic radius and their particular electronic configuration, f-element based research can be extremely challenging from a synthetic point of view. Scientific exchange and collaboration are therefore essential in this field of research. EUFEN offered for the first time a platform for f-element researchers from all over Europe to join forces and tackle the pending questions, such as: What are the principles/mechanisms behind the use of lanthanide complexes as catalysts? How can f-element compounds be treated in computational chemistry and how can we predict the formation of new complex compounds (COST, 2014)?

Due to their special physical and chemical properties, rare earth elements were almost unused and less popular in the field of crystal engineering (Broker, Klingshirn & Rogers, 2002). Novel developments and results in terms of f-element crystal engineering are therefore also an important part of EUFEN.

As EUFEN is the first and only collaborative research network in the field of f-element chemistry, a very large part of the European community of f-element scientists is involved. Over 120 researchers from 24 nations form part of this COST action. Three main working groups were set up (COST, 2014):

a) Synthesis and Structure: the aim is to undertake the synthesis, structures, and chemical bonding in new f-element compounds, with the scope of discovering novel structures and bonding and rationalising the observed phenomena.

b) Spectroscopy and Computation: the aim is to focus on exploiting existing and new f-element compounds for novel spectroscopic and computational investigations which hitherto have no precedent.

c) Applications: the aim is to explore existing and new chemical bonding and reactivity, or other phenomena, with a view to ultimately delivering research which can benefit EU science, industry, and society as opportunities to exploit new findings become available.

Three different major published outcomes of collaborative EUFEN research will be described below in order to showcase the COST action. Subsequently, our own scientific contribution will be summarised.

2 Supramolecular Sensors

One of the most prominent properties of both 4f- and 5f-elements is their large atomic or ionic size. As a consequence their metal ions can usually coordinate to a higher amount of ligands (coordination numbers vary between 7 and 12) than is usually the case for transition metals. The geometries around the f-element centre are less restricted and this makes 4f-element complexes in particular a well-known component in supramolecular chemistry.

Three research participants of EUFEN (Universities of Strasbourg and Bretagne Occidentale, France; University of Coruña, Spain) collaborated on a project which examined the effect of fluoride addition to aqueous solutions of luminescent lanthanide complexes (Ln = Eu, Tb, Yb).(Liu et al., 2014) Upon addition, the formation of a dimeric europium complex **Eu-1** was observed. X-ray crystal structure determination and luminescence spectroscopy revealed not only the geometry of the complexes but also that i) fluoride anions were confined into the cavity that was formed by the two complex molecules and ii) luminescence intensity increased significantly. Synergistic effects of the Eu-F-Eu bridging motif, π stacking interactions, and a four-component hydrogen-bonding network which control the assembly of the two complex molecules around the fluoride ion, are crucial for the above (see Figure 1).

The exact sensing of fluoride in aqueous solutions or solvent mixtures is of great importance for public health (Liu et al., 2014). Fluoride in small quantities can have a positive impact on teeth and bones, whereas higher concentrations of fluoride can result in serious health problems. According to the World Health Organisation the fluoride content in drinking water should be lower than 1.5 ppm (World Health Organization, 2004). How-

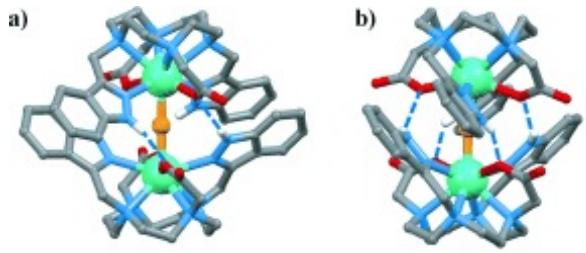

Figure 1: Crystal structure of the **Eu-1** dimer viewed perpendicular to (a and b) the main pseudo-C_2 axis.(Liu et al., 2014)

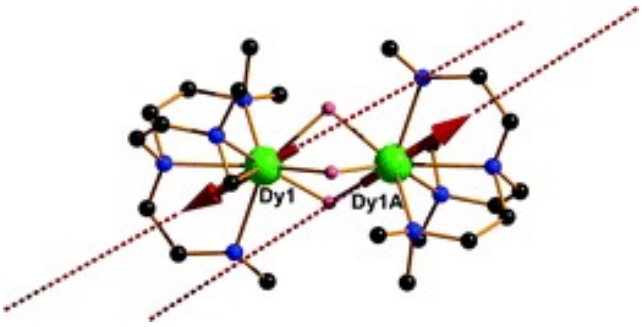

Figure 2: Orientation of the magnetic moments in **Ln-2** (dashed lines). The arrows show the antiferromagnetic coupling. Pink atoms = hydride atoms, blue = N atoms, grey = C atoms (Venugopal et al., 2013).

ever, so far there are just a few procedures known to quantitatively analyse the fluoride content in aqueous solutions.

The authors' new approach is to use the lanthanide complexes described above to sense the fluoride content in aqueous solutions using fluorescence spectroscopy (Liu et al., 2014).

3 Single Molecule Magnets

Single-molecule magnets (SMMs) are metal-organic compounds that exhibit paramagnetic behaviour on a molecular level. They can be described as the smallest possible magnetic devices. Potential applications of SMMs are quantum computing, high-density information storage and magnetic memory devices. Lanthanide-SMMs play a leading role in this field of research as the presence of 4f-elements in a complex molecule alters the magnetic behaviour drastically due to the presence of 4f-electrons and thus show a particularly high magnetic moment and single ion anisotropy compared to transition metal SMMs (Christou, Gatteschi, Hendrickson & Sessoli, 2000). However, also systematic research in the field of actinoid (5f) based SMMs emerged in the literature, recently. The outcomes discussed in these papers have resulted from STSMs (Short-Term Scientific Mission) of EUFEN (Liddle, Mills & Wooles, 2010, 2011; Meihaus & Long, 2015; Mougel et al., 2012, 2012).

Okuda (RWTH Aachen, Germany), Layfield (University of Manchester, UK) and coworkers have collaborated within EUFEN and carried out an experimental and ab initio computational study of an asymmetrical, hydride-bridged di-dysprosium single-molecule magnet (Venugopal et al., 2013).

Hydride ligands can be of particular interest for the development of SMMs as they exhibit very strong ligand-field effects. Subsequently, this could have a different effect on the relaxation times in Ln-SMMs compared to metal-organic compounds with oxygen-donor ligands (Luzon & Sessoli, 2012; Rinehart & Long, 2011; Sorace, Benelli & Gatteschi, 2011).

The reported compounds in this study were the first hydride-ligated SMMs. The compounds consist of hydride bridged complexes $[Ln(Me_6trenCH_2)(\mu{-}H)_3Ln(Me_6tren)][BC6H3(CF3)2_4]_2$ **Ln-2**,where Ln = Gd(III), Dy(III) and Me_6tren = *tris*[2-(dimethylamino)ethyl]amine (see Figure 2).

4 Uranium(VI)–nitride triple bond

The chemistry of uranium is not only related to its role as a radioactive element, but it is in fact an element well known for its extraordinary coordination chemistry (Bart & Meyer, 2008; Hayton, 2010; King & Liddle, 2014; Lu, 2014; Van Horn & Huang, 2006). Uranium can exist in different oxidation states and can form complexes with high coordination numbers around the actinide (5f) centre due to its large atomic and ionic size.

Whereas the bonding of ligands to 4f elements (lanthanides) is generally known to be mainly ionic, the nature of 5f-element (actinides) ligand bonds is still discussed in recent literature. One of the most controversially analysed aspects of this is the existence of uranium-ligand multiple bonds (Baker, 2012; Bart & Meyer, 2008; Hayton, 2010; King & Liddle, 2014).

During the past years uranium(V)-OR and –NR were reported (R = alkyl group) and thus, the synthesis of a uranium-ligand triple bond was one of the hot topics of EUFEN as it is of fundamental importance to the study of f-orbital participation in metal-ligand multiple bonding (Hayton, 2013).

It was therefore a big break-through when Liddle (University of Nottingham, UK), McInnes (University of Manchester, UK) and coworkers reported the preparation of a complex with a uranium(VI)–nitride triple bond: the terminal uranium(V) nitride complex $[UN(TrenTIPS)][Na(12\text{-crown-}4)_2]$ (TrenTIPS = $[N(CH_2CH_2NSiiPr_3)_3]_3$ - and ^{i}Pr = isopropyl) **U-3**. Synthesis was achieved as summarized in Figure 3 (King et al., 2012, 2013).

The existence of a triple bond was proven by single-crystal X-ray diffraction. The uranium-terminal nitride bond length of 1.825 Å is significantly shorter than the

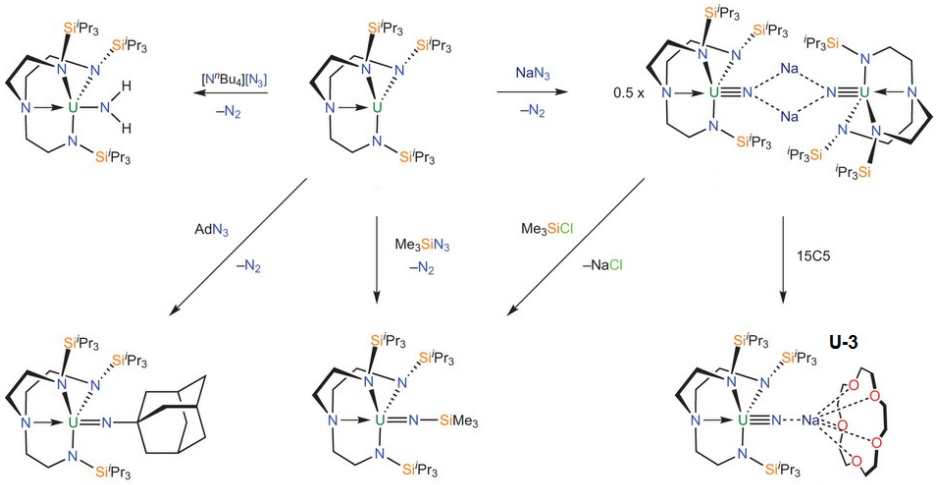

Figure 3: Reaction scheme for the synthesis of multiple uranium-nitrogen bonds (**U-3**) (King et al., 2013).

distances reported for uranium amides and uranium amines, which average at around 2.33 Å and 2.66 Å (Lu, 2014).

A thorough understanding of the nature of uranium ligand bonds could result in new developments for nuclear-waste clean-up, as well as new applications in the field of catalysis or in the synthesis of polymeric uranium nitride $[UN]_n$ for use as a ceramic nuclear fuel (Streit & Ingold, 2005).

5 Lanthanide Crystal Engineering in Malta

Solubility is one of the biggest challenges industrial chemistry has to face day by day. Prominent examples are paints, coatings, pigments, and last but not least, pharmaceutically active compounds. Almost all of these compounds are obtained as a solid at some stage during the manufacturing process. Solids are often preferred as the final product because of the obvious advantages in transporting and storing. However, almost all of the above-mentioned products are applied in the form of solutions or suspensions: e.g. drugs are swallowed as a solid and then will have to be dissolved in the stomach to enter the blood stream; paints have to be applied as solutions or suspensions.

Thus, almost all of the solids produced will have to be dissolved or suspended again when used. The solubility of a solid depends mainly on the amount of attraction between the individual molecules, ions or atoms in the crystal lattice, which again depends on their 3D arrangement. Crystal chemistry of molecular networks, where molecules are linked to each other by non-covalent interactions (H-bonds, π-interactions) or coordinative bonds involving multidentate ligands, can be considered as one of the most promising and appealing branches in modern solid-state chemistry. The formation and recombination of these molecular building blocks permits a unique fine-tuning of the physical-chemical properties of the synthesized compound in the solid-state without altering the actual molecule itself.

During the past decade a new field of science emerged from the above-mentioned necessity to control the arrangement of molecules and ions in crystalline solids: Crystal Engineering. It is "the understanding of the intermolecular interactions in the context of crystal packing and the utilization of such understanding in the design of new solids with desired physical and chemical properties" (Desiraju, 1989). This area of research does not fall neatly within the classical fields of chemistry (organic chemistry, inorganic chemistry, physical chemistry, materials chemistry, biochemistry). It cuts across these traditional vertical subdivisions. Analytical knowledge (e.g. crystallisation techniques, X-ray diffraction, biological processes, and thermo-analytical methods) and the practical experience to synthesise organic, complex, and organometallic compounds are equally important.

In spite of its great potential, very few research groups have applied a crystal engineering approach to molecular metal-based compounds in general (Braga, Grepioni & Maini, 2010; Evans & Lin, 2002; Kanaizuka et al., 2008; Mınguez Espallargas et al., 2010; Zaworotko, 2006). Their inclusion into cocrystal and polymorph screening for the development of the treatment is still to be exploited both from a theoretical and practical point of view.

This is true in particular in the field of 4f-element chemistry. Lanthanide elements are chemically very similar to alkaline and alkaline earth elements and thus can also influence the solubility of a compound signi-

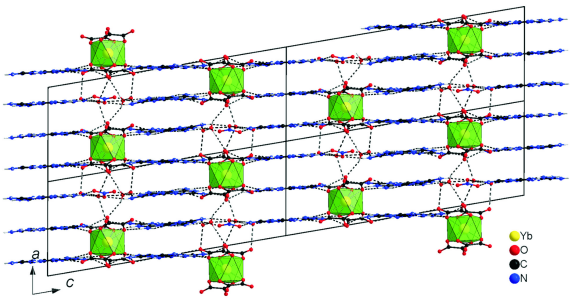

Figure 4: Crystal structure of **Yb-4** with view along cell axis b, dashed lines depict hydrogen bonding (Baisch & Braga, 2009).

ficantly. When surrounded by ligands, these elements have very low toxicity apart from being generally very stable towards oxidation.(Dyson & Sava, 2006; Mewis & Archibald, 2010) They could be utilised effectively not only in the formation of cocrystals or salts to alter the physical properties of drugs, but also to alter crystal growth/solubility of solids which are harmful to the body (e.g. urate crystals). There is currently no precedent literature for this approach.

Our group at the University of Malta functions as a collaborator in all aspects of Crystal Engineering for various 4f-element complexes (ionic and neutral) in the EUFEN COST action. Polymorph screening is carried out in order to explore the existence of new crystal forms with potentially different solubility properties.

The synthesis of lanthanide containing supramolecular compounds with exceptional materials properties has already been achieved as can be seen below (Figure 4) (Baisch & Braga, 2009). This compound has been synthesized by the reaction of melamine with ytterbium oxalate **Yb-4** in boiling water. Single-crystal X-ray analysis of suitable crystals revealed a structure in which melamine forms layers via an extended hydrogen bond network. The lanthanide complex molecules connect these layers vertically by multiple hydrogen bonds.

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Review Article

Symbiotics of History and Social Psychology: Understanding Social Representations of History in Europe

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Abstract. COST Action IS1205 aims at advancing knowledge and promoting networking among historians and social psychologists to analyse the role played by social representations of history in Europe. Social representations of history are central to the identity of groups that may or may not form the majority in any given country. In Europe, these representations are at best diverse, at worst fragmented, among various national and ethnic groups, either in the same country or across the continent. If left unexplored and unexplained, these social (mis)representations can incite adverse emotions, in turn influencing group behaviours and possibly leading to intergroup rivalry. Bridging the two disciplines through representatives from 28 countries, Action IS1205 addresses this issue by coordinating research on the role of: social cognitive processes in shaping lay representations of history; lay representation of history through the concepts of nationhood and identities; social-psychological studies of the narrative transmission of history through textbooks and the media; lay representation of history and group-based emotions in shaping attitudes, intergroup conflict and reconciliation processes.

1 Introduction

1.1 What is the relationship between History and Social Psychology?

The field of psychology is largely defined as 'the science of human behaviour'. Social psychology is that branch which deals specifically with human interaction. It seeks to establish general laws which describe and explain social relations. If such general principles of human behaviour could be established, it becomes more possible to determine social contexts in such a way as to offer optimal benefits to members of society, predict social behaviour, and reduce conflict Gergen, 1973.

History as a discipline is concerned with the course of international relations, nations and social institutions (political, financial, educational structures), economic and social development, social groups and movements (civil right protestors, student activists, strikers, trade unions), and of groups of people (women, minorities, children, migrant labourers). Historians focus on the history of particular systems, analyse the history of periods and events, and study the processes and actions of men/women cutting across various system levels Runyan, 1988. Therefore, history is fundamentally a study about people, unpredictable beings who make this discipline an investigation of angles and curves, rather than linear developments. Time and place are two crucial factors that any historian has to consider when trying to make sense of the experiences of the past Buttigieg, 2011.

Even from these very broad definitions, the relationship between the two disciplines could be easily drawn. The disciplines of social psychology and history share a fundamental concern with the human condition, be it in the form of 'individual and collective beliefs, mentalities, human behaviour and motivation, memory, personalities, emotions and feelings.' Tileagă and Byford, 2014. In spite of the long history of mutual suspicion and interdisciplinary uneasiness, recent works and projects are seeking to underline that the similarities between these two sciences outnumber their differences, and that subsequently, there are more benefits to derive from dialogue than from competition. This is the core remit of COST Action IS1205.

As economic historian Koji Yamamoto and social psy-

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chologist Vlad Glaveanu note:

> Social psychologists can benefit from engaging with historical sources by being able to contextualise their findings and enrich their theoretical models.... On the other hand, historians can enhance their analysis of historical sources by drawing upon the conceptual tools developed in social psychology [and] to 'test' these tools and contribute to their validation and enrichment from completely different perspectives. Glăveanu and Yamamoto, 2012

The attempt of bridging history and social psychology is not particular to this Action. This exercise of bringing the two disciplines closer together is embedded in a rich academic body of research and analysis from both ends of the spectrum. Scholars in social psychology, such as Münsterberg (1899), Gergen (1973), Runyan (1988), and historians such as Bloch (1924), Scott (2012), Millstone (2012) have long underlined the need of mutual exploration of the two disciplines, primarily in the analysis of collective memory.

These previous attempts notwithstanding, the distance between the two sciences remained, in that the effort was largely made by one side only, and not simultaneously. The process therefore has been fraught with theoretical and epistemological tensions, misunderstandings and mutual suspicion. Indeed, this is the gap that Action IS1205 tries to bridge. By providing a solid framework and forum for experts from both fields, this COST Action is ensuring two-way coordination of the bridging process. Research is still underway. Yet, constructive and promising results already point towards a hopefully successful exercise.

1.2 What is COST and Action IS1205?

COST is an intergovernmental framework for European Cooperation in Science and Technology.[1] Its multidisciplinary element is clearly reflected in the nine key domains which it promotes, ranging from biomedicine and molecular biosciences to transport and urban development, from physics and nano-sciences to individuals, societies, cultures and health. Action IS1205[2] is specifically on social psychological dynamics of historical representations in the enlarged European Union and it aims at advancing knowledge on the role played by social representations of history in processes of ethnic and national constructions of European identities, and intergroup conflicts. The Action is stimulating scientific cooperation among social psychologists and historians from around 27 European countries and Israel, along with the Palestinian Authority, New Zealand, and Argentina.

The Action also plans to influence and shape both scientific and public domains through academic and public dissemination. Throughout the estimated four years allocated for this Action (2012-6), the participating historians and social psychologists seek to: complement and expand existing knowledge regarding the psychological processes involved in the development and maintenance of lay representations of history; understand how representations of the past are collectively elaborated and remembered; complement and expand existing social psychological theories of intergroup relations and prejudice by integrating a historical dimension; identify, through concerted data collection, representations of the past that inform contemporary political conflicts across Europe; provide insight into how group-based emotions influence processes of collective remembering, identities, and intergroup relations; provide guidelines for the teaching of history of intergroup conflicts through reviewed pedagogical methods in the teaching of history; inform political decision-makers about the influence of lay representations of history, and history education on identities and intergroup relations through a brief policy briefing; prepare a concerted research project that will be submitted to a series of research-funding sources, such as Horizon 2020 and Eurocores.

2 Working Groups

2.1 The Working Groups (WGs): Work in Progress

The four working groups (WGs) through which these objectives will be achieved share the ultimate goal of exploring the interplay between lay representations of history, social identities and intergroup conflict. Although working relatively independently to ensure free choice of the most appropriate methods according to the respective group objectives, the four teams are bound by a set of common tasks. All four groups set off by reviewing the relevant literature from both disciplines in a concerted effort to increase mutual awareness of what already exists in the respective fields. This proved crucial to the build-up of ideas and the planning of future projects within the respective remits, while also establishing opportunities of cooperation with the other research groups.

By pooling in the individual research agenda and expertise, the participants collaborated to consolidate common research projects(s) which run in line with the individual area of interests so that contribution would be more forthcoming. It is noteworthy that in all WGs, more than one research project has been identified, which has motivated the formation of subgroups. These have already developed or are still in the process of de-

[1] http://www.cost.eu/ [Last accessed: 28 Aug. 2014]

[2] http://costis1205.wix.com/home [Last accessed: 28 Aug. 2014]

veloping research methods and designs that would contribute to updating the broader research agenda of each of the WGs.

Indeed, it is an important requisite that all subgroups of each of the WGs meet at the same time and in the same location so that, while getting enough space to work on their own, the subgroups would be able to mutualise their research findings and build a comprehensive and compact picture of their results. In other instances, two WGs may even plan to hold their meetings contemporaneously or to hold joint sessions where necessary. This in turn facilitates coordination of studies among the four WGs. Such orchestrated research is then aimed at informing the public, academics, journalists and decision-makers about the social psychological correlations of lay representation of history.[3]

2.1.1 Working Group 1

The first WG focuses on the role of social cognitive processes in shaping lay representation of history. It is coled by social psychologists Olivier Klein, Karen Douglas and Susanne Bruckmüller and includes six subgroups. It investigates the psychological processes that explain how communities forge collective memories around historical events. This in turn is meant to assist historians in avoiding biases in historical accounts.

Subgroup 1 is concerned with 'examining how ordinary people interpret historical events in a general sense by investigating what in history is considered important and worthy of an explanation – and what is not.' A study has already been undertaken to gauge how 'ordinary' people assess a given set of important events in European history. The second part of the project focuses on people's construals of the beginning versus the ending of an historical event. The initial hypothesis is that people would attach more importance to the beginnings of the events rather than the endings.

Subgroup 2 examines the influence of labelling a piece of information as a 'conspiracy theory' on the way that it is received. In the upcoming studies, the group plans to directly manipulate rationality by instructing participants to behave in a rational versus an intuitive way. This is meant to further explore how 'people's self-concepts of rationality interact with the way information is presented', in turn offering a deeper insight into people's interpretation of political and historical events.

Subproject 3 is concerned with the epistemic and affiliative functions of collective memory or what has been termed 'mnemic neglect'. The main hypothesis is that positive behaviours of in-group members will be better remembered than negative behaviours, especially by highly identified participants. The tool used in this study is a computer-administered personality inventory, the Michigan Omnibus Personality Inventory (MOPI). Each participant answers a number of items and reads a set of statements about citizens from two different countries: the participant's country and another country. This survey would then explore the way people process and remember information about their own and other cultural groups.

As for subgroup 4, it is organised in two groups. The first one is concerned with the effects of exposure to counter-stereotypical narratives on attribution. The second deals with the effects of exposure to counter-stereotypical narratives on cognitive processing by exploring the relationship between the commitment to the group narrative and the level of interest in the facts depicted by the narrative.

Subproject 5 is focused on the influence of historical analogies on current political judgement and attitudes. An online study on the current Ukraine conflict (Crimea crisis) has been undertaken. The conclusions allowed for too many interpretations. This has given rise to several follow-up studies during the autumn of 2014.

As for Subproject 6, it has prepared a theoretical paper on historical culture.

2.1.2 Working Group 2

Social psychologist Denis Hilton, and historians Chantal Kesteloot and Alberto Sá head WG2 which deals with lay representation of history in Europe, and focuses on concepts of nationhood and identities. It is concerned with 'the content, structure, and properties of social representations of history, and how they relate with ethnic, national and European identities.' A study around this issue has already been undertaken by analysing the conceptions of world history from data collected from 30 counties in Europe, Asia, Australasia, North and South America Liu et al., 2012. This WG2 now aims at conducting a similar study on Europe only.[4]

Three subgroups have been formed to facilitate this exercise, each focusing more specifically on WWI, colonialism and social representations of European history. Jointly, these subgroups are interested in uncovering the commonalities and differences in representations of historical events across Europe; analysing how the identification with the nation and with the continent correlates with differences in conceptions of national and European history, respectively; exploring the 'moral lessons' that people draw from historical events; evaluating how conceptions of nationhood and lay representations of national and continental history relate, or otherwise, with existing intergroup attitudes; and exploring how these conceptions relate with attitudes towards immig-

[3] http://costis1205.wix.com/home#!working-groups/ciop [Last accessed: 28 Aug. 2014]

[4] http://costis1205.wix.com/home#!working-groups/ciop [Last accessed: 28 Aug. 2014]

rants and acculturation processes in the receiving countries.

The first subgroup honed in on WWI by launching a Europe-wide survey among the participants and any other interested third parties. The participants collected/are collecting survey data about the way this war is represented nowadays, and how it relates with current attitudes and ideological positioning. This survey will ultimately help members to draw European comparisons, and to assess how commemorations will affect those representations and attitudes. The data is collected from undergraduate psychology ($n \geq 100$) and history ($n \geq 100$) students in each participating country.

Three countries (Serbia, Belgium and Finland) have collected big enough samples to start some analysis work. Although incomplete, preliminary conclusions are indicative of certain results. It turns out that the respondents have very minimal knowledge of WWI, that they believe that the war was the result of animosities between the leaders of different nations rather than a conflict between peoples, and that a nation's people and soldiers were constrained – rather than willing – to live the war and to fight for their country.

Subgroup 2 runs on the same lines as the World History Survey, but is focused on Europe instead. It is in course of drafting a similar survey as that of the World History Survey for circulation among its participants.

The third subgroup is concerned about social representations of colonialism and their implications in contemporary intercultural relationships. Historically, colonialism had a profound impact on the way formerly colonised peoples, as well as formerly colonising ones, represent themselves, the others, and the world Volpato and Licata, 2010. However, the history of colonialism is hardly taught in most European countries. This subgroup is investigating how collective memories of the colonial period in different settings relate with current intergroup representations and attitudes. A text-book analysis in a selection of former colonised and colonising countries is currently underway.

2.1.3 Working Group 3

Co-led by Tibor Polya and Eva Fülöp, WG3 deals with social-psychological studies of the narrative transmission of history. The participants are chiefly working on narratives as presented in history textbooks to study how institutional presentation of the past is diffused and consumed by younger generations. The initial assumptions are two: history textbooks currently used in schools serve as materials of social representations; historical texts, because of their inevitable narrative style, are conducive to evoke empathy. Based on these two points, history textbooks become sources of identification, which may project nationalism and therefore create distortions of social identity and representation. To this effect, the group has selected particular events, namely WWI and WWII and the colonial past, and a number of selective media about them, namely novels, movies and textbooks, to study their content and narratives.[5]

This WG is also using computer-assisted techniques of text analysis. This is the linguistic development environment NOOJ that includes large-coverage dictionaries, grammars and parses corpora in real time. Dictionaries and grammars are applied to texts in order to locate morphological, lexical and syntactic patterns and tag simple and compound words. NOOJ dictionaries and grammars can be built by users and they can process a dozen languages, including some Roman, Germanic, Slavic, Semitic and Asian languages, as well as Hungarian. It will be used to analyse various psychological dimensions in history textbooks.[6]

2.1.4 Working Group 4

The fourth and final WG, headed by Michał Bilewicz & Sabina Čehajić-Clancy, covers the roles of lay representations of history and group-based emotions in intergroup conflict and reconciliation processes. This group is working on the interplay between such specific history-related emotions as collective guilt, shame and pride, and intergroup mediation or confrontation.

Research on collective guilt has already yielded important results. What WG4 is in the course of doing is to extend this research further to cover the following areas: the conditions under which intergroup apologies and reparations satisfy their target groups; why some people defend against national guilt and others accept it; how victimisation influences intergroup attitudes; and how to temper the role of historical moral schemes in present day political attitudes.[7]

WG4 has established several project lines: to explore the extent to which and how intergroup relations in ethnically-mixed countries are determined by the different historical beliefs about ownership or authoctony; to look into the adherence to the 'official' historical narrative; to evaluate the dimensions of national identification; to assess the impact of official apologies on victim or perpetrator group members; or to investigate the role played by historical moral exemplars in historical narratives. Besides this, WG4 has also performed several studies in countries where historical genocides took place (Bosnia & Herzegovina, Poland), looking into how perpetrator, victim and bystander groups construe the past, with a view to analyse which strategies can be employed to foster reconciliation between them. The main

[5] http://costis1205.wix.com/home#!working-groups/ciop [Last accessed: 28 Aug. 2014]

[6] www.noojnlp.net/ [Last accessed: 28 Aug. 2014]

[7] http://costis1205.wix.com/home#{}!working-groups/ciop [Last accessed: 28 Aug. 2014]

subgroups deal with the following specified areas: victims and perpetrators - contemporary social perceptions of Fascism in different European countries; responsibility displacement; moral exemplars in history; historical representations of the Holocaust; parrhesiastic reconciliation.

3 Conclusion

The interaction of history and social psychology in this Action is seeking to find ways in which the past can be better understood in the present, while acknowledging that the present is forever informing perceptions of the past. The work in progress is successfully exploring the healthy symbiosis between the two fields and the participants are already reaping the benefits of this intensive dialogue. Some limitations have emerged, such as the numerical imbalance of social psychologists over historians, and the theoretical versus the interpretational priorities of social psychologists and historians, respectively. However, the increasing mutual awareness of the common grounds between the two disciplines means that the participants are constantly seeking to work on the strong points between them in search of the broader objective of Action IS1205.

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Research Article

Numerical Modelling of Soil Erosion Susceptibility in the Maltese Islands using Geographic Information Systems and the Revised Universal Soil Loss Equation (RUSLE)

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Abstract. The Mediterranean region is subject to various factors that exacerbate soil erosion pressures. Such factors include agricultural land fragmentation and abandonment, unsustainable agricultural practices and rapid urbanisation. Soil erosion in the Maltese Islands has been identified as a predominating land degradation process and a major threat to the sustainability of the agricultural sector. The small scale of the Maltese Islands facilitates an in detail national study of soil erosion processes and contributing socio-economic dynamics. The research methods, erosion rate values and controlling dynamics discussed in this work have a particular relevance to the Mediterranean area.

1 Introduction

Soil erosion is triggered by a combination of natural and anthropogenic factors that include steep slope gradients, intense precipitation, low vegetation cover and inappropriate land use (Renschler, Mannaerts & Diekkrüger, 1999; Wischmeier & Smith, 1978). Prolonged erosion leads to an irreversible loss of ecological and agricultural soil function and associated ecosystems services. Erosion reduces agricultural productivity, posing limitations to sustainable agricultural use. The aspect of soil erosion that causes most concern is the loss of topsoil, the most fertile part of the soil profile (Gobin et al., 2004).

The Global Assessment of Human-induced Soil Degradation (GLASOD) map estimates that 114 million hectares are affected by human-induced soil erosion (Oldeman, Hakkeling & Sombroek, 1991). The principal drivers of soil erosion in the European Union are unsustainable agricultural practices, overgrazing, deforestation and construction activities (Oldeman et al., 1991). The Mediterranean region is particularly susceptible to erosion (EEA, 1999). High erosion rates, in conjunction to slow soil formation, lead to irreversible reductions of Mediterranean soil quality and quantity.

The Maltese Islands (Figure 1) are located in the centre of the Mediterranean Sea. The Islands have a total land area of 316 km^2 and comprise three main islands, Malta, Gozo and Comino, and a number of outlying islets.

Figure 1: Map of the Maltese Islands (from (Ezilon, 2009))

The Maltese Islands have a semi-arid Mediterranean climate, with mild, wet winters and hot, dry summers. The average annual rainfall is around 524 mm and the average yearly temperature is 22.5 °C. Rainfall is characterised by storms of high intensity and relatively short

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duration (Government of Malta, 2002).

The Maltese Islands, and indeed the rest of the Mediterranean, are subject to various local factors which exacerbate soil erosion pressures. Such factors include agricultural land fragmentation and abandonment, limited soil agriculture suitability, unsustainable agricultural practice, rapid urbanisation, limited water resources and rapidly modernising social structure. As a consequence of the above interacting factors, soil erosion has been identified as a predominating land degradation process and a major threat to the sustainability of the agricultural sector (Tanti, Role, Borg & Calleja, 2002).

Maltese soil erosion risk modelling predating this article consisted of a numerical model developed by Tanti et al. (2002) assessing the northwestern region of Malta. The model identified areas threatened by soil erosion on the basis of geological substrate, slope, retaining rubble walls state and land cover. Model results clearly indicated that the assessed area was subject to high soil erosion rates (Tanti et al., 2002).

Presently, a large variety of empirical, semi-empirical, and physical process-based soil erosion risk models are available (Gitas, Douros, Minakou & Silleos, 2009; Erkal & Yildirim, 2012). The most widely applied empirical model (Fistikoglu & Harmancioglu, 2002) for assessing soil erosion by water driven mechanisms is the Universal Soil Loss Equation (USLE), developed by Wischmeier and Smith (1978). The USLE and its revision RUSLE (Renard, Foster, Weesies, McCool & Yoder, 1997) apply more than 40 years of experimental field observations gathered by the Agricultural Research Service of the USDA (Novotny & Olem, 1994). The RUSLE is applied is this study.

The dynamic relationship between human activities and resulting soil erosion requires that erosion be monitored. Regular monitoring allows competent authorities to appreciate the influence policy and land use change mechanisms have on soil erosion. Our study aims to provide quantitative estimates of soil erosion by water of the Maltese Islands for the year 2013. The discussion section examines the interaction between the socio-economic situation and consequent effects on soil erosion. This approach ties environmental science to policy issues and provides an integrated approach through which professionals and government may prioritise and present context specific erosion control measures. In this framework, high erosion risk areas are singled out, the physical, socio-economic and policy mechanisms influencing the area identified, and erosion control measures, via policy and physical intervention, suggested and implemented to reduce risk.

2 Methods

The RUSLE technique was applied and built into a GIS-based model. Relevant model input parameters were prepared separately and stored as GIS vector layers. Five vector layers, each representative of RUSLE factors, were converted to raster layers with a grid resolution of 50 metres. Each cell has a value representative of the area's factor value. Each raster layer was then combined in the GIS model to calculate soil loss for each cell in the study area for the year 2013. The predicted soil losses were verified against field observations of soil erosion made at the end of the 2013 winter season. The section below discusses the method followed to obtain the five RUSLE factor values.

2.1 RUSLE factors

The factors assessed in the RUSLE are rainfall erosivity (R), soil erodibility (K), slope length and steepness (LS), cover and management practices (C) and conservation practices (P) (Wischmeier & Smith, 1978). These factors are combined in a numerical formula (equation 1). The computation returns soil loss per unit area, equivalent to predicted erosion in $\mathrm{ton\,hectare^{-1}\,year^{-1}}$ (Gitas et al., 2009).

$$A = R \times K \times LS \times C \times P \tag{1}$$

where A = average annual soil loss ($\mathrm{t\,ha^{-1}\,yr^{-1}}$), R = rainfall/runoff erosivity ($\mathrm{MJ\,mm\,ha^{-1}\,h^{-1}yr^{-1}}$), K = soil erodibility ($\mathrm{t\,h\,MJ^{-1}mm^{-1}}$), LS = slope length and steepness (dimensionless), C = cover management (dimensionless), P = support practice (dimensionless)

2.1.1 Rainfall Erosivity (R factor)

Rainfall erosivity is a climatic factor that takes into account the erosive capacity of rainfall (D'Odorico, Yoo & Over, 2001; Le Bissonnais, Montier, Jamagne, Daroussin & King, 2002). The factor is determined as a function of total storm kinetic energy (E) and its maximum 30-min intensity (Imax30) (Wischmeier, 1959; Wischmeier & Smith, 1958).

The authors refer to the method employed by (Iraldo et al., 2013) in the calculation of the Maltese R factor. Iraldo et al. (2013) applied a modification of the Fournier index F, developed by (Arnoldus, 1980), the modified F index (F_F) (Ferro, Porto & Yu, 1999) (equation 2). The method uses average monthly ($\mathrm{p_{ij}}$) and annual precipitation (P). This approach is thought to be better correlated with rainfall erosivity.

$$F_F = \frac{1}{N}\sum_{j=1}^{N}\sum_{i=1}^{12}\left(\frac{\mathrm{p_{ij}^2}}{\mathrm{P_j}}\right) \tag{2}$$

where $\mathrm{p_{ij}}$ is the rainfall in month (mm) of the year j and P is the total rainfall per year.

Iraldo et al. (2013) calculate the F_F index using rainfall data for Malta (http://www.maltaweather.com) over the period 1985–2012. The R value was estimated using the average of R: F_F relationships adapted for Sicily (Ferro et al., 1999; Renard & Freimund, 1994). $R = 0.612\ F^{1.56}$ (Sicily). The R factor value defined by Iraldo et al. (2013) for Malta was 832.16 MJ mm ha^{-1} h^{-1} yr^{-1}.

2.1.2 Soil erodibility (K factor)

Soil erodibility expresses the intrinsic capacity of the soil to be eroded and reflects the effect of the average long-term soil profile response to rainfall and runoff erosion. The main soil properties affecting K are soil texture, organic matter, structure, and soil permeability (Erkal & Yildirim, 2012). High organic content decreases soil erodibility (F.A.O, 1996).

An extensive national soil survey, assessing over three hundred data points in a grid distribution of 1 km spacing, was carried out in 2002 (MALta Soil Information System, 2003). Amongst other soil parameters, the survey defined organic matter content and texture. Following Table 1, the MALSIS (2002) data set was used to define two hundred and sixty eight K factor values (Figure 2) for the Maltese Islands. Soil erodibility factor (K) value varies from 0.12 to 0.6 t ha h MJ^{-1} mm^{-1} and the mean value is 0.29 t ha h MJ^{-1} mm^{-1}.

The data points were then interpolated following GIS kriging method. The resulting GIS layer was converted to a raster map with a grid resolution of 50 metres. Each cell has a value representative of the area's K factor value (Figure 2).

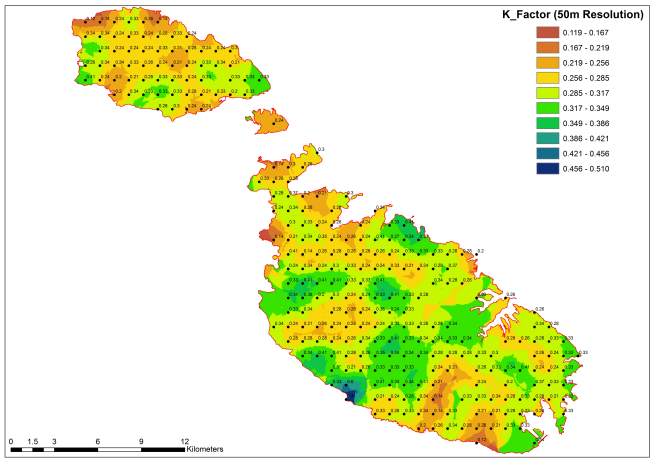


Figure 2: K factor raster map showing K factor values.

2.1.3 Slope length and steepness (LS factor)

Slope length and steepness reflect the proportional effect topography has on erosion (Foster & Wischmeier, 1974; Wischmeier & Smith, 1978). For this study, the LS factor was computed from a Digital Elevation Model (DEM) with the ArcGIS Spatial Analyst extension. The LS factor was calculated at a 10 m horizontal spacing from a 1:1500 scale topographic DEM following equation 3.

Table 1: K values as they were calibrated according to specific soil parameters

Textural classes	Organic matter less than 2 %	Organic matter more than 2 %
Clay	0.24	0.21
Clay Loam	0.33	0.28
Coarse Sand Loam	/	0.07
Fine Sand	0.09	0.06
Fine Sandy Loam	0.22	0.17
Heavy Clay	0.19	0.15
Loam	0.34	0.26
Loamy Fine Sand	0.15	0.09
Loamy Sand	0.05	0.04
Loamy Very Fine Sand	0.44	0.25
Sand	0.03	0.01
Sandy Clay Loam	/	0.20
Sandy Loam	0.14	0.12
Silty Loam	0.41	0.37
Silty Clay	0.27	0.26
Silty Clay Loam	0.35	0.30
Very Fine Sand	0.46	0.37
Very Fine Sandy Loam	0.41	0.30
Loamy sand	0.05	0.04
Silt	0.43	0.60
Sandy clay	0.10	0.14
Clay and heavy clay	0.24	0.21

$$LS = \left(\frac{\text{Slope} - \text{length}}{22.1}\right)^{0.5} 0.065 + 0.0456(\text{slope}) + 0.00654(\text{slope})^2 \qquad (3)$$

where Slope−length is in meters and slope is in %.

The computed LS factor GIS layer was converted to a raster map with a grid resolution of 50 metres. Each cell has a value representative of the area's LS factor value (Figure 3).

2.1.4 Crop and Vegetation management (C factor)

The management factor reflects the effect cropping and management practices have on erosion rates. The C factor is closely linked to land-use types and is a factor in soil erosion vulnerability (Wischmeier & Smith, 1978; Beskow et al., 2009). For this study, the C factor of the study area was obtained from a high resolution aerial orthophoto set produced in June 2012. The orthophoto

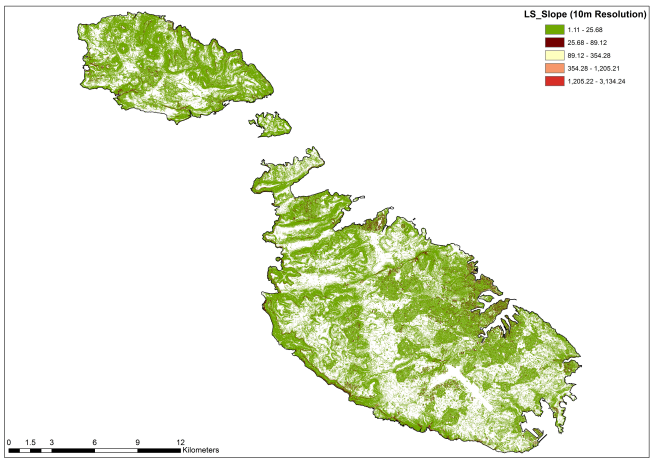


Figure 3: *LS* factor raster map.

data set was manually interpreted by the author, a land cover expert (Figure 4). Ground truthing surveys and land cover reports were consulted as a means of ensuring correct orthophoto land cover interpretation. A minimum mapping unit of 10.000 m^2 was applied following a classification system compliant to the CLC 2006 technical guidelines. A number of additional layers were defined in view of the large scale of orthophoto interpretation (1:5000). Maltese Island cover was divided into twenty two land-use types (Figure 4).

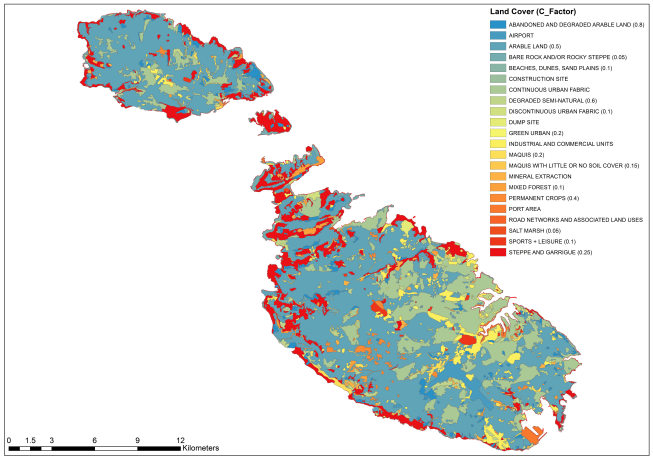


Figure 4: Detailed map showing spatial distribution of land use and cover in the Maltese Islands.

C factor values, corresponding to land cover classes (Table 2), were defined following expert defined descriptions (e.g., Wischmeier and Smith (1978), Morgan (1995), Pasák, Janeček and Šabata (1983), Alena (1991), Mališek (1992). The classification scheme excludes urbanised areas, bare rocks and water surfaces from evaluation since these surfaces contain no soil. Discontinuous urban, green urban (semi-permeable surfaces), mixed forest (moderate soil cover), maquis (with little to no soil cover), beaches, dunes, sand plains, rocky steppe and salt marshes as cover types of good management practice with a *C* factor value of less than 0.1. Maquis (with moderate soil cover), green urban (semi-permeable surfaces), sports and leisure (semi-permeable surfaces), pastures and permanent crops are categorised as land cover of moderate management practice with a designated *C* factor value between 0.11 and 0.4. Arable land, abandoned and degraded agricultural areas, and degraded semi-natural areas are categorised as land cover or low management practice of *C* factor values between 0.41 and 0.8. Land-use classes were allocated *C* values without considering seasonal variance.

Agricultural practice is of particular relevance when defining agricultural *C* factor. A large portion of Maltese arable soils are exposed, have no vegetation cover, and are deep-ploughed in anticipation of the first torrential September rains (RDP, 2007–2013). This agricultural practice intensifies water erosion and, as a consequence, a high *C* factor value was assigned to areas covered by arable land. Abandoned/ degraded agricultural areas and degraded semi-natural areas have the highest *K* factor values. Vegetation cover in these areas is often entirely removed to accommodate bird trapping, parking and other such activities that contribute towards accelerated soil erosion.

The land cover dependent *C* factor values were mapped in GIS. The resulting layer was converted to a raster map with a grid resolution of 50 metres. Each cell has a value representative of the area's C factor value (Figure 5).

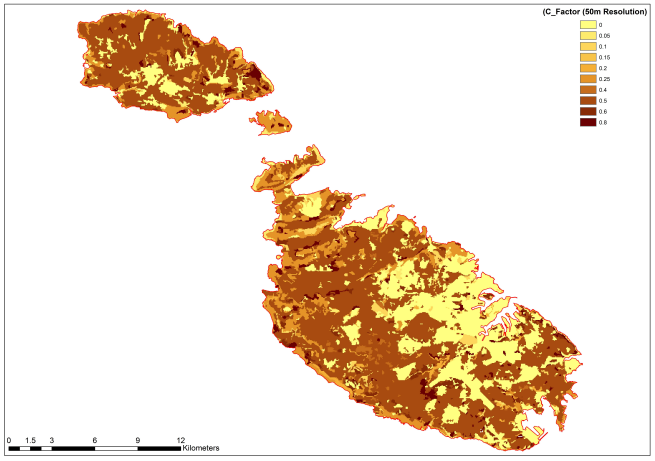


Figure 5: *C* factor raster map.

2.1.5 Erosion control (*P* factor)

Erosion control represents the effects various practices have on preventing soil erosion by water run-

Table 2: Land-use classes, cover in km^2 and allocated C value (land-use listed below only includes those with a C value)

Land cover type	Cover km^2 (% of total land)	C factor value
Discontinuous urban	3.36 (1.06%)	0.10
Green urban (semi-permeable surfaces)	0.26 (0.08%)	0.15
Sports + Leisure (semi-permeable surfaces)	1.95 (0.62%)	0.20
Arable land	164.50 (52.1%)	0.50
Permanent crops	2.91 (0.92%)	0.40
Abandoned + Degraded Agricultural areas	9.56 (3.03%)	0.80
Mixed forest (moderate soil cover)	4.20 (1.33%)	0.10
Steppe + Garrigue	36.07 (11.42%)	0.25
Maquis (with moderate soil cover)	4.69 (1.49%)	0.20
Maquis (with little to no soil cover)	0.08 (0.03%)	0.15
Beaches, dunes, sand plains	0.12 (0.04%)	0.10
Bare rock / rocky steppe (little to no soil cover)	5.83 (1.83%)	0.05
Degraded semi-natural areas	7.28 (2.31%)	0.60
Salt marshes	0.15 (0.05%)	0.05

off. Wischmeier and Smith (1978) discuss that control structures, which include *inter alia* improved tillage practices, strip cropping and terraces, should significantly contribute towards erosion control and frequently provide the major control in a farmer's field. The lower the P values, the more effective the conservation practice is deemed to be in reducing soil erosion (Erkal & Yildirim, 2012).

Appropriate farming practices may positively influence countryside and landscape quality, and sustain key environmental resources such as biodiversity, soil and water. Terraced agricultural fields are recognised as a characteristic feature of Mediterranean landscapes (Whitelaw & French, 1999; Frederick & Krahtopoulou, 2000; Grove & Rackham, 2001; Price & Nixon, 2005). Terraces adjust hillslopes into stepped, contour parallel, agricultural units of relatively flat ground suitable for cultivation. In the Mediterranean, terrace construction has typically involved the use of interlocked dry stone risers, rubble walls. These walls act as retainers to support back-lying beds of level soil. Although the original purpose of terrace construction is the increase of agricultural areas, these structures provide a necessary means of soil erosion control (Bevan & Conolly, 2011).

Maltese agricultural practices have significant control on agricultural land susceptibility to degradation and soil erosion. Tanti et al. (2002) identify retaining rubble walls in terraced fields as the most important water and soil erosion control method structures in the Maltese Islands (Tanti et al., 2002). Contour ploughing was also identified as a key erosion mitigation practice.

A national survey was conducted by the author over four months, starting in June 2013. The survey assessed rubble wall state following a classification scheme consisting of three potential rubble wall states. In this classification scheme, rubble walls in a good state contain a maximum of 1 breach showing half of the soil profile; rubble walls in moderate state contain more than 1 breach, but no more than 3, showing half of the soil profile, and rubble walls in a poor state contain more than 3 breaches showing half of the soil profile or 1 or more that show the whole soil profile. Each state is attributed P factor values; good state P factor value 0.3, moderate state P factor value 0.5, and poor state P factor value 0.7. The resulting GIS rubble wall state layer was converted to a raster map with a grid resolution of 50 metres. Each cell has a value representative of the area's P factor value (Figure 6).

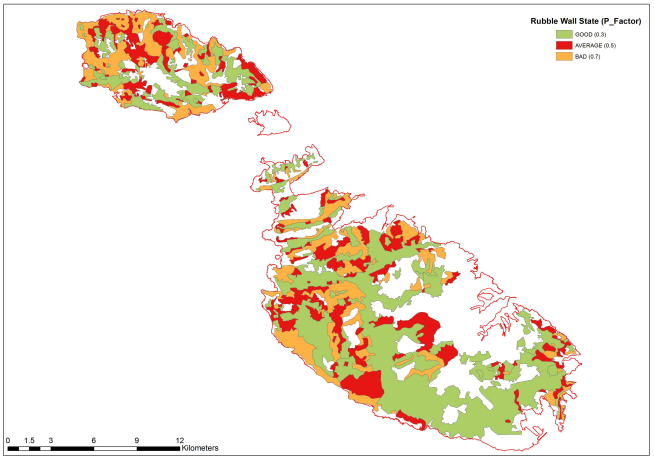


Figure 6: P factor raster map. Blank (white) areas represent urban areas or areas that contain no rubble walls.

3 Results

3.1 Computed soil loss

The average annual soil loss was computed on a cell-by-cell basis following equation 1. The five factor raster maps representing R, K, LS, C and P factors, were overlain and multiplied with the ArcGIS Spatial Analyst extension. The erosion map (Figure 7) shows the spatial distribution of soil loss in the Maltese Islands expressed as annual average soil loss in tonnes per hectare per year. The values should however be considered in a comparative manner rather than absolute values. This is due to the generalisation of the used input data as well as the nature of the model.

In order to obtain a better general understanding and be able to carry out a National comparison, the quantitative output of soil loss prediction was classified in eight categories of increasing soil loss severity: < 1 (none), 1 to 2, 2 to 5 (very low), 5 to 10 (low), 10 to 25 (moderate), 25 to 45 (high), 45 to 75 (very high), $>75\,\mathrm{t\,ha^{-1}\,yr^{-1}}$ (severe). Erosion severity thresholds are consistent with those presented by various experts (e.g., Iraldo et al. (2013)). Such a classification is consistent with the RUSLE model's role as a conservation management tool, where relative comparisons among areas are more significant than any assessment of the absolute soil loss in a particular location.

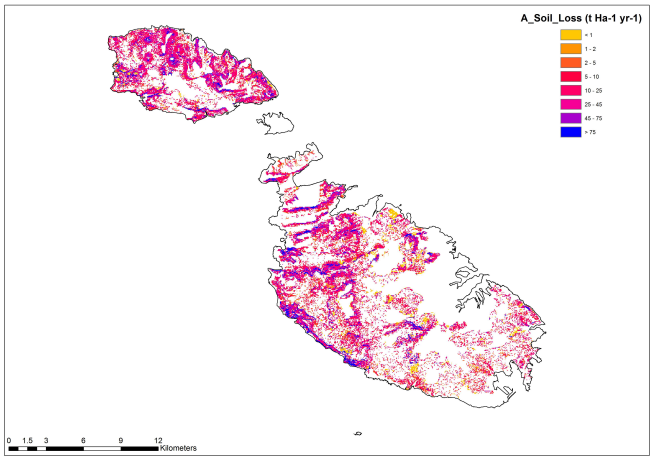
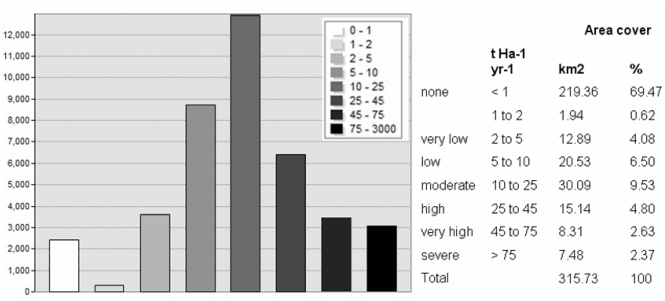


Figure 7: Average annual soil loss ($\mathrm{t\,ha^{-1}\,yr^{-1}}$) in the Maltese Islands following RUSLE equation.

3.2 Areas at risk

Calculated National annual soil loss (Figure 7 and 8) indicates that $61.01\,\mathrm{km^2}$, 19.33 % of total National land area, are at risk of moderate (10 to $25\,\mathrm{t\,ha^{-1}\,yr^{-1}}$) to severe ($>75\,\mathrm{t\,ha^{-1}\,yr^{-1}}$) soil erosion.



	t Ha-1 yr-1	Area cover km2	%
none	< 1	219.36	69.47
	1 to 2	1.94	0.62
very low	2 to 5	12.89	4.08
low	5 to 10	20.53	6.50
moderate	10 to 25	30.09	9.53
high	25 to 45	15.14	4.80
very high	45 to 75	8.31	2.63
severe	> 75	7.48	2.37
Total		315.73	100

Figure 8: Soil loss potential histogram. X-axis number of cells ($2500\,\mathrm{m^2}$) showing erosion values that fall within erosion rate categories, Y-axis soil erosion rate categories.

4 Discussion

Maltese central and north-eastern areas show the lowest erosion risk. These areas are characterised by relatively flat topographies, good land management and erosion control measures. Maltese north-western and Gozitan areas are characterised by a large range in erosion rates. Within this area, low erosion risk occurs in plateaus comprising low topographic gradients, and the application of good land management and erosion control measures. Plateau flanks typically consist of exceptionally high erosion rates, characterised by high topographic gradients, inappropriate cultivation practices and poor erosion control measures. Steeply inclined plateau flanks demonstrating low erosion risk are associated with areas demonstrating adequate vegetation cover, and effective management and conservation practices.

The spatial pattern of modelled potential soil erosion (Figure 7) is clearly proportional to slope gradient. Field work and national reports also identify land use (C factor) and control measures (P factor) as being critical. The highest estimates of quantitatively measured and predicted erosion rates occur in steeply inclined arable land where poor management and conservation practices are applied (e.g., Tanti et al. (2002).

A large portion of Maltese arable soils are exposed, all vegetation cover is removed and deep-ploughed in anticipation of the first torrential September rains. This agricultural practice intensifies water erosion. The author proposes that strip contour ploughing, where vegetation cover is retained between ploughed areas, is applied to reduce water induced soil erosion. Field evidence also clearly identifies agriculture retaining rubble walls as a key soil erosion control method in steeply inclined agriculture areas. The reasons leading to inappropriate agricultural practices, and consequent soil erosion, are closely tied to the National socio-economic situation often common to the Mediterranean region.

Agricultural land ownership is a key issue in the Maltese Islands. Two thirds of the agricultural land

is owned by the State and the remaining one-third by the private sector. Eighty percent of cultivated agricultural land is rented and twenty percent is occupied by owners or under a freehold basis (National Statistics Office, 2003). In accordance with the Agricultural Leases (Re-letting) Act drawn up in 1967, government and privately owned land is automatically re-let to the existing tenant or descendants. Law impedes the eviction of tenants or any substantial increases of rent, even on privately owned land. Given the low prices at which land is rented and prospects of strong land speculation, both tenants and private landowners tend to hold on to leased land. This situation has significant national consequences. Should leased agricultural land not provide a significant source of income, the arable land may be disused or used for other purposes. This process leads to accelerated land degradation and reduces the economies of scale potential, reduced production, of the Maltese agricultural sector (Rural Development Plan, 2007–2013).

Statistical results from the 2010 census indicate that seventy four percent (9.203 ha) of all agricultural holdings cover less than one hectare (census, 2010). Significantly contributing to this issue is land fragmentation, brought about by inheritance and parcel sale. This process significantly reduces the total exploitable land and thus diminishes economic viability of agriculture production. This may consequentially lead to land abandonment or change in land use.

Land abandonment may also be the consequence of increased international agricultural product cost competitiveness. Mediterranean agriculture is faced with severe limitations in this area. Naveh (1991) estimates that more than half of Mediterranean land is of marginal economic agricultural potential, characterised by steep, rocky uplands and poor soils. This setting often presents insurmountable economic obstacles for the introduction of modern agricultural techniques necessary in modern markets (Pinto Correia, 1993). For the Mediterranean regions, the trend towards land abandonment is accentuated by increasing competition with the highly productive agriculture of northwestern Europe (Pinto Correia, 1993). Agricultural activity survived in the Maltese Islands in the past fifty years as a result of protective measures, namely price guarantees and quota restrictions on imports, aimed at encouraging production by ensuring a regular income flow for local farmers. Maltese entry to the EU (2004) led to the dismantling of various protective levies and extensive sector restructuring to adhere to EU legislation (RDP, 2007–2013). These developments adversely influenced net farmer income. As a consequence, landowners may have to sustain net income through alternative employment, leading to reduced land management and land degradation. When agricultural exploits do not produce a source of revenue, farmers may also resign and abandon the area altogether.

The socio-economic conditions discussed above, agricultural land ownership, increased international agricultural product cost, agricultural holding size and land fragmentation, constrain net farm income. The generation of income via agricultural practices is central to understanding whether agricultural land is used and invested upon or abandoned and erosion processes potentially intensified.

The effects of land abandonment on soil quality and soil erosion may be either positive or negative. The key control on soil regenerative capacity is vegetation cover, controlled by climatic conditions and soil quality. Numerous authors have demonstrated that in a wide range of environments both runoff and sediment loss decrease exponentially as the percentage of vegetation cover increases (e.g., Elwell and Stocking (1976), Lee and Skogerboe (1985), Francis and Thornes (1990). Consequently, should the applied agricultural management practices have been unfavourable, the re-establishment of natural vegetation cover may reduce soil erosion. Central Mediterranean climatic conditions, dry summers reduce vegetation cover and winter flash floods exacerbate soil erosion, however do not favour natural vegetation reclamation. Another key parameter, significantly contributing to soil erosion in terraced fields, is the degradation of soil retaining rubble walls.

Soil retaining rubble walls in terraced fields are anthropogenic structures characteristic of sloped Mediterranean agricultural areas. Although the original purpose of terrace construction is the increase of agricultural areas, these structures provide a necessary means of soil erosion control (Bevan & Conolly, 2011). In Malta, significant expanses of garrigue were reclaimed for agricultural use; rubble material was used for levelling, topped-off with soil and retained by rubble walls (Rural Development Plan, 2004-2007). Under natural conditions, these soils would not accumulate in such areas, and given the opportunity, gravitational processes would transport soils to more stable areas. Unfavourable climatic conditions hinder natural vegetation reclamation and as a consequence, once rubble walls are breached and not restored, intensive soil erosion occurs.

The Maltese National survey, carried out by the author assessing land use (Figure 4) and rubble wall state (Figure 6), concludes that the majority of agricultural terraces on inclined surfaces are disused and retaining rubble walls in a derelict state. These steeply inclined agricultural fields show the highest National soil erosion rates (Figure 7). These areas are subject to various socio-economic conditions that constrain net farm income. Such hindering conditions include agricultural land ownership legislation, increased international agri-

cultural competition, agricultural holding size and land fragmentation. In the Maltese Islands, these fields, of limited size, low accessibility, and requiring high rubble wall maintenance, may have once been economically exploitable. However, with changes in socio-economic dynamics, the economic incentive for tending these marginal fields was lost and the fields abandoned. It is proposed that in most cases, these socio-economic factors, common to Mediterranean countries, significantly contributed towards agricultural land disuse and dilapidation of soil retaining structures. These conditions are the main drivers leading to accelerated soil loss. The socio-economic dynamics, and the consequent effects on agricultural practices and soil erosion processes discussed in this study are characteristic of the Mediterranean region.

There is an urgent need for an update of national legislation to alleviate the adverse effects socio-economic parameters have on agricultural practices. The author proposes that ameliorating net farmer income will directly increase agricultural land use, reversing the current abandonment trend. This mechanism will indirectly increase the maintenance of key soil erosion control structures, the terraced field rubble wall, and reduce the current alarming rate of soil erosion.

5 Conclusion

Calculated National annual soil loss (Figure 7) indicates that 61.01 km^2, 19.33 % of total National land area, are at risk of moderate to severe soil erosion. Maltese central and north-eastern areas show the lowest erosion risk. These areas are characterised by relatively flat topographies, good land management and erosion control measures. Maltese north-western and Gozitan areas are characterised by a large range in erosion rates. Within this area, low erosion risk occurs in plateaus comprising low topographic gradients, and the application of good land management and erosion control measures. Plateau flanks typically consist of exceptionally high erosion rates, characterised by high topographic gradients, inappropriate cultivation practices and poor erosion control measures. Steeply inclined plateau flanks demonstrating low erosion risk are associated with areas demonstrating adequate vegetation cover, and effective management and conservation practices.

Analysis of the Maltese National land cover (Figure 4) and rubble wall state (Figure 6) survey, carried out by the authors, concludes that the majority of agricultural terraces on inclined surfaces are in a derelict state and in most cases disused. These steeply inclined agricultural fields show the highest National soil erosion rates (Figure 7). These areas are subject to various socio-economic conditions that constrain net farm income. We propose that in most cases socio-economic factors, common to Mediterranean countries, significantly contribute towards agricultural land disuse, dilapidation of soil retaining structures and accelerated soil erosion.

Soil is a limited resource in the Mediterranean area both in terms of quantity and quality. Soil resources support agriculture, maintain ecosystem health and are central to hydrological processes. Although of great importance, soil resources are relatively mismanaged and are threatened by accelerated erosion rates. There is an urgent need for an update of national legislation to alleviate the adverse effects socio-economic parameters have on agricultural practices. Ameliorating Mediterranean net farmer income will directly increase agricultural land use, reversing the current abandonment trend. This mechanism will indirectly increase the maintenance of key soil erosion control structures, the terraced field rubble wall, and reduce the current alarming rate of soil erosion.

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Review Article

3D Video Coding and Transmission

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Abstract. The capture, transmission, and display of 3D content has gained a lot of attention in the last few years. 3D multimedia content is no longer confined to cinema theatres but is being transmitted using stereoscopic video over satellite, shared on Blu-Ray^{TM}disks, or sent over Internet technologies. Stereoscopic displays are needed at the receiving end and the viewer needs to wear special glasses to present the two versions of the video to the human vision system that then generates the 3D illusion. To be more effective and improve the immersive experience, more views are acquired from a larger number of cameras and presented on different displays, such as autostereoscopic and light field displays. These multiple views, combined with depth data, also allow enhanced user experiences and new forms of interaction with the 3D content from virtual viewpoints. This type of audiovisual information is represented by a huge amount of data that needs to be compressed and transmitted over bandwidth-limited channels. Part of the COST Action IC1105 "3D Content Creation, Coding and Transmission over Future Media Networks" (3D-ConTourNet) focuses on this research challenge.

Keywords: 3D video transmission, multi-view video coding, quality of services

1 Introduction

Multimedia communications has been improving over the years, starting from the broadcasting of black and white television to today's ultra high definition colour transmission and stereoscopic video. These improvements, together with the availability of more services and use of different devices to view the content, including mobile equipment, require more and more data to be transmitted, increasingly demanding more bandwidth from the telecommunication networks. Recent surveys (CISCO, 2014) expect that video traffic will reach around 79% of all the consumer generated Internet traffic in 2018.

To date most of the 3D multimedia experiences have been limited to cinema viewing and controlled environments. This is mainly attributed to the high investments needed to develop these environments and bandwidth demands. However, technologies across the whole chain from capture to transmission to displays have been advancing at a high rate and stereoscopic video has become available for home consumption with content transmitted over satellite, Blu-Ray^{TM}, and Internet technologies (Vetro, Tourapis, Müller & Chen, 2011). In general, viewing this type of video requires the use of special glasses to filter the content towards the correct eye of the viewer to obtain the 3D perception. However, the experience of the viewer can be further improved by transmitting more camera views of the same scene and use displays which do not need glasses. If depth data is added to the multi-view stream, virtual views can be generated using Depth-Image-Based Rendering (DIBR) at the display allowing the user to determine a personal viewing angle, known as Free-viewpoint Tele-Vision (FTV) (Ho & Oh, 2007). All the data generated has to generally pass through a limited bandwidth channel and therefore adequate coding must be performed.

Transmission of 3D and immersive multimedia services and applications over heterogeneous networking technologies includes broadcasting channels, wideband backbone links, bandwidth-constrained wireless networks, among others (Lykourgiotis et al., 2014). At transport level, three main system layers have been considered in the recent past, as the most adequate for 3D media delivery: MPEG-2 systems, Real-time Transport Protocol (RTP) and ISO base media file format (Schierl & Narasimhan, 2011). However, these legacy technologies are now facing new challenges as a result of fast

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evolution towards future media networks. For instance, 3D multimedia streaming requires flexible adaptation mechanisms capable of delivering subsets of 3D data according to network constraints or users' preferences and robust coding and transmission schemes are necessary to cope with error-prone channels and dynamic networking such as Peer-to-Peer (P2P) (Gurler & Tekalp, 2013). In this context, the challenge of achieving an acceptable level of Quality of Experience (QoE) has been evolving from a technological perspective (Cubero et al., 2012) by including an increasing number of human factors (Taewan, Sanghoon, Bovik & Jiwoo, 2014) and acceptance in different usage scenarios (Wei & Tjondronegoro, 2014).

The COST Action IC1105 "3D Content Creation, Coding and Transmission over Future Media Networks" (3D-ConTourNet) aims at bringing together researchers from all the spectrum of the 3D technology chain to discuss current trends and research problems. It also provides, through dissemination of findings, information for stakeholders on the state-of-the-art technology and services. This article deals with the 3D video coding and transmission part of this COST Action.

The paper is divided into five sections. The next section gives information related to the available 3D video formats. Section 3 deals with the coding of 3D videos while section 4 focuses on the transmission of the 3D content. At the end, a conclusion is given.

2 3D Video Formats

2.1 Stereoscopic Representations

The most cost effective way to transmit 3D videos is using stereoscopic representation. This only needs to transmit two views, one intended for the left human eye and the other one for the right eye. The transmission is done sequentially. These two views can be transmitted at a lower resolution in the same space dedicated for a high definition television frame and positioned either side-by-side or in top-and-bottom fashion. In (Zhang, Wang, Zhou, Wang & Gao, 2012), the authors propose the transmission of one single video plus the depth information. In this case the second view is generated at the display using DIBR techniques. In all cases, the video can either be viewed using a normal television by decoding one of the views or in 3D using any type of stereoscopic display.

2.2 Model-based Representation

This approach considers the video as a sequence of 2D projections of the scene. It uses closed meshes, such as triangle meshes (Theobalt, Ziegler, Magnor & Seidel, 2004), to represent generic models. Adaptation through scaling of the segments and deformation of surfaces is then applied to better represent the objects in the scene. The input streams are mapped into texture space transforming the 3D model into 2D. The texture maps of each camera view are encoded at every time stamp using 4D-SPIHT (Theobalt et al., 2004; Ziegler, Lensch, Magnor & Seidel, 2004) or similar methods. Semantic coding can also be used for model-based representations, where detailed 3D models are assumed to be available (Kaneko, Koike & Hatori, 1991). The drawback of semantic coding schemes is that it can only be used for video having known objects.

2.3 Point-sample Representation

2D video can be mapped to 3D video polygon representation using point sample methods. Such a technique is applied in Würmlin, Lamboray and Gross (2004), where a differential update technique uses the spatio-temporal coherence of the scene captured by multiple cameras. Operators are applied on the 3D video fragments to compensate for the changes in the input and are transmitted with the video stream. The 3D video fragment is defined using a surface normal vector and a colour value. This method also needs the transmission of camera parameters and identifiers together with the coordinates of the 2D pixels.

2.4 Multi-view Video Representation

This representation considers the capturing of a scene from multiple cameras placed at different angles. This generates a huge amount of data proportional to the number of cameras capturing the scene. To reduce this huge data and provide for better scalability Multi-view Video Coding (MVC) can be used (Vetro, Tourapis et al., 2011). Furthermore, the Multi-View (MV) representation is an extension of the High Efficiency Video Coding (HEVC) standard. An overview of HEVC can be found in Sullivan, Ohm, Han and Weigand (2012).

2.5 Multi-view Video Plus Depth Representation

The Multi-view Video plus Depth (MVD) format includes the transmission of depth maps with the texture video. The depth information adds geometrical information that helps in achieving better encoding and view reconstruction at the displays. This format supports the use of less views, as intermediate views can be constructed at the display, ideal for wide angle and autostereoscopic displays (Vetro, Yea & Smolic, 2008). This format will probably be the main format for transmission of 3D videos for HEVC coded content.

3 3D Video Coding

3.1 Stereoscopic 3D Video Coding

The current way of transmitting 3D video is using stereoscopic technology. This mainly involves the capture of the scene using two cameras similar to the human vision system. These sequences are then separately presen-

ted to the left and right eye of the viewer. In this case, the video is either coded by means of simulcasting, where each view is compressed using H.264/AVC or HEVC, or by placing the two images, one from each stream, in a single high definition frame. In the latter, known as frame compatible format, the resolution is decreased, but is an efficient way of coding since the bandwidth required is similar to the single-view transmission.

3.2 Multi-view Video Coding

This coding scheme allows for a more efficient way to transmit multiple views compared to simulcasting each individual view. This is done by exploiting the redundancies available between camera views. Thus, H.264/MVC and MV-HEVC use spatial, temporal and inter-view predictions for compression. An overview of the MVC extension to the H.264/AVC can be obtained from Vetro, Weigand and Sullivan (2011). The multi-view video can be coded using different structures; the most commonly used in literature being the low latency structure and the hierarchical bi-prediction structure. The low latency structure, shown in Figure 1 for 3 views, uses only previously encoded blocks for its predictions in the time axis. Bi-prediction is still applied in between views, but this is done at the same time instant and therefore the decoding does not need to wait for future frames and needs a smaller buffer. On the other hand, the hierarchical bi-prediction structure uses future frames in the encoding as shown in Figure 2. This implies that a larger buffer is needed and the decoding has to wait for the whole group of pictures to start decoding. The advantage of this structure is that it provides a better coding efficiency and therefore less data needs to be transmitted.

3.3 Video-plus-depth Coding

Even though current multi-view encoders can provide very high compression ratios, transmission of the multiple views still needs huge bandwidths. However, to satisfy the need of a high number of views to generate an immersive 3D experience, a lower number of views can be transmitted together with the depth data. The missing views can then be interpolated from the transmitted views and depth data. This can be done using a synthesis tool such as DIBR with the geometry data found in the depth maps. The texture and depth videos can be encoded using the 3D video coding extensions discussed above and then multiplexed on the same bit stream. Otherwise, they can be jointly encoded such that redundancies inherent in the texture and the depth videos can be exploited for further coding efficiencies. An example of such a coding method is found in Müller et al. (2013) and is now an extension of the HEVC standard. The HEVC extension for 3D (3D-HEVC) improves the coding efficiency by exploiting joint coding of texture images and the corresponding depth maps (Tech et al., 2015).

3.4 Research Trends in Video Coding

Although a lot of work has been done in 3D video coding, more research is still needed to provide for fast, more efficient and cheap encoders. This can be done by reducing further the redundancies in the videos, applying more parallel algorithms, simplifying processes, catering for scalability due to the different display resolutions, applying more prediction schemes, and other ideas. The 3D-ConTourNet COST Action members are discussing these issues and are working to address these in order to get better 3D video transmission closer to the market.

4 3D Video Transmission

Three-dimensional video delivery is mainly accomplished over broadcasting networks and the Internet, where the IP protocol prevails in flexible platforms providing IPTV and multi-view video streaming. In broadcasting and IPTV services, 3D video streams are encapsulated in MPEG-2 Transport Streams (TS) and in IPTV TS that are further packetised into the Real-Time Protocol (RTP) and User Datagram Protocol (UDP), which provide the necessary support for packet-based transmission and improved QoS. Since TS and RTP provide similar functionalities at systems level, this type of packetisation introduces some unnecessary overhead, which is particularly relevant in multi-view video due to the increased amount of coded data that is generated. In the case of Internet delivery, HTTP adaptive streaming is becoming more relevant, since it allows low complexity servers by shifting adaptation functions to the clients, while also providing flexible support for different types of scalability under user control, either in rate, resolution and/or view selection, besides improved resilience to cope with network bandwidth fluctuations.

Since the term "3D video" does not always correspond to a unique format of visual information, the actual transport protocols and networking topologies might be different to better match the compressed streams. For instance, enabling multi-view 3D video services may require more bandwidth than available if all views of all video programs are simultaneously sent through existing networks. However, as mentioned above, if DIBR is used, a significant amount of bandwidth may be saved, because the same performance and quality might be kept by simply reconstructing some non-transmitted views at the receivers, from their nearby left and right views. Such possibility is enabled by the MVD format, which allows reconstruction of many virtual views from just few of them actually transmitted through the communications network.

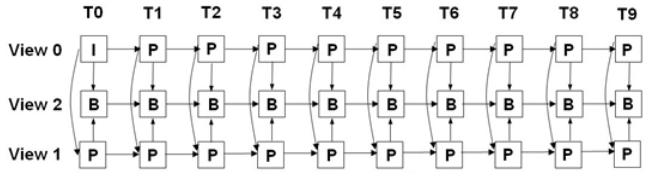

Figure 1: The low latency MVC structure. I represents an Intra coded frame, P represents a predicted frame, and B represents a bi-predicted frame.

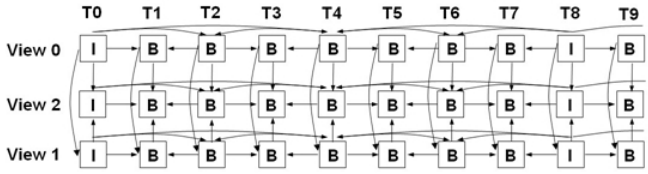

Figure 2: The hierarchical bi-prediction MVC structure.

Interactive streaming also poses specific transmission requirements in 3D multi-view video. In non-interactive services, multiple views can be sent through a single multicast session shared simultaneously by all clients, while interactivity requires each view to be encoded and transmitted separately. This allows users to freely switch between views by subscribing to different multicast channels. Multipath networks, such as P2P, can also provide the necessary support for interactive multi-view 3D video streaming by assigning the root of different dissemination trees to different views, which in turn can even be hosted in different servers (Chakareski, 2013). In the case of mobile environments, there are quite diverse networking technologies that might be used to provide immersive experiences to users through multi-view video, but the huge amount of visual data to be processed and the limited battery-life of portable devices is pushing towards cloud-assisted streaming scenarios to enable deployment of large-scale systems where computational power might be provided at the expense of bandwidth (Guan & Melodia, 2014).

Figure 3 summarises the main protocol layers used in 3D video broadcasting and streaming services. In the left side, the traditional DVB, including satellite, terrestrial and cable is shown. Basically, the Packetised Elementary Streams (PES) are encapsulated in TS before transmission over the DVB network. An extension of the classic 2D MPEG-2 Systems was defined to support multi-view video, where different views may be combined in different PES to provide multiple decoding options. The right side of Figure 3 shows a typical case of IP broadcasting and/or streaming of 3D multi-view video. Multi-Protocol Encapsulation (MPE) is used to increase error robustness in wireless transmission (e.g. DVB-SH), while Datagram Congestion Control Protocol (DCCP) may be used over Internet. In this case, MPEG-2 TS encapsulation may not be necessary. In the case of multi-view streaming using RTP, either single-session or multisession may be used to enable a single or multiple RTP flows for transport of each view. The underlying communication infrastructure can be quite diverse (e.g. cable, DVB, LTE). Like in classic 2D video transmission, dynamic network conditions fluctuation, such as available bandwidth, transmission errors, congestion, jitter, delay and link failures are the most significant factors affecting delivery of 3D video across networks and ultimately the QoE.

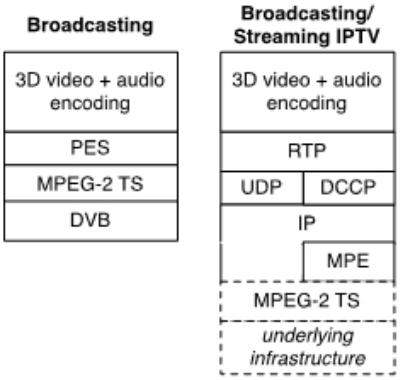

Figure 3: Generic protocol stack for 3D video services.

However, the increased amount of coded data and high sensitivity of 3D video to transmission errors requires robust coding techniques and efficient concealment methods at the decoding side because the perceived QoE in 3D video is known to be more sensitive to a wider variety of quality factors than in classic 2D (Hewage, Worrall, Dogan, Villette & Kondoz, 2009). Two robust coding techniques suitable for such purposes are scalable 3D video coding and Multiple Description Coding (MDC). In both of them several streams are produced and transmission losses may only affect a sub-

set of them. In the case of scalable 3D video coding, there is one main independent stream (base layer) that should be better protected against transmission errors and losses while the other dependent streams, or layers, can be discarded at the cost of some graceful degradation in quality. In MDC, each stream is independently decodable and can be sent over different paths to avoid simultaneous loss. This is particularly efficient in multipath transmission over P2P streaming networks (Ramzan, Park & Izquierdo, 2012).

4.1 Research Trends in 3D Multimedia Transmission

Current research trends in 3D and multi-view transmission span over several key interdisciplinary elements, which aim at the common goal of delivering an acceptable QoE to end-users. Heterogeneous networks comprising hybrid technologies with quite diverse characteristics and the increasing dynamic nature of 3D multimedia consumption (e.g. mobile, stereo, multi-view, interactive) pose challenging research problems with regard to robust coding, network support for stream adaptation, scalability and immersive interactive services, packet loss and error concealment. Hybrid networks and multipath transmission in P2P is driving research on MDC of 3D multimedia combined scalability and P2P protocols. While MDC is certainly better for coping with dynamic multipath networks, scalability might offer the most efficient solution for pure bandwidth constraints. Network-adaptation by processing multiple streams in active peer nodes is also under research to ensure flexibility and acceptable QoE in heterogeneous networks with different dynamic constraints and clients requiring different sub-sets of 3D multimedia content. The problem of accurate monitoring of QoE along the delivery path has been an important focus of the research community, but no general solution has yet been devised, so much more research is expected in the near future in this field. Synchronisation of the video streams across the different network paths is another open problem which can lead to frequent re-buffering and jittering artifacts. Overall, joint optimisation of coding and networking parameters is seen as the key to accomplish high levels of QoE, validated through widely accepted models.

5 Conclusion

An overview of the most important elements of 3D video coding and transmission was presented with emphasis on the technological elements that have open issues for further research and development. 3D video formats have evolved from simple stereo video to multi-view-plus-depth, which leads to a huge amount of coded data and multiple dependent streams. The need for robust transmission over future media networks using multiple links, providing in-network adaptation functions and coping with different client requirements was also highlighted as necessary for achieving and acceptable QoE. As an active multidisciplinary field of research, several promising directions to carry out further relevant investigations were also pointed out.

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Note Article

CM1106 STEMCHEM: Chemical Approaches to Targeting Drug Resistance Cancer Stem Cells

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Abstract. STEMCHEM is a COST action aiming to target causes of drug resistance in cancer stem cells. Cancer stem cells are cells which are believed to be responsible for the larger part of the regenerative capacity of cancers. They are also thought to be similar to adult stem cells in that they do not proliferate most of the time and are thus resistant to many kinds of chemotherapy. The action brings together labs around Europe in both biological and chemical fields to work together in this regard. Biologists targeting individual stem-cell related molecules as well as stem cell phenotypes (like the undifferentiated state), test chemicals from numerous labs for activity in high throughput screens, with the aim of identifying new drug targets. This COST action, like most others, offers opportunities for Malta, both in a general way and also particularly for a small country with small labs.

Keywords: Stem cells; Cancer; Differentiation; COST; drug resistance

COST (European Cooperation in Science and Technology) is one of the longest running European Scientific programmes, which allows for better coordination of nationally funded research at a European level.

Unlike the FP7/Horizon 2020 instruments which work in a top-down manner with calls being issued by the Commission as to what areas of research it is interested to fund, COST works in a bottom up or grass-roots approach. Any group of labs or interest groups from a few different countries can get together, start up a COST action, after it is vetted by the COST central administration in Brussels and then open it up to other partners to join in. This information is usually passed on the national contact point organisation (in Malta the MALTA COUNCIL FOR SCIENCE AND TECHNOLOGY), who informs interested potential participants on a regular basis of the newest set of COST actions to be set up.

For a small country like Malta, with small labs and minimal funding, where entry into the big consortia characteristic of the FP programmes has always been difficult (and I speak from experience here), COST allows the development of important scientific contacts, which may later lead to participation in such collaborations. It also allows increased scientific exposure of research done in our laboratories.

The University of Malta's Anatomy Department through a number of graduate students under Pierre Schembri-Wismayer's supervision has been working for some time on inducing differentiation of leukaemia cell lines using a variety of natural extracts, some of which have been published (Agius Anastasi, Cassar & Schembri-Wismayer, 2012) and others of which not yet due to the possibility of developing intellectual property for the University. The research uses the HL60 leukaemia cell line as an initial screen since it allows differentiation into both monocytic and granulocytic cell types.

When reviewing newer COST actions, Prof. Janet Mifsud, COST coordinator in Malta brought to our attention an action entitled "Chemical Approaches to Targeting Drug Resistance in Cancer Stem Cells" in December 2011. This seemed interesting so I, (Pierre Schembri-Wismayer) approached Prof. Mifsud about joining and was elected a member on the managerial board of the Action, representing Malta. I attended the

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kick-off meeting in Brussels, with the rest of the management committee since we got involved early on. The meetings of the management committee (which tend to be combined with workshops in this Action in order to allow more to get done at once) allow the regulation of the budget and of how the action is set to work.

Cancer stem cells (CSC) are a subpopulation of cells within tumours that exhibit enhanced tumor-initiating attributes and are a major contributing factor to current cancer therapy failure. The CSC phenotypic state comprises distinct molecular and functional differences that underpin resistance to current treatments and the unique ability spread and to seed new tumors throughout the body. This insight of this particular subpopulation of cells and its capability of repopulating tumours where most cells have been killed by conventional therapy, necessitates an entirely new approach to cancer drug development. This action aims to unite expertise in rational drug design and medicinal chemistry with biomedical investigators dedicated to understanding the mechanisms governing drug resistance in cancer stem cells. Thus it aims to develop effective methods for identifying novel compounds and drug candidates that target these drug-resistant cancer cells. One such way in fact would be to cause these cancer stem cells to differentiate into less stem-like cancer cells so that they can then be killed by more standard chemotherapy. The rationale and basis behind the action are reviewed in more detail in a recent publication from the consortium (Sotiropoulou, Christodoulou, Silvani, Herold-Mende & Passarella, 2014).

During the first meeting in Brussels we also chose a number of important positions such as the Chair (Prof. Daniele Passarella, a chemist from University of Milan, Italy who had initiated the action) and Vice Chair (Prof. Marija Balic, a medical doctor and biologist from Austria) as well as a number of important positions relating to specific instruments of the actions, (such as Dr Gabriela Almeida from Portugal who is in charge of short term scientific missions - STSMs - more about these later).

It was decided for example that members were divided into three working groups, one of chemists, one of biologists and one of pharmaceutical and medicinal chemists, including specialists in computational prediction of drug-target interactions. Our lab joined working group one, primarily for biologists. However, even within this group, there were numerous variations, which enriched the meetings since attending the various workshops not only allowed sharing of our own expertise but learning from others.

Amongst the biological experts were clinicians developing novel treatments for brain cancers, biologists developing *in vitro* models for various cancers, scientists developing *in vitro* systems of testing different well known stem-cell related candidate targets like Notch and Hedgehog, those screening for epigenetic modifiers of the stem cell phenotype and others like ourselves involved in phenotypic screening.

Chemists also hailed from different branches, including synthetic chemistry, producing steroids, retinoids and other potential drugs, pharmaceutical chemistry, *in silico* screening and development of chemical libraries.

These three groups worked within themselves, each setting up different workshops at the different group meetings. They also set up collaborative activities, such as the development of a chemical database of different kinds of agents for testing, from the chemists. However, it later became clear that the best work and publications would result from the development of cross-speciality collaborations where biologists tested new chemicals in their various test systems. In fact the three work-group system whilst still a functional grouping, became less significant as time went by. Numerous collaborations have been developed over the period of the COST action (although it is still ongoing) resulting in various publications (Madeira et al., 2014; Christodoulou et al., 2013; Majchrzak et al., 2013; Porcile et al., 2014).

Once the action was established, the first workshop was held in Milan in July 2012 and two members from our research group, Dr Pierre Schembri-Wismayer and Dr Krystle Blaire Theuma attended. The meeting was very well organised and showed many different approaches to tackling stem cells in Cancer, including both the targeting of specific stem cell-related molecules using chemical approaches and the wider search of natural or synthesised products for anti-stem cell activity using a particular biological model system.

One of the main points about such meetings is that apart from the consortium members themselves contributing, external speakers (usually experts in the field who can contribute a new view point or angle to ongoing research) are invited (and funded) to attend the different meetings.

We presented some research work, using natural extracts for leukaemia differentiation. An oral presentation presented by PSW was entitled "Insect cell-extract induces increased expression of differentiation markers in HL60 leukaemia cells" whilst Dr Theuma (also a managerial committee member in the STEMCHEM consortium) presented a poster entitled "Combination of DNA modifying agents and differentiation inducers can enhance differentiation in HL60 leukaemia cells", which was work she had done in collaboration with Ms Analisse Cassar (Fig. 1).

The advantage of this was that since we were working using phenotypic screening (looking for a biological effect which could lead to candidate compounds for drug development and since we (mostly A. Cassar) has de-

Figure 1: Dr Schembri-Wismayer and Dr Theuma presenting their poster and discussing with Prof. Navakauskiene.

veloped this phenotypic screen into a relatively high throughput system, many chemists in particular, were interested in talking to us to assess their compounds. The fact that the group's area of expertise is also the less commonly targeted mechanism of differentiation therapy rather than cytotoxicity also increased interest.

Basically, we spectrophotometrically assess a leukocyte differentiation-related enzymatic activity (NBTZ). This is then divided by a mitochondrial activity which acts as a surrogate for cell number giving us and indication of the average differentiation marker per cell (Fig. 2).

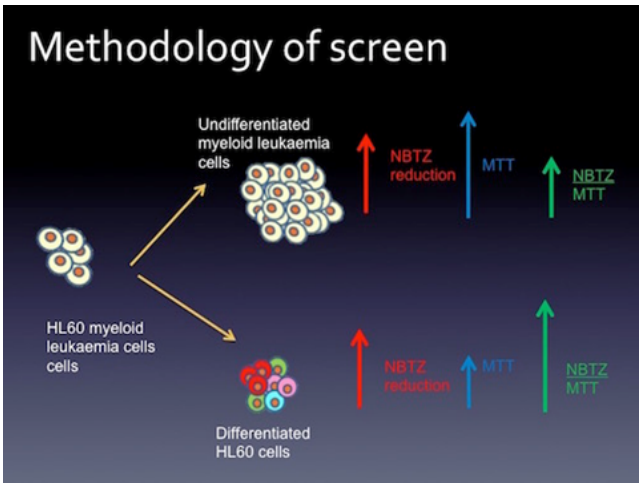

Figure 2: Schematic showing rationale behind screening tests.

At a second workshop meeting in Porto in February 2013, Analisse Cassar, another graduate student from the same lab presented her paper "Effects of Insect Conditioned Medium in Combination with Chromatin-Modifying Agents on the Terminal Differentiation of Leukaemia". She was given a lot of good feedback and I (PSW) received glowing reports of her very good presentation of very interesting work from other very authoritative STEMCHEM colleagues.

Analisse presented this work in the same session as a colleague form Lithuania, Ruta Navakauskiene from Vilnius University, who presented complimentary and similar work entitled "Effects of HDACI, HMTI and HMTI in combination with retinoic acid on granulocytic differentiation of human promyelocytic leukemia cells". Our lab and that of Prof. Navakauskiene have often found that we are working on similar areas so sharing our expertise and skills in STEMCHEM has been useful in turning us from primarily competitors to collaborators.

In fact, another post-graduate student in the lab, Mr Sherif Suleiman, supervised by Dr Jean Calleja Agius, the present head of the Anatomy department will hopefully be benefiting form a short term scientific mission to Prof. Navakauskiene's labs in Lithuania, where he will be learning how to test for different chromatin-modifying agents using molecular biology techniques not presently in common use in our own labs. We on the other hand have benefited from colleagues in Europe visiting our lab, from Greece (officially funded as an STSM) , Ireland and Serbia, through the STEMCHEM consortium.

This is in fact the aim of a short terms scientific mission. This is funded as part of the action and involved up to €300 for travel and €60–€90 as a daily allowance up to a maximum of €2500 for a period up to 3 months and in the case of early stage researchers (within 8 years of a PhD), even up to €3500 for longer periods.

I should at this point make a little note as to the various benefits of COST actions for our little nation. Many countries have two management committee members from two different labs. In our consortium, the management committee members are always funded in whatever conference or visit is organised, except when specific meetings for young scientists for example are organized when there is no management committee meeting . When possible, the Action also funds 1-2 participants from each other member (i.e. each lab or university involved in the action, since of course these may involve numerous labs from the same country). Since it is uncommon that too many researchers are involved in a particular area, in Malta and since one is allowed to have alternative management committee members, should one be able to attend, then it allows good funding locally for research staff, and even students to have the opportunity to present their work on a much wider stage.

In fact following the different talks by the group, collaborations initially started as contacts from individual labs who asked us to send certain chemicals for testing.

One of the earliest of these was Dr Danijel Kikelj who sent 12 chemicals for differentiation testing. Later Nadine Martinet (initially an invited speaker from INSERM in Nice in France and later a member of the consortium) asked all chemistry labs to contribute towards a large central chemical database which she administered and this was made available to all the various biology labs. As a result, we ended up screening over 600 chemicals (and still have more to work on) from numerous labs around Europe. Due to the large number of chemicals needing work-up and due to the limited hands on deck in terms of graduate students, more than 30 undergraduate students participated in this research, also offering undergraduate students (some of which are shown in Fig. 4) an opportunity to understand the research component of the University of Malta and contribute to international science. More than 80 very interesting hits (which cause leukaemia differentiation to different degrees) have been identified and many of these are being followed up.

The first 400 or so chemicals screened were presented in a fourth meeting in Budapest in March 2014 (no one from Malta attended the third meeting in Warsaw) and again this garnered a lot of attention with various labs interested in continuing collaborations. Some of the chemicals presented are indicated in Fig. 3 where different novel synthesised chemicals (named only by their catalogue number) are shown to be active in the screening method developed in our lab, again compared to positive and negative controls. The more active of these chemicals are then assessed by morphology – i.e. the effects of differentiation are assessed visually. Should this also be interesting, further testing can be done by means of flow cytometry (Fig. 5), locally to confirm the expression of differentiated chemistry markers. In fact this has resulted in students and post-docs from labs in Ireland and Serbia visiting our labs and more should be coming in the near future, from Greece and possibly elsewhere.

Figure 4: The students from the preclinical medical years who performed the bulk of the work in testing the first 400 chemicals on the STEMCHEM database.

Some of the chemicals from these labs have already shown activity in 2 or more assays and will be followed up for publication. The 80 or so hits from the first four hundred screen and all the new hits from the last 200 chemicals will also be assessed with morphology and flow cytometry and followed up accordingly with the

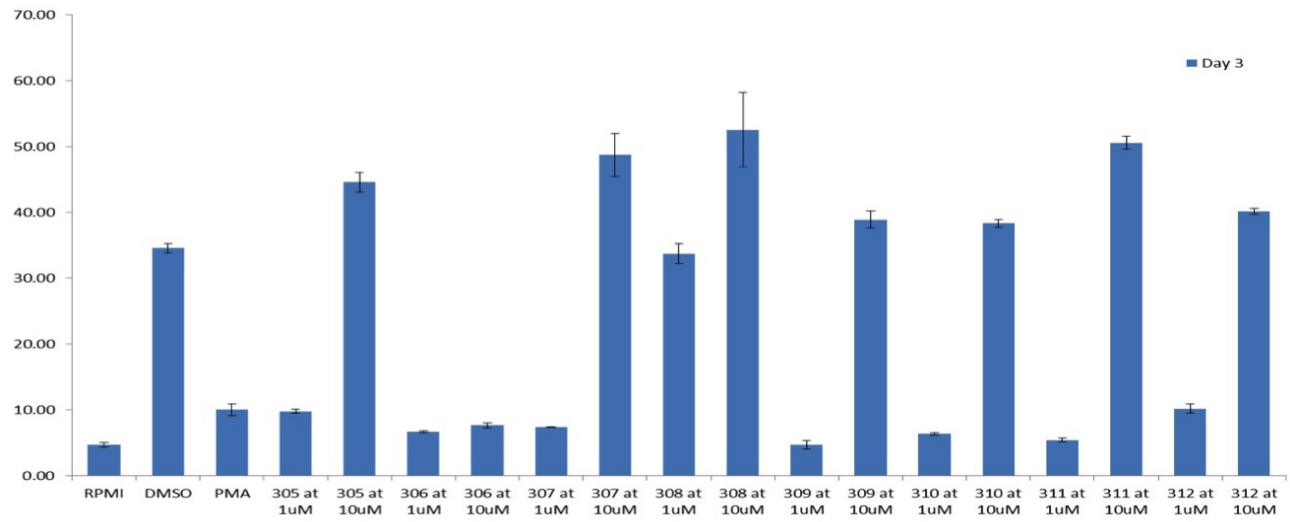

Figure 3: Some results out of the first 400 chemicals in the STEMCHEM chemical library, tested in the summer of 2013. Results show an index of differentiation created by dividing a leukocyte differentiation marker (NBTZ reduction function) by an indicator of cellular activity and thus of cell number (MTT activity). Results are an average of 3 replicates at each of two concentrations 1 and 10 mM. The first three columns show the results of negative control (normal culture medium) as well as two positive controls which however cannot be used in the clinic due to toxicity, PMA and DMSO.

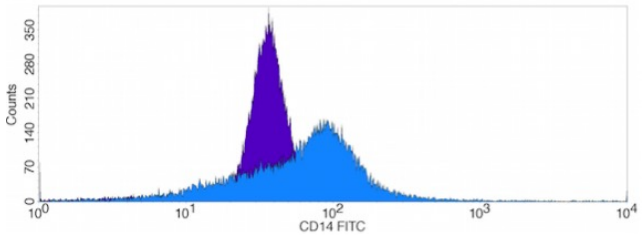

Figure 5: Changes in the expression of CD14 marker (monocytic differentiation) between untreated (violet) and treated (blue) HL60 populations.

chemists who produced them.

The common way that this is done is that the chemists make similar molecules to the hits and then these are once again screened to see which variations are even more active. The most active of these agents can then be tested on numerous leukaemia cell lines (we usually test 4 rather different myeloid leukaemias initially) and then can even be tested on primary leukaemia blasts from patients to see if the effect is also possible on patient cancers rather than the more artificial cell lines. All this information enhances the possibility of developing effective drugs for inducing differentiation, a rather novel kind of therapy for cancers, started a number of years ago with all-trans retinoic acid, which converted the normally fatal Acute promyelocytic leukaemia into a manageable disease where more than 90% of patients are cured nowadays.

Expanding this interesting branch of cancer therapy is one of the main interests of our lab at the Anatomy department and STEMCHEM has given us the capability to open up our expertise to collaborations all across Europe and to many more possible candidate drugs, apart from our own in-house derived extracts.

Another development in our own lab is the indication that many of these chemicals may also work on other solid tumours apart from leukaemias, especially the serious solid tumours of childhood. There is already evidence that all trans retinoic acid works to some extent on brain and bone tumours (Choschzick et al., 2014; Yang et al., 2012) but so do some of the extracts and drugs we are testing. In fact our preliminary work on osteosarcomas (bone tumours) was presented by another graduate student, Mr Sherif Suleiman (Fig. 6) at the last meeting of the Action in October in Tenerife where a good group of Action members enjoyed a meeting at one of Europe's more exotic destinations (Fig. 7).

Overall, COST actions offer extra funding for cash-strapped local research groups to travel, and train, and more importantly the opportunity to share data, discuss in a wider pan-European forum and set up collaborations which allow one to access larger sources of funding such as Horizon 2020 or Innovative Medicines

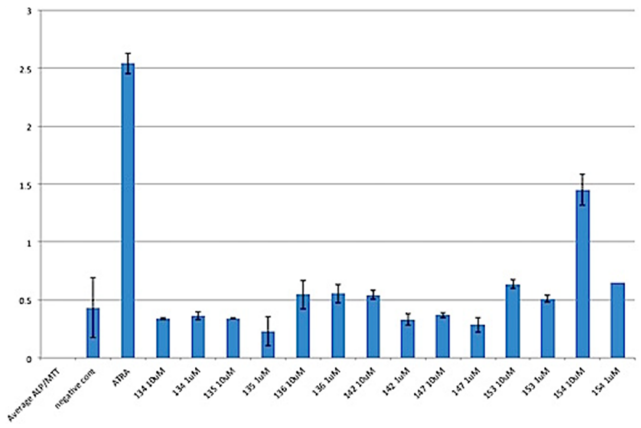

Figure 6: Changes in Alkaline phosphatase expression divided by indicator of total cell number assessing differentiation related changes in osteosarcoma.

Figure 7: The poster of the last action workshop meeting in Tenerife.

Initiative 2 funding. Importantly for our small country, they also provide an opportunity for students to travel and present their work. This was my first experience in COST but it will definitely not be my last.

Acknowledgments

The Authors would like to acknowledge the numerous laboratories who provided chemicals for testing to the STEMCHEM database and also to many other students (apart from those pictured), both Maltese and international who contributed to the screening of all of the chemicals.

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Review Article

Supramolecular Chemistry in Water: Self-Assembly of Multi-Component Fluorescent Molecular Logic Gates in Micelles

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Abstract. A recent strategy for developing supramolecular logic gates in water is based on combinations of molecules via self-assembly with surfactants, which eliminates the need for time-consuming synthesis. The self-assembly of surfactants and lumophores and receptors can result in interesting properties providing cooperative effects useful for molecular information processing and other potential applications such as drug delivery systems. This article highlights some of the recent advancements in supramolecular information processing using microheterogeneous media including micelles in aqueous solution.

Keywords: supramolecular chemistry in water, molecular recognition, chemosensors, micelles, molecular logic gates, biomedical diagnostics

1 Introduction

The objective of COST Action CM1005 is the development of supramolecular systems that work in water (Oshovsky, Reinhoudt & Verboom, 2007; Zayed, Nouvel, Rauwald & Scherman, 2010, http://supracheminwater.wordpress.com/). The COST Action is divided into three working groups (i) the molecular recognition of biologically and environmentally relevant species in water (ii) the selective control of reactions in water, and (iii) the self-assembly of organized structures in water that are stimuli responsive and can be used for programming functions in materials and devices. The Action aims to improve the understanding of multiple non-covalent weak bonds (hydrogen bonding, electrostatics, Van der Waals forces, pi-pi interactions etc.) that are collectively powerful interactions for selective recognition of chemical analytes and processes in water.

The majority of molecular receptors for recognition of physiologically important cations, anions and neutral analytes are not readily soluble in water (Magri & Mallia, 2013; Schneider, 2013). One working group within the COST Action is designing and synthesising novel intelligent molecules readily soluble in water, which is not always an easy task even for skilled organic chemists (Magri, 2012). A simple way to circumventing the issue of poor solubility of receptors in water is to incorporate them in micelles to form water-soluble nanoscale supramolecular devices (Pallavicini, Diaz-Fernandez & Pasotti, 2009). Micelles result from the spontaneous association of surfactants to form dynamic spherical conglomerates above the critical micelle concentration (cmc), and other shaped assemblies at higher concentrations, which are representative biomimetic models of biological membranes (Turro, Grätzel & Braun, 1980). In the case of ionic micelles, the micelle interface has an electrical double layer and a potential difference on the order of several hundred millivolts. The electric field can modulate the sensitivity of ion determination due to an amplifying effect on the local ion concentration, Moreover the receptor-micelle nanodevices often show enhanced binding properties as will be discussed.

This article highlights examples of supramolecular multicomponent systems with stimuli-responsive properties that perform molecular computation-based logic (de Silva, 2013; Szaciłowski, 2008). The examples illustrated are presented according to increasing complexity of the logic system (de Silva & Uchiyama, 2007). A common theme throughout is the use of micelle media, which introduces synergistic effects. Examples are included representative of fluorescent sensing devices for various kinds of chemical species as inputs including protons, cations and anions. Readers with a desire for background literature on fluorescent probes can view the cited references (Bissell, Bryan, de Silva & McCoy, 1994; Callan, De Silva & Magri, 2005; de Silva et al., 1997; Valeur & Berberan-Santos, 2012).

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2 Single-input Logic Gates

There are four possible single-input logic gates: PASS 0, PASS 1, YES and NOT. PASS 0 is the simplest of Boolean logic operations and appears trivial. Any molecule that is non-fluorescent remains so independent of the absence or presence of an input. PASS 1 is another trivial logic gate exemplified by a fluorophore that emits fluorescence on excitation independent of the absence or presence of an input. The design of 'fluorophore–spacer–receptor' and 'fluorophore–receptor' molecules allows for YES and NOT logic to be demonstrated. The standard molecular YES logic gate is based on the competition between photoinduced electron transfer (PET) and fluorescence yielding an *off-on* switching action of the fluorescence intensity ideally with no change in the wavelength (de Silva et al. 2009). A NOT gate, also referred to an inverter operates by *on-off* switching.

Akashi's team demonstrated a viable way for detecting barium by using an ether crown-based fluorophore **1** in aqueous solution (Nakahara, Kida, Nakatsuji & Akashi, 2004). The pyrene-functionalised monoaza-18-crown-6 ether derivative is a 'fluorophore-spacer-receptor' system with poor water solubility and Ba^{2+} binding properties in water. Addition of the non-charged detergent Triton X-100 above the cmc allows the chemosensor to position itself in the less polar micellar location, yielding a supramolecular assembly which results in binding of Ba^{2+} by the cryptand. The amino nitrogen atom is involved in the complexation of Ba^{2+}, which cancels the PET from the tertiary amine to the pyrene fluorophore with a high fluorescence output. Although the experiment is conducted at pH 10 due to the sensitivity of **1**, the strategy exemplifies a selective way of detecting barium by YES logic.

Bhattacharya and Gulyani are perhaps the first to develop the concept of multifunctional hydrophobic probe design (Bhattacharya & Gulyani, 2003). The method was demonstrated by detecting for Zn^{2+} in micelles and vesicles with 1-pyrenyl-methyl-*bis*(2-picolyl) amine **2**. In water the chemosensor aggregates as observed by an excimer emission about 500 nm. In micelles, however, aggregation of the probe molecule **2** is prevented such that no excimer emission is observed, while the monomer emission in the presence of Zn^{2+} at 400 nm is substantially enhanced. Large fluorescence enhancements were observed in polyoxyethylene (20) sorbitol monolaurate (Tween 20) micelles and in dipalmitoyl phosphatidylcholine vesicles.

A salophen-UO_2 complex has been demonstrated to exhibit a remarkable increase in binding of fluoride in CTAB micelles (Cametti, Dalla Cort & Bartik, 2008). In water alone, the salophen-UO_2 complex is not soluble. UV-visible titration studies of **3** in 50 mM CTAB

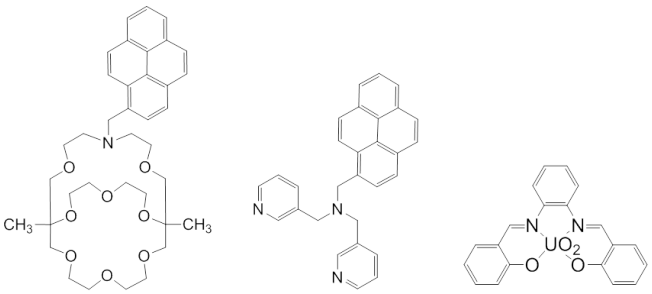


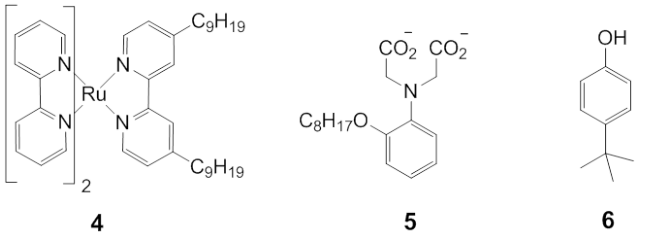
Figure 1: Chemosensors for detection Ba^{2+} **1** and Zn^{2+} **2** and F^- **3** with the assistance of micelles.

were consistent with a 1:1 binding isotherm with fluoride with a binding constant of 10 800 M^{-1}. Sulfate, acetate and phosphate also bind, but one to two orders of magnitude lower. An understanding of the spatial orientation of the salophen-UO_2 receptor **3** in the micelle was determined by NMR paramagnetic relaxation enhancement (PRE) and Nuclear Overhauser Effect (NOE) measurements (Keymeulen, De Bernardin, Dalla Cort & Bartik, 2013). It was discovered that **3** preferentially locates near the micelle interface orientated with the oxygen-linked aromatic rings facing the bulk aqueous solution and the nitrogen-linked phenyl ring backed into the hydrophobic core. The techniques presented by the collaboration of Dalla Cort and Bartik bring a fresh perspective with respect to shedding light on the location and spatial orientation of probes in miceller media. PRE and NOE experiments could be used to complement fluorescent mapping studies near micellar membranes (Bissell et al., 1994; Uchiyama, Iwai & de Silva, 2008).

By self-assembly of a lumophore and receptors with micelles, logic gates can be constructed in a 'plug and play' fashion (de Silva, Dobbin, Vance & Wannalerse, 2009). Triton X-100 is used to solubilize a hydrophobic tris(2,2'-bipyridyl)Ru(II) complex **4**, a lumophore with both a long excitation state lifetime of 200 ns and a long emission wavelength about 625 nm. The elemental PASS 0 and PASS 1 logic gates were mentioned as the micelle alone and the micelle containing **4**. YES logic is demonstrated using a 2-nitrophenyl-*n*-octyl ether receptor **5**, which is emissive on protonation of the aromatic amine at pH 2. Ligand **5** also binds Ca^{2+} at pH of 8 with YES logic behavior by a five-fold emission enhancement. This approach of using separate components for the lumophore and receptors allows for the configuration of new modules enabling new functions in the supramolecular ensemble. To reiterate, the microheterogeneous media is an essential component for enhanced luminescence to be observed.

Table 1: General truth tables for seven two-input logic gates.

$Input_1$	$Input_2$	AND	NAND	OR	NOR	XOR	XNOR	INH
0	0	0	1	0	1	0	1	0
0	1	0	1	1	0	1	0	0
1	0	0	1	1	0	1	0	1
1	1	1	0	1	0	0	1	0



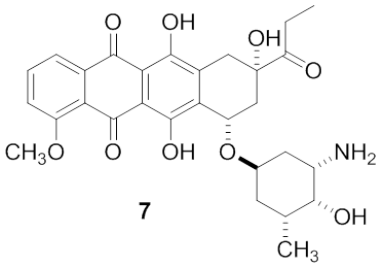
4 5 6

Figure 2: Components **4**–**6** for a supramolecular 'plug and play' logic device.

3 Two-Input Logic Gates

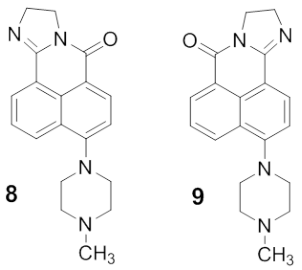
Double input logic gates were also demonstrated in Triton X-100 surfactant with **4** and **5** (de Silva et al., 2009). For referral, Table 1 summarises the two-input logic for seven types of logic gates. When both H^+ and Ca^{2+} are present as inputs, the assembly is an OR logic gate as the presence of either input or both provides a fluorescence output. AND logic is observed when *para-tert*-butylphenol **6** is added as a new module to the assembly. In basic solution, **6** is deprotonated to the negatively charged phenolate, which acts as an electron donor to the excited ruthenium complex rendering the luminescence *off* at pH 12. However, on decreasing the pH to 8, a six-fold luminescence improvement is observed at 625 nm.

A stimuli-responsive polymeric micelle was demonstrated by the Wang group (Wei, Guo & Wang, 2011) as a novel strategy for developing an intelligent drug delivery system (Alvarez-Lorenzo, Bromberg & Concheiro, 2009). Certain tumor cells are known to have characteristically high reductive environments and high proton concentrations. The Wang team developed polymeric crosslinked micelles with Andriamycin **7** conjugated to the micelles. The pH and reduction conditions are the key stimuli-based parameters for an AND logic result. The drug is initially doubly trapped in the micellar system by hydrazone and disulfide bonds. Drug release is achieved at pH 4 and in the presence of 15 mM of the redox agent, dithiothreitol. Addition of acid cleaves the hydrazone bonds while dithiothreitol cleaves the disulfide bonds. When both input chemicals are present, **6** is severed from the polymers, which disperse as smaller fragments. Liberation of the drug by both stimuli allows for selective release of the drug at the target tumor cells.



7

Figure 3: The molecular structure of the anti-cancer drug Adriamycin **7**.



8 9

Figure 4: The molecular structures of the naphthalimide regioisomers **8** and **9**.

The team of Qian demonstrates up to ten logic functions with the reconfigurable molecules **8** and **9** in water, and extends the use of sodium dodecyl sulphate (SDS) surfactant as an additional input using both absorbance and fluorescence outputs (Qian, Qian, Xu & Zhang, 2008). Six two-input gates are configurable for AND, NAND, OR, NOR, XNOR, INHIBIT logic in additional to the four one-input gates. The versatility of these naphthalimide-based molecules for logic applications is due to the two accessible sites of protonation according to a 'receptor$_1$-fluorophore-spacer-receptor$_2$' design (Zammit, Pappova, Zammit, Gabarretta & Magri, 2015). Though regioisomers, the fluorescence quantum yields of **8** and **9** are significantly different at 0.218 and 0.055 in water; however, implemented as logic devices the characteristics are similar. Addition of anionic SDS (low 0, high 8.2 mM) and hydroxide (pOH of 7 and 4) provides INH and XOR using the absorbance

at 425 nm and negative logic convention for the fluorescence output to form the basis of a half-subtractor. Dual protonation of both compounds provides pathways that change the absorbance and emission spectra, as well as the interaction of SDS below and above the cmc.

The theme of naphthalimides and SDS surfactant and protons is continued with the addition of using the inorganic salt Na_2SO_4 as an alternative input (Qian et al., 2008). With inputs SDS and Na_2SO_4, OR and AND logic are exhibited for **10** and **11**, respectively. XOR and INH logic gates can also be interpreted from the output **11** due to the additional PET pathway from the tertiary amine. Exploiting both the PET and ICT push-pull channels, the authors share their interpretation of a half-adder and half-subtractor functions (Pischel, 2007).

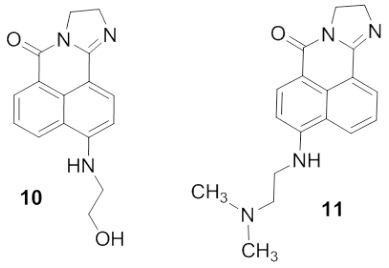


Figure 5: Examples of a two-input OR gate **10** and AND gate **11** with SDS and Na_2SO_4 as inputs.

The team of Uchiyama and de Silva demonstrated the first example of dual-input molecular computation within a small defined nanospace of 3 nm volume (Uchiyama, McClean, Iwai & de Silva, 2005). A lipophilic molecule **12** consisting of a benzo-5-crown-15 ether, an anthracene fluorophore, a tertiary amine and an octyl hydrocarbon chain was used as a probe of the micelle environments. The molecular device contains two classic electron donors used in PET systems. Self-assembly of the molecular probe in cationic and neutral micelles of cetyltrimethylammonium chloride (CTAC), octyl β-D glucopyranoside and Triton X-100 yielded no observable fluorescence response. However, in tetramethylammonium dodecyl sulfate (TMADS) micelles the molecular probe exhibits a ten-fold fluorescent enhancement in the presence of H^+ and Na^+ at elevated concentrations. The reason is the binding constant of benzo-5-crown-15 ether, at only $\log K$ of -0.3 in water, increases by two order of magnitudes ($\log K = 1.9$) due to the local concentration of Na^+ at the micelle interface. At pH 3 and 0.4 M sodium ions, the H^+ and Na^+ input concentrations are high resulting in a substantial fluorescent output due to the sodium ions binding to the benzo-5-crown ether and the protons to the tertiary amine, which in both cases, prevents PET to the anthracene fluorophore reminiscent to a AND logic gate. The strategy illustrates the ability to sense within a nanometer radius (a dimension where silicon-based electronic devices cannot approach) and opens up the possibility of molecular computation in other microheterogeneous (i.e. liposomes and vesicles) and biological systems.

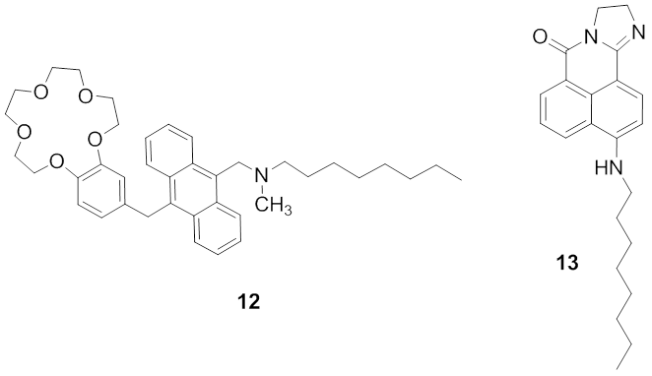


Figure 6: Hydrophobic molecular probes **12** and **13** for information retrieval in nanospaces.

4 Three-input Logic Gates

The first reported example of the potential cross-fertilization between Boolean algebra and biomedical sensing was reported for a three-input AND 'lab-on-a-molecule' based on a competition between PET and fluorescence (Magri, Brown, McClean & de Silva, 2006). In this instance, three receptors are incorporated within a single molecule: a benzo-15-crown-5 ether for Na^+, a tertiary amine for H^+, and a phenyliminodiacetate for Zn^{2+}. The modular covalent arrangement of the receptors, spacers and fluorophore facilitates the cooperative sensing algorithm (Magri & de Silva, 2010). Consideration of **12** with the micelle as one of the inputs illustrates a supramolecular system as an example of a 3-input AND logic gate with the inputs Na^+, pH and TMADS.

Three-input IMPLICATION logic is demonstrated using a naphthalimide probe with an octyl hydrocarbon chain **13** (Qian, Xu, Zhang & Qian, 2011). This type of logic is similar to an IF-THEN operation. However, in this example the fluorescence is modulated by the inputs SDS, CTAC and temperature. The molecule is fluorescent in water with a quantum yield of 0.135. Addition of SDS just below the cmc concentration $\sim$ 8.0 mM quenches the fluorescence. Subsequent addition of 100 µM cetyltrimethylammonium bromide (CTAB) causes a 25-fold fluorescence enhancement. The rational for these observations is that the opposite charges of SDS and CTAB in addition to the hydrophobic alkyl chains common to both surfactants results in aggregation between the two micelles, and consequently, liberation of the fluorescence probe. An increase in temperature enhances the fluorescence by making the probe molecule more soluble. The net outcome is that in the

Table 2: Truth table for the supramolecular three-input AND logic gate **12**.

$Input_1$	$Input_2$	$Input_3$	Output
Na^+ [a]	H^+ [b]	TMADS [c]	Fluorescence
0 (low)	0 (low)	0 (low)	0 (low)
0 (low)	0 (high)	1 (low)	0 (low)
0 (low)	1 (low)	0 (high)	0 (low)
0 (low)	1 (high)	1 (high)	0 (low)
1 (high)	0 (low)	0 (low)	0 (low)
1 (high)	0 (low)	1 (high)	0 (low)
1 (high)	1 (high)	0 (low)	0 (low)
1 (high)	1 (high)	1 (high)	1 (high)

[a]High input level of 0.4 M of NaCl. Low input level maintained with no added NaCl. [b]High input level 10^{-3} M acid. Low input level 10^{-11} M acid. [c]High input level of 20 mM TMADS. Low input level no TMADS.

Table 3: Truth table for the supramolecular three-input IMPLICATION logic gate **13**.

$Input_1$	$Input_2$	$Input_3$	Output
SDS [a]	CTAB [b]	T [c]	Fluorescence
0 (low)	0 (low)	0 (low)	1 (high)
0 (low)	0 (high)	1 (low)	1 (high)
0 (low)	1 (low)	0 (high)	1 (high)
0 (low)	1 (high)	1 (high)	1 (high)
1 (high)	0 (low)	0 (low)	0 (low)
1 (high)	0 (low)	1 (high)	1 (high)
1 (high)	1 (high)	0 (low)	1 (high)
1 (high)	1 (high)	1 (high)	1 (high)

[a]High input level of 10 µM of SDS. Low input level maintained with no added SDS. [b]High input level 20 µM CTAB. Low input level with no CTAB added. [c]High input level at 75 °C and low input level at 25 °C.

presence of SDS alone, the fluorescence (and absorbance) is low, while in the other seven combination of SDS, CTAB and temperature, the fluorescence in high (Table 3). We recently reported a colorimetric and fluorimetric inverted enabled OR logic array with CTAC, Triton X-100 and hydroxide as inputs using a rhodamine B probe derivatised with a hexane chain (Caruana, Camilleri Fava & Magri, 2015).

5 Multi-level Logic

In the previous section supramolecular systems were illustrated that detect for three input conditions and with only two output results of either a low fluorescence or a high fluorescence. Now we demonstrate systems with three output levels. Traditionally, the design principle for these systems is based on 'fluorophore-spacer$_1$-receptor$_1$-spacer$_2$-receptor$_2$' and 'receptor$_1$-spacer$_2$-fluorophore-spacer$_2$-receptor$_2$' strategies to develop *off-on-off* ternary systems (de Silva, Gunaratne & McCoy, 1996). At a low input level the switch is *off*, at a medium input level the switch is *on*, and at a high input level the switch is *off* again (Pais et al., 2013). The regulation of analytes in living things is governed by ternary logic. Too little or too much of analyte results in illness, and in extreme situation even death. Thus, good health requires the right balance of each analyte within a specific concentration range (Burtis & Ashwood, 2001).

Off-on-off systems were first demonstrated with **14** based on a fluorophore-spacer$_1$-receptor$_1$-spacer$_2$-receptor$_2$' design consisting of an anthracene fluorophore, a tertiary amine and pyridine as the receptors (de Silva et al., 1996). Pallavicini also demonstrates an easy-to-assemble approach with no synthetic effort in a 'plug and plug' fashion (Pallavicini et al., 2009). In the analogous supramolecular version, the fluorophore is pyrene **15** and the two receptors are the lipophilic bases *N*,*N*-dimethyl-*N*-dodecylamine **16** and 2-dodecylpyridine **17**. Assembled in Triton X-100 as the surfactant, and anionic SDS as the co-surfactant (at various concentrations), the multicomponent system is a tuneable *off-on-off* micellar sensor device with the capability of shifting the *on* window along the pH axis with the curve apex ranging between pH 5 to 10. In another *off-on-off* example from the Pallavicini group, the polyaspartamide based co-polymer, PHEA-PEG$_{5000}$C$_{16}$ is used as the surfactant and SDS and CTAC as the co-surfactants (Diaz-Fernandez et al., 2010).

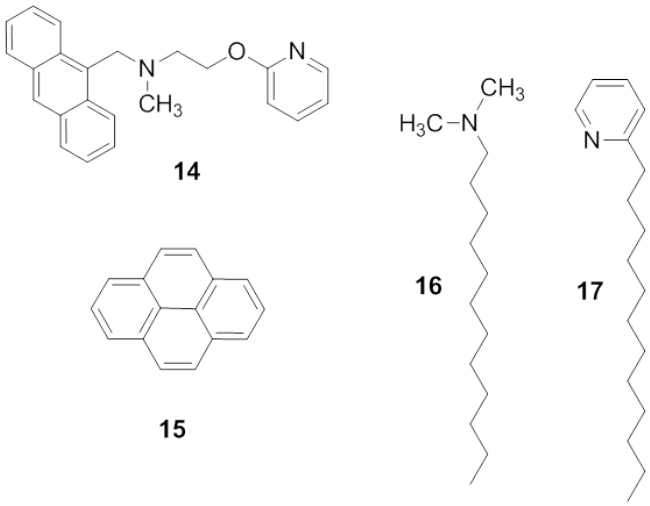


Figure 7: An *off-on-off* molecular device **14** and the components of a supramolecular device consisting of pyrene **15**, *N*,*N*-dimethyl-*N*-dodecylamine **16** and 2-dodecylpyridine **17**.

Das reported pH dependent fluorescence switching of salicylideneaniline in micelles according to *on-off*, *off-on*, and *off-on-off*. pH profiles (Das & Dutta, 2014). Salicylideneaniline **18** behaves as an *off-on* switch in 1:1 acetonitrile/H_2O and 3% negatively charged SDS aqueous solution. At pH 6 or lower, the fluorescence is *off* while at pH 10 the fluorescent is *on*. However, in CTAB and Triton X-100 ternary *off-on-off* behaviour is admirably observed with the fluorescence turning *off* pH 10. In CTAB a distinct *on* pH window is observed between pH 7–11. The differing chemistry is attributed to the equilibrium between the keto (fluorescent) form **18** and the enol form (non-fluorescent) **19** by acid and base catalysis.

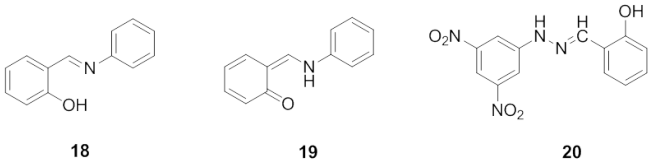


Figure 8: Examples of hydrazones **18** and **20** with *off-on-off* behaviour in micelles.

In another recent study, Goswami and Das also report the dinitrophenolhydrazone derivative **20** in 1:1 CH_3OH:H_2O, SDS, CTAB and Triton X-100 (Goswami & Das, 2011). In 1:1 CH_3OH:H_2O, a broad pH window from 5 to 12 is observed with *off-on-off* behaviour, while with CTAB a much narrow pH window is observed between 4 and 7. In SDS a *low-medium-high* response is observed on increasing pH. The sites of protonation are thought to be the phenol and the secondary amine. At pH 12 fluorescence quenching results from the phenolate to the 2,4-dinitrophenyl moiety. Below pH 6, fluorescence quenching is postulated to result from protonation of the dinitroanilic nitrogen, which lowers the oxidation further allowing for excited electron transfer from the phenol.

Pallavicini have both independently demonstrated *on-off-on* pH window sensing molecular devices (Denat, Diaz-Fernandez, Pasotti, Sok & Pallavicini, 2010). A multicomponent approach consisting of Coumarin 343 **21**, Cu^{2+} ions and *N*-dodecylated trimethylcyclen **22** are self-assembled in Triton X-100 micelles. At low pH the two organic components do not interact, and the fluorescence from 21 is high. At intermediate pH, 21 is deprotonated and coordinates to Cu^{2+} ions resulting in fluorescence quenching. At high pH, the carboxy end of **21** is displaced from Cu^{2+} by the formation of complex with hydroxide reviving the fluorescence. In both examples, the *off* window is between pH 6 and 8, which is the physiological pH sweet spot of 6.8 to 7.4. The lipophilicity of nonsteroidal anti-inflammatory drugs (NSAIDs) is also measurable by expressing an *off-on* fluorescent signal correlating the fluorescence increase with the logarithmic water/octanol partition coefficient ($\log P$).

The emissive and absorptive properties of **13** and other related members of the naphthalimide-based fluorescence sensors were investigated as chromogenic and fluorogenic sensors for anionic surfactants (Qian, Qian & Xu, 2009). The probe **13** is an *on-off-on* fluorescence sensor for SDS. Interpretation of the spectroscopic output provided for multiple output readouts at 430 nm by UV-visible absorption and 525 nm by fluorescence spectroscopy with SDS, CTAB and Triton X-100 allowing for a sensor array, which also discriminates SDS at different concentration ranges. The octyl hydrocarbon chain was found to be an important parameter as other model probes with butyl and dodeceyl hydrocarbon chains exhibited inferior emission switching properties.

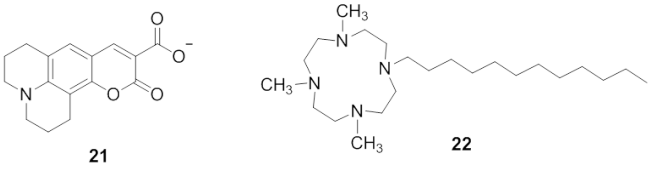


Figure 9: The components Coumarin 343 **21** and *N*-dodecylated trimethylcyclen **22** as part of an *on-off-on* supramolecular nanodevice for gauging drug lipophilicity.

6 Conclusion

The self-assembly of surfactants provides an alternative strategy for information processing applications at the molecular level. Supramolecular systems can be designed that respond to various chemical inputs such as cations, anions, pH as well as physiochemical parameters. Advantageously, supramolecular assemblies require minimal synthetic effort. Most of the one- and two- inputs logic gates have been demonstrated in micellar media as well as examples of supramolecular systems that exhibit *off-on-off* and *on-off-on* profiles within narrow pH windows and *low-medium-high* ternary pH profiles on sequential addition of proton inputs. Applications in drug delivery and smart materials are just a sliver of potential uses.

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Research Article

Accessibility as an indicator of transport equity. The case of public transport infrastructure in Malta, and its impact on the elderly

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Abstract. The concept of equity is essential in transport because inequities lead to the formation of transport-disadvantaged groups, such as the elderly, disabled and low-income people. This paper focuses on the elderly. Due to age-related circumstances, several elderly persons have to surrender on driving, consequently they become highly dependent on public transport. Hence, accessible public transport is crucial to provide them with the necessary mobility. This research considers accessibility as a key indicator for transport equity, since the latter primarily deals with the provision of equal access to opportunities. The study focuses on the case of Malta's public transport system, which is composed of the bus service. The uniqueness of the Maltese case is that transport policy is fragmented, and is not focused on equity. This paper looks at three aspects of accessibility related to road infrastructure, public transport infrastructure, and the bus fleet. The first aspect refers to accessibility at the macro scale, for instance, pavements may not be solely designed to cater for the bus service, but they are an integrative part of it. The meso scale refers to accessibility of infrastructure in physical and cyber form, such as access to and on bus stops and access to online travel information. The bus fleet refers to the micro scale of accessibility, which may include boarding and alighting the vehicle, and access on the vehicle. The research approach involves a review of existing Maltese public transport policy, with specific focus on whether accessibility for the elderly is considered in the context of the afore-mentioned scales. It is envisaged that the minimal or non-existent policy on accessibility in public transport that focuses on elderly, makes this population segment at a double disadvantage. The research concludes with implications for policy related to public transport accessibility in a Maltese ageing society.

Keywords: transport equity, accessibility, public transport infrastructure, elderly people, transport policies for elderly, Malta

1 Introduction

Accessibility refers to the ability of reaching goods, services, and destinations. It is linked with mobility, which provides the opportunity for people to move from an origin to a destination (Litman, 2016). Hence, accessibility and mobility are two interdependent concepts that encourage independent living (Suen & Mitchell, 2000).

Accessibility is a necessity for people to reach their destinations, whether they are daily commuters or not. A non-commuting group is the elderly population. Due to age-related circumstances, several elderly persons have to surrender on driving, consequently they become highly dependent on public transport (Whelan, Langford, Oxley, Koppel & Charlton, 2006). This is one reason why elderly are one of the transport disadvantaged groups in society (Wixey, Jones, Lucas & Aldridge, 2005). In fact, older people use public transport more than younger generations (Goodwin & Lyons, 2010).

Hence, equity is essential in public transport because it ensures that the population segments that are at a disadvantage are provided with the same opportunities as other population segments. In fact, the concept of transport equity is built upon connecting citizens to key activity destinations by means of public and private transport infrastructure (Di Ciommo & Lucas, 2014). Consequently, it is necessary to include the assessment

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of equity as part of the monitoring programme of a bus service. Two important factors that are used to gauge equity are accessibility and mobility (Litman, 2016).

This research focuses in particular on public transport accessibility as a key indicator for transport equity. It seeks to identify the availability of transport policy on different levels of the bus system's infrastructural accessibility. The first level refers to accessibility at the macro scale, for instance, pavements may not be solely designed to cater for the bus service, but they are an integrative part of it. The meso scale refers to accessibility of infrastructure in physical and cyber form, such as access to and on bus stops and access to online travel information. The bus fleet refers to the micro scale of accessibility, which may include boarding and alighting the vehicle, and access on the vehicle.

The case study is Malta's public transport system, namely the bus service. Hence, the premise for this research is that an accessible bus service is crucial to provide the elderly with the necessary mobility that retains their independence.

The Maltese case provides the opportunity to explore a fragmented transport policy in which equity is not at the top of the policy agenda. This makes the elderly segment at a double disadvantage. The paper concludes with implications for policy related to public transport accessibility in a Maltese ageing society.

Malta has several geo-demographic characteristics that make it a good candidate to have high public transport patronage. However, it is not the case as the modal split is 75 per cent car users and 15 per cent bus users (Transport Malta, 2010). Such factors include a population of 0.4 of a million residing on a land area of only 316 km^2, one of the highest population densities in the EU (1,317 persons per km^2). Moreover, with particular relevance to this paper, Malta has an ageing population that is increasing at a fast rate. For the first time in history the 65+ age group in Malta is exceeding the 0–14 age group (1901: 0–14 age group - 34.1%, 65+ age group - 5.4%; 2012: 0–14 age group 14.5%, 65+ age group - 17.2%) (National Commission for Active Ageing, 2013). Fig. 1 illustrates the distribution of the elderly population in Malta in 2011. The Northern Harbour District, followed by the Southern Harbour District has the highest elderly population. Malta's conurbation is found in these districts.

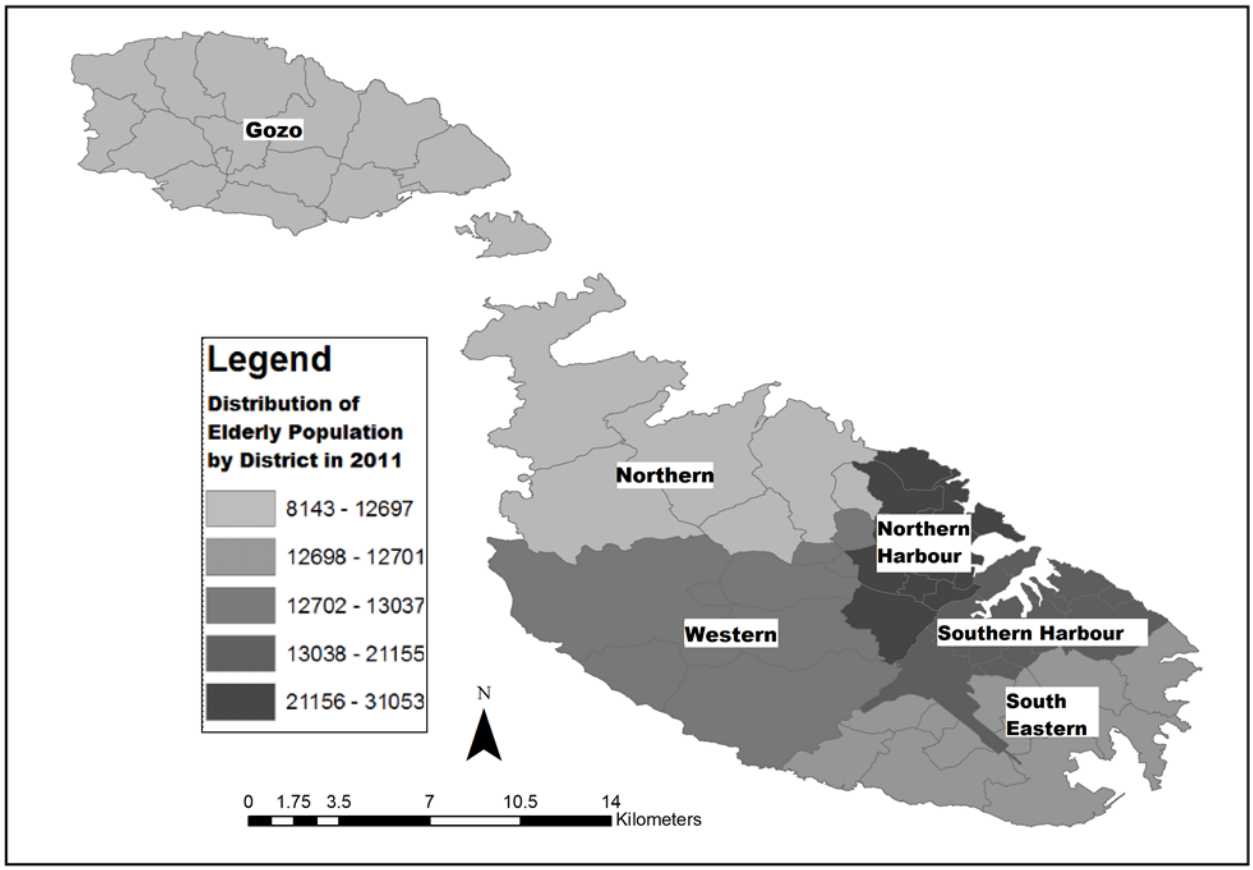

Figure 1: Map of Malta indicating the distribution of the Elderly Population by District in 2011 (Adapted from National Statistics Office, 2012a).

The paper is organised in five sections. Following this introduction, the second section provides a background literature review on transport equity in the context of elderly people as a socially excluded group, and referring in particular to work on accessibility. Section three provides an overview of the case study, the bus service in Malta. Section four explores the evaluation of Maltese transport policy in the context of elderly people and accessibility related to bus use. The fifth section provides the discussion and conclusion in view of land transport policy in Malta that relates to evaluation discussed in section four.

2 Literature Review

The concept of equity in transport research is relatively new (Trinder, Hay, Dignan, Else & Skorupski, 1991; Banister, 2000; Lucas, 2006; Martens, 2006; Mavoa, Witten, McCreanor & O'Sullivan, 2012). It has been classified into two dimensions. The first one is 'horizontal equity', which refers to an equal distribution of resources between individuals; and the second one is 'vertical equity', whereby resources are distributed according to similar abilities and needs (Litman, 2016; COST, 2012; Martens, Golub & Robinson, 2012). This study refers to 'vertical equity', because it focuses on elderly people who are a segment of the population with the same capabilities and requirements.

2.1 Transport Equity

Transport equity is considered as a way of providing social justice (Martens, 2006), and where transport equity is missing in terms of transport distribution, social exclusion takes place (Lucas, 2012). Factors that are used to gauge equity in public transport are system reliability, environmental impact (Bocarejo S. & Oviedo H., 2012), and accessibility, which affects the opportunities and capabilities of individuals to use the bus (Litman, 2016). This research focuses solely on accessibility because it is an important factor for elderly persons, since an inaccessible bus service impairs their mobility (Hanson & Giuliano, 2004).

2.2 The elderly as a socially excluded group

Older people who use public transport can become socially excluded due to limitations regarding the choice of other modes. Restricting factors include: age, income or lack of access to private transport (Beimborn, Greenwald & Jin, 2003).

Opportunities for various demographic groups are often reduced due to a reduction in accessibility, affordability and availability of transport (Church, Frost & Sullivan, 2000; Sen, 2000; Wixey et al., 2005). Elderly women tend to suffer more than men as they are more likely to spend more time relying on public transport after retirement (Foley, Heimovitz, Guralnik & Brock, 2002; Stutts, Wilkins, Reinfurt, Rodgman & Van Heusen-Causey, 2001).

2.3 Elderly and public transport accessibility

Social injustice is experienced when, for example, elderly people suffer from difficulties in mobility and feel insecure while waiting for the bus (Dunbar, Holland & Maylor, 2004). Such problems lead to inaccessibility, which hinders the quality of life of elderly people (Peel, Westmoreland & Steinberg, 2002; Hess, 2009; Frye, 2012). This can lead to social isolation, depression, and general health deterioration (Marottoli et al., 1997; Victor, Scambler, Bowling & Bond, 2005; Siren & Hakamies-Blomqvist, 2009). Hence, in an ageing country like Malta, it is important to consider and plan for equity in the public transport system, and gauge equity through factors such as transport accessibility.

Accessibility can be measured using different factors. The first factor is infrastructure accessibility. Pedestrians require accessible walkways, suitable traffic signals and street crossings (Suen & Mitchell, 2000). Older persons appreciate an accessible walking environment with pedestrian crossings and signs, much more than younger adults. They are more cautious and try to avoid crossing roads without pedestrian facilities (Bernhoft & Carstensen, 2008).

Additionally, accessibility comprises the ability to move from one bus stop to another, within a specific timeframe, particularly if a person is interchanging from one mode to another. In fact, the distance to bus stop, waiting time and ease of transfers are major factors that attract elderly persons to use public transport (Wardman, 2001). Since people in public transport services often cite the elderly population as one of the major rider segments (Carr, 2003) it is important that such infrastructure is suitable to accommodate the elderly.

Another key concept is knowledge. Bus users must be well-informed about the service before scheduling a trip, such as knowing the location of the bus stop and travel times (Beimborn et al., 2003). Information can attract more people to use public transport (Beirão & Sarsfield Cabral, 2007). A study carried out in Luqa, Malta, identified that lack of information was one of the factors that hindered elderly persons from using the bus service (Mifsud, 2013).

Moreover, people should find it easy to board on and off the vehicle (Beimborn et al., 2003); particularly old persons who generally suffer from health problems such as, arthritis, rheumatism and cardiac conditions (Smith, 2001). In Nigeria, 46 per cent of the transport constraints for elderly were related to boarding problems and inappropriate vehicle conditions (Olawole & Aloba, 2014), such as absence of low floor buses (Wixey et al., 2005). The feelings of resentment from other passengers if old persons take too long to access the vehicle are an-

other common problem that elderly people face when using public transport (Wixey et al., 2005).

2.4 Polices on Transport Equity

Different countries have adopted various policies and programmes related to elderly mobility and accessibility. In the United States equity in public transport provision is required by the legislation SAFETEA[1] (Delbosc & Currie, 2011).

The ECMT[2] has identified these main policy areas: ensure an accessible mobility environment and legislative reforms that address elderly transport issues, such as improving accessibility to public transport (European Conference of Ministers of Transport Council of Ministers, 2003), and monitoring the progress of accessibility policies (European Conference of Ministers of Transport Council of Ministers, 2006). Moreover, the European Commission (2011) acknowledges the difficulties that elderly persons encounter in their walking environment, and highlights the need to improve the accessibility of transport infrastructure for elderly and disabled passengers.

The current generation of elderly people is healthier than prior ones, and they have a more mobile lifestyle. However, there is still need to focus policies on the ageing population. Such policies are often lacking, as often only short-term goals of transport are considered. In Ontario (Canada), for instance, the ageing population is not even considered in transport policies on long-term basis (Mercado, Páez & Newbold, 2010). This is due to political and economic bias. Priority is given to economic and environmental issues, leaving the ageing population perspective behind. Additionally, most transport policies for elderly people are just related to private cars (example, screening drivers to analyse whether they should stop driving) (Mercado, Paéz, Scott, Newbold & Kanaroglou, 2007).

Furthermore, most of the current policies related to elderly in the transport environment are just concerned with disability aspects. In Ontario, the AODA[3] published in 2005, aims that by 2025 the province's infrastructure is accessible to elderly with impairments. Transport policies should go beyond limiting the elderly within the policy framework of disabled persons (Mercado et al., 2010). In fact, developed countries, which take primarily into consideration the needs of the elderly population, such as Japan serve the general public better. However, when referring to a public transport service one needs to consider the particular context and necessities of the country (Mercado et al., 2007).

In 2013, Malta launched *The National Strategic Policy for Active Ageing* 2014–2020 (National Commission for Active Ageing, 2013). This shows how lack of access often leads to social exclusion. Unfortunately, although the policy tackles independent living amongst the elderly, the transport dimension is not given detailed consideration. Hence, although Malta has a projected increase in the elderly population, it is clear that more national plans dealing with this population segment's transport necessities are lacking. Therefore, in 2012 the University of Malta joined the Transport Equity Analysis: assessment and integration of equity criteria in transportation planning (TEA) Cost Action №1209 to acquire an understanding on the equity implications of transport policies. This is a positive step in the interest of increasing awareness about equity in transport policy.

2.4.1 The need for stakeholder involvement

There is the need for integrating various stakeholders in order to have a more inclusive approach in transport policy (Smith, 2001; Mercado et al., 2010). For instance, an accessible walkable environment is both a transport and a health concern. Therefore, links between health and transport institutions should be accentuated.

This means that transport policies should take into consideration a holistic approach of the older persons' necessities that comprise their lifestyles, health, and physical abilities. They should support an integrated mobility approach. If all the laws are in place and the concerned institutions are interrelated, they can contribute to secure consistency in policy directions and trigger accessibility innovations. A comprehensive literature review has recently been finalised as part of one of the milestones of the TEA Cost Action (Bastiaanssen, Lucas & Martens, 2014) that refers to the inclusion of accessibility in equity appraisal. Reference to this work can lead to new ideas on how to evaluate and improve equity in the field of transport.

3 The Case Study of the Bus Service in Malta

The Malta bus service has gone through radical changes in the past three years. Table 1 shows the timeline of the bus service until January 2014.[4] Following nationalisation of the bus service, the Maltese government has issued an expression of interest to find a new operator.

Although the bus service reform failed in achieving modal shift, it led to the improvement of some bus service quality characteristics. Such achievements were acquired through the onerous service level agreement that was included in the contract.

[1] Safe, Accountable, Flexible, Efficient Transportation Equity Act: A Legacy for Users

[2] European Conference of Ministers of Transport

[3] Accessibility for Ontarians Disability Act

[4] This research was carried out in 2014.

Table 1: Timeline of events related to the bus service in Malta.

Month/July	Description
2008	A policy document entitled "Public Transport in Malta: A vision for Public Transport which fulfils public interest in the context of environmental sustainability" (Ministry of Infrastructure Transport and Communications, 2008), paved the way for the bus service reform.
Pre-July 2011	Bus services provided by the Public Transport Association, comprised by 400 bus owners/drivers, operated under the form of a monopoly.
July 2011	Commencement of the bus service reform.
December 2013	"Arriva Malta" bowed out of the country.
January 2014	Bus service nationalised.

3.1 Bus Service Quality Characteristics

An increase in bus patronage means that customers are satisfied (Eboli & Mazzulla, 2007). The bus service quality characteristics that improved after the reform were comfort, fare, and customer care (Attard, 2013). Infrastructural changes included low kerbsides in some areas where there are main bus stops, tactile surfaces, and low floor buses. These improvements were important milestones that made the bus service better, at least for those users who have no other mode of transport available. The factors that need immediate improvement are punctuality related issues (Attard, 2013).

3.2 Effects of the Malta bus service on the elderly

An increase in longevity allows the elderly generation to have a more mobile lifestyle (Alsnih & Hensher, 2003; Banister & Bowling, 2004). In a decade (between 2001 and 2011), Malta witnessed an increase of 19,279 driving licence holders for people aged 60 years and more (National Statistics Office, 2012b).

Yet, the elderly population still represents the highest number of bus users. When compared to other age groups, a minimal distinction between males and females using the bus service is found in older people (Transport Malta, 2010). Since 2011, The Public Transport Customer Satisfaction Survey (Institute for Climate Change and Sustainable Development, 2013) shows that the majority of the frequent elderly bus users are actually non-car owners; hence, as seen in other cases, such as Portland, Oregon, they are potential captive bus users (Beimborn et al., 2003).

Elderly bus users appreciate customer care assistance, fare structure (value for money), and comfort (Institute for Climate Change and Sustainable Development, 2013; Mifsud, 2013). Users also rated positively accessibility in terms of low-floor buses (Mifsud, 2013), which is convenient for elderly persons as seen in cases around Europe and North America (Suen & Mitchell, 2000).

In Malta, negative factors include unreliability, inaccessible and out-dated travel information, lack of safety, fear to travel alone, low frequency of services, and inappropriate bus driver travel behaviour (Institute for Climate Change and Sustainable Development, 2013). Despite the fact that the elderly travel mostly for medical issues and errands, the temporal accessibility to reach Malta's general hospital is still not sufficient, as all the desired time budgets of the elderly are exceeded (Mifsud, 2013).

The time ratio between bus use and car use is significant when considering the locations where elderly people reside and their travel destination. In a study in Madrid, it has been identified that in the congested section of the M40 the travel time ratio of public transport and car is on average 1.62 (Di Ciommo & Lucas, 2014). Thus, the long journeys associated with bus use contribute more to social exclusion, in this case when elderly find it more difficult because of time issues to reach their destinations.

Moreover, elderly persons are well concerned about the inappropriate distribution of bus stops, which does not cater for their needs. Inaccessibility to bus stops is also expressed through difficulties in crossing roads that have high traffic volumes.

4 Maltese Transport Policy

The Structure Plan of the Maltese Islands (Buchanan, 1990) is one of the earliest policy documents that looks holistically at land-use planning policies, including land transport (Buchanan, 1990, Section 14). The policies refer particularly to the land transport matters listed in Table 2.

Table 2: Land Transport Policies referred to in the Structure Plan (Buchanan, 1990)

Land Transport Policies referred to in the Structure Plan (1990)
Development and maintenance of a hierarchical network of roads
Traffic and environmental management
Public transport
Legal and educational measures

Over the past twenty-four years, implementation of these policies was restricted since the aim of this document was to provide a larger planning vision for Malta. Additionally, organisational fragmentation contributes to a disjointed transport policy (Attard, 2005) that is divided between the Planning Authority (PA), Transport Malta (TM) and the Ministry of Transport. Hence, an update of the Structure Plan was long overdue.

The SPED[5] has been issued for public consultation in March 2014 (Malta Environment and Planning Authority, 2014), and is the follow up to the Structure Plan (Buchanan, 1990). The issues related to transport are reported as key issues under the section Travel Patterns (Malta Environment and Planning Authority, 2014, p. 13). They echo the same problems discussed in the original Structure Plan and refer to the white paper that triggered the bus service reform (Ministry of Infrastructure Transport and Communications, 2008).

In fact, the white paper (Ministry of Infrastructure Transport and Communications, 2008) is the only policy document that is directly related to public transport in Malta. Additionally, TM had issued the Accessible Public Transport Infrastructure Policy, Design Guide (Transport Malta, 2009).

The following section discusses these three policy documents in terms of the three different scales of accessibility discussed in Section 1: the macro scale, meso scale and the micro scale. The initial observation when looking at the three policy documents is the absence to the reference of social equity when discussing land transport policy.

4.1 A Methodological Approach: The Three Scales of Accessibility

The structure plan refers to the configuration of the road network as the major criterion that affects the accessibility level (Buchanan, 1990). It focuses on the arterial and distributor road network that forms the main roads in Malta, and link the urban and rural areas.

Apart from this focus, the structure plan mentions the elements listed in Table 2. With the exception of the improvement of public transport, the other elements influence indirectly the bus system since it operates on this road network.

Hence, the policies that reflect these elements affect the bus service and its accessibility. Table 3 shows the policies related to both the macro scale and the meso scale. The macro scale is the shared infrastructure between the public transport system and the road network, as found in the Structure Plan of the Maltese Islands (Buchanan, 1990).

The meso scale refers to infrastructure, both physical and online, that is directly related to the bus system. The physical form is referred to in the Structure Plan (Buchanan, 1990) and in the Accessible Infrastructure for Public Transport Policy Design Guide (Transport Malta, 2009). The cyber form is mentioned briefly in the white paper (Ministry of Infrastructure Transport and Communications, 2008), stating that information technology should be applied at all levels, and give more facilities and information to the public. Although this provides more accessibility, it fails to address equity, and direct access to elderly people. In a world where elderly people are becoming more capable of using technology that aids in increasing accessibility (Mikkonen, Vayrynen, Ikonen & Heikkila, 2002), this concept is even more important to integrate in a transport system that provides services to the elderly. The importance of this is related to the possibility of increasing opportunities and abilities to elderly persons (Geurs & van Wee, 2004).

Table 3 also shows the inclusion of road transport policy that is at the meso scale. All the references made to the meso scale are generic policies that fail to address accessibility for elderly persons. The main barriers related to developing further such transport policies are linked to lack of proper and accurate information; there is a deficiency in transfer of knowledge that is related to a small number of transport professionals in Malta. Another issue arises from the two-party political situation, so politicians do not embrace projects that impose a cost on the population, because the projects may influence whether they are elected in the next legislation. Another issue arises from the lack of infrastructural and professional investment in public transport operations (Attard, 2005).

Micro scale accessibility refers to the ability to move easily when boarding and alighting the bus and on the vehicle itself. The policy documents mentioned in Table 3 do not mention in detail the requirements for an accessible service. However, the service level agreement signed in the contract (Transport Malta, 2009) specifically required low floor buses that are easily accessible by vulnerable groups of society, such as elderly persons.

5 Discussion and Conclusion

This research shows that in Malta land transport policy is limited (Attard, 2005) and public transport policy is even more restrained. Some examples include deficiencies in waiting time conditions and interchanging facilities that increase accessibility to elderly persons. Moreover, policy is fragmented between different institutions within government, namely PA (Buchanan, 1990), TM (Transport Malta, 2009) and the Ministry of Transport (Ministry of Infrastructure Transport and Communications, 2008). This fragmentation leads to a lack of detail in land transport policy that focuses particularly on public transport and on the availability of

[5]Strategic Plan for Environment and Development

Table 3: Policy Documents that address the Macro and Meso Scale of Public Transport in Malta.

	Scales			
	Macro		Meso	
Policy Document	Policy	Comment	Policy	Comment
Structure Plan of the Maltese Islands	**TRA3:** During the time that urban development takes place, developers are subject to fund necessary remedial road works. **RDS7:** The extension of pedestrian priority and access only restrictions in UCAs (Urban Conservation Areas), including areas suffering from the environmental impact of traffic. **TEM1:** Design of traffic management will conform to agreed standards for road design and construction.	There is no specification to accessibility or equity, and such works tend to be temporary, which might imply that provision of necessities that cater for vulnerable groups of society might not be implemented. Elderly people require pedestrian priority, because they have to walk to reach the nearest bus stop. Moreover, UCAs are the core of urban areas, where generally elderly people live. This policy focuses on the benefit of the environment but fails to address the element of equity. The design guide for public transport infrastructure was only prepared nineteen years later. During these years, as still happens with road infrastructure, the designs are known and adapted by road engineers, and knowledge is transferred from one person to the other. This leads to the possibility of omitting equity measures that could improve, amongst other factors, accessibility for the elderly.	**TEM7:** Bus priority lanes and other priority measures in locations where they are feasible, and where the time and cost savings to the bus operators and passengers exceed the equivalent delays to other road traffic. **PTR2:** Appropriate bus fleet for the narrow road types. **PTR3:** Studies that minimise interchange. **PTR5:** Efficient intermodal interchanges. **PTR7:** Smaller bus terminus in Valletta, the Capital City and main hub of the bus service. **PTR8:** Better accessibility during and after operations of major developments. **PTR9:** Improved waiting conditions with reliable passenger information on shelters at bus stops.	All these policies are generic and address accessibility without going into detail of usage for elderly persons, hence omitting the concept of equity particularly for vulnerable groups.
Public Transport in Malta: A Vision for Public Transport which fulfils public interest in the context of environmental sustainability	N/A	N/A	Addressed inter-modal accessibility, new bus fleet, express and regular bus service, a service that caters for peripheral destinations, and night services, discount schemes for frequent users, and provision of information technology services.	These factors were mentioned briefly and there were no policies that referred particularly to each point, and even more so that addressed equity to vulnerable groups.
Accessible Public Transport Infrastructure Policy, Design Guide	N/A	N/A	Details specifications and measurements for the design of bus stop signs and information signs, bus stop and bus shelter designs, including kerb dimensions and bus priority dimensions. It follows the framework of national accessibility guidelines for disabled persons	It primarily focuses on infrastructure design it does not refer directly to usage of the infrastructure by elderly persons.

public transport to elderly people.

In Malta it is necessary to focus more on land transport policy making. There is the need of having an integrated approach to the formulation of land transport policy. Policy should directly address equity and vulnerable groups in society, including elderly persons. This can be done by providing additional policy documents and guidelines to the existing documents. This measure would allow more focus that is direct on equity issues, such as long walking distances to bus stops, which could be identified by using time ratios (Di Ciommo & Lucas, 2014).

However, the SPED (Malta Environment and Planning Authority, 2014) does not seem to address these issues. It builds upon the Structure Plan (Buchanan, 1990) and refers to the public transport policy document (Ministry of Infrastructure Transport and Communications, 2008). The objectives for transport and public transport reproduce the objectives of these two documents, and there is limited direct addressing to accessibility in general and for the elderly.

Meanwhile, TM is in the process of designing the National Transport Strategy and Transport Master Plan. This process is still in its early stages; TM is proposing that an SEA is undertaken as part of the development of the master plan (Transport Malta, 2014). This will allow for the evaluation of policy within the transport framework. Consequently, it is essential that at this stage the relevant stakeholders meet to discuss the needs for improving accessibility to elderly persons and address transport equity issues.

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Note Article

Reviewing the review process: New Frontiers of Peer Review

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Abstract. This news article introduces a new COST Action entitled PEERE (TD1306), which stands for New Frontiers of Peer Review (PEERE). PEERE is a trans-domain proposal which brings together researchers from various different disciplines and science stakeholders for the purpose of reviewing the process of peer review. PEERE officially began in May 2014 and will end in May 2018. Thirty-one countries, including Malta, are currently participating in the Action. In order to set the context in which this COST Action was initiated, we first look very briefly at the history of the process of peer review and various models of peer review currently in use. We then share what this COST Action hopes to achieve.

1 Introduction

As researchers, we are no doubt all too familiar with the feelings of euphoria associated with having a paper accepted for publication in a peer-reviewed journal, especially if the journal in question happens to be the top journal in our academic field, which is likely to be a journal with a high "impact factor"[1]. Sadly, probably even the best among us would also have experienced the sting of having a paper rejected, although these feelings can be somewhat mitigated if we feel that the paper has undergone an impartial peer-review process, and we are provided with good review comments that can help us to improve the paper for submission to the next journal on our list.

Of course, this is from our own viewpoint, as researchers. As researchers, what we sometimes forget to dwell on is the important "gatekeeper" function peer review can play (deciding which information "deserves" to be disseminated). Wrong or misleading information can have a huge impact on the daily life of people, from medical treatments to recovering from the economic crisis. Therefore, the correct functioning of the peer review process is in the interest of science and of society as a whole.

Although the need for some form of peer review (either pre- or post-publication) is acknowledged by most researchers, the system of peer review is far from perfect and there have been numerous high-profile cases of fraudulent publications that have passed the peer review process (Martin, 2012; Storbeck, 2011, July 7).

[1] The "Impact Factor" is probably the most commonly accepted, if controversial, way of rating the quality of academic journals. It is a quantitative tool for ranking, evaluating, categorizing, and comparing journals. It is a measure of the frequency with which the "average article" in a journal has been cited in a particular year or period. The annual Journal Citation Report impact factor is a ratio between citations and recent citable items published. Thus, the impact factor of a journal is calculated by dividing the number of current year citations to the source items published in that journal during the previous two years. An Impact Factor of 1.0 means that, on average, the articles published one or two years ago have been cited one time. An Impact Factor of 2.5 means that, on average, the articles published one or two years ago have been cited two and a half times. http://admin-apps.webofknowledge.com/JCR/help/h_impfact.htm

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Problems are frequently attributed to the social and subjective dimensions of the process (e.g. bias and conflict of interest; Lipworth, Kerridge, Carter & Little, 2011). Other common criticisms levied against the peer review process include unacceptable delays in publication, expense, inconsistencies, fraud/plagiarism, nepotism, and counter to innovation – and the list goes on!

2 What exactly is scientific peer review?

Peer review or refereeing is the process of subjecting an author's work, research, or ideas to the scrutiny of experts (peers) in the same field, traditionally, before the work is published in a journal.

In its most basic form, peer review is the evaluation of an author's manuscript by identified reviewers, who make recommendations to the journal's editor as to whether or not a manuscript should be accepted as is, revised prior to publication or rejected, based on the quality, originality and importance of the manuscript (Sense about Science, 2009). Peer review is one significant method by which research grants are allocated, papers published, academics promoted, and Nobel and other major prizes won (Smith, 2006).

Peer review concerns all of us. As aptly stated by The Publishers Association in response to the UK House of Commons Science and Technology Committee consultation on Peer review in scientific publications conducted in 2010–2012, peer review is "a duty and a skill, performed by researchers, for researchers. It is a system that has been developed by the academic community, for the academic community over centuries and it is established practice that professional scientists are prepared to engage in peer review as a service to the community at large and as a contribution to the progress of science" (The National Archives of the UK, 2011).

The first recorded use of peer review is ascribed to Ishaq bin Ali Al Rahwi (AD 854–931). In his book, Ethics of the Physician, Al Rahwi apparently encouraged doctors to keep contemporaneous notes on their patients, later to be reviewed by a jury of fellow physicians. Journal peer review followed much later, when Henry Oldenburg, editor of Philosophical Transactions of the Royal Society, adopted peer review in the seventeenth century (The National Archives of the UK, 2011). Since then, peer review has played an increasingly important role in scientific publishing: in 2008, 1.3 million learned articles were published in peer-reviewed journals. Peer review is now fundamental to the integration of new research findings into established knowledge, enabling other researchers to analyse or use findings and, in turn, society at large to access and interpret research claims (Sense about Science, 2009).

Several different types of peer review process are now available (see Box 1). It is an evolving process, with continuous attempts being made to find better, more effective models of peer review. The peer review process has also been complicated by the increasing use of institutional repositories, self-archiving, data sharing, social media, and other tools. However, the underlying assumption in all situations is that, since peer review is based on human labour and judgement, it is unlikely that a perfect system can ever be found.

3 Why PEERE?

This COST Action aims to improve the peer review process, potentially increasing the credibility of science in Europe in an era of increasing scandals and public concern. The main objectives of the Action are given in Box 2.

In order to achieve these objectives, three working groups (WG) have been created. They will be working in the following areas:

- WG1: Theory, analysis and models of peer review (Analysing peer review by integrating qualitative and quantitative research and incorporating advanced computational and experimental investigation; Testing implications of different peer review models).
- WG2: Data sharing and testing (Establishing standards and appropriate Information and Communications Technology (ICT) applications to treat, manage and share data on peer review between stakeholders; Providing guidelines and protocols for data sharing; Developing quality and efficiency indicators and monitoring measures to evaluate the potential impact of new models).
- WG3: Research and implementation agenda (Defining and monitoring challenges and prospects for an evidence-based evolution of peer review; Leveraging existing resources and identifying new opportunities for collaboration and research).

4 Opportunities for Malta

As this is a new COST Action, Malta currently has only one representative on the Management Committee. The Action currently includes researchers from diverse disciplines such as computational sociology, economics, basic sciences, etc. Some members have experience as journal editors. Important stakeholders such as the publishers Elsevier, Springer and Wiley, and partners from the US, Canada and Brazil are also included. Whatever your research background, you may have ideas that can help to improve the process of peer review, which is (arguably) the cornerstone of science! If you are interested in joining this Action, please contact Prof. Janet Mifsud (janet.mifsud@um.edu.mt), COST Malta Country National Contact. More information on the Action is

available at: http://www.cost.eu/COST_Actions/tdp/TD1306 and from http://peere.org/.

Box 1: Types of peer review

"Single blind" peer review: The author's name and institution is known to the reviewer, but not vice versa. This is the most common form of peer review, especially in the sciences.

"Double blind" peer review: This system is fully anonymised i.e. the authors are unaware of the identity of the reviewers, and vice versa. This is more common in the social sciences.

"Open" peer review: In which the authors' and reviewers' names are revealed to each other. This is not too common, but is used in some biomedical journals, such as BioMedCentral journals and the British Medical Journal (BMJ).

Post-peer review or post-publication peer review: Different models can be found under this title, for example, Review by formally invited reviewers, after publication of the un-reviewed article; Review by volunteer reviewers, after publication of the un-reviewed article; and Comments on blogs or third party sites, independent of any formal peer review that may have already occurred on the article. Post-publication peer review can be named or anonymous, and reviews can in some cases be written by uninvited reviewers who may not necessarily be literal "peers" in the field (Amsen, 2014).

Cascading peer review (or cascading reviews between linked journals): This is a system whereby a publishing house redirects rejected manuscripts to related journals that have lower rejection rates, in the same field. Advantages to the publisher are reduced cost and higher efficiency, while the advantages to the author is faster publishing (Davis, 2010).

Pre-print servers such as the arXiv repository of electronic preprints (http://arxiv.org/), where the e-prints are commented on by the community, and can later be submitted to a journal and published. Some of the benefits of the arXiv system are that it "allows the scientists to publish research quickly and get informal feedback and identify any weaknesses. This is then followed by formal peer review in a journal" (The National Archives of the UK, 2011).

Box 2: Objectives of PEERE

The main objective of the Action is to improve efficiency, transparency and accountability of peer review through a trans-disciplinary, cross-sectorial collaboration. This is will be achieved through:

- analysing peer review by integrating qualitative and quantitative research and incorporating advanced computational and experimental investigation;
- testing implications of different peer review models (e.g., open vs. anonymous, pre vs. post publication) and different scientific publishing systems (e.g., open vs. subscription based publication systems) for the rigour and quality of peer review;
- discussing present reward structures, rules and measures and exploring new solutions to improve collaboration in all stages of the peer review process; and
- developing a coherent peer review framework (e.g. principles, guidelines, indicators and monitoring activities) for stakeholders that truly represents the complexity of research in various fields.

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Review Article

Role of Protein Structure in Drug Discovery

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Abstract. Many pharmaceuticals currently available were discovered either during the screening of natural of synthetic product libraries or by serendipitous observation. Such a "random" approach entails testing numerous compounds and developing countless high-throughput screening assays. On the other hand, a "rational" approach involves the structure-based route to drug discovery, where the structure of a target protein is determined. Hypothetical ligands may be predicted by molecular modelling, while movement of a molecule may be predicted by Molecular Dynamics Simulations prior to synthetic chemical synthesis of a particular molecule. Here, we will be discussing protein structure-based approaches to drug discovery.

Keywords: Protein Structure, X-ray crystallography, Molecular Dynamics Simulations, Drug design

Introduction

Proteins are complex molecules composed of long strings of twenty different types of amino acid. The length of the string and the order of amino acids are vitally important for the protein to function properly in its biological role. This part of the process of protein function, the gene encoding the protein determines these factors. A single mistake (mutation) in the gene may cause the wrong amino acid to be incorporated into the sequence or a nonsense mutation may cause the protein to be truncated. However, protein function is more directly determined by the protein's three dimensional shape, the protein structure, and the availability of non-protein cofactors.

1 Protein Structure - why is it important?

As can be seen in Fig. 1, a complex protein such as xanthine oxidoreductase (XOR) forms a highly convoluted structure, but one which accommodates cofactors and substrates perfectly. Protein structure is typically determined by one of three methods today; X-Ray crystallography, Nuclear Magnetic Resonance spectroscopy (NMR) or cryoelectron microscopy. The former is the oldest and most commonly used technique, while the latter is only just becoming available for the analysis of proteins at atomic resolution. X-Ray crystallography relies on the ability of the protein to form a regular molecular array and crystallise; a completely biologically unnatural condition for any protein. Even so, it is possible and there are now over one hundred thousand entries in the biological structures databank, RCSB (Deshpande et al., 2005). The advantage of NMR over X-Ray crystallography is that it can be performed in solution (no crystals required) but the major problem is size; NMR cannot be used to determine the structure of large proteins. Electron microscopy will soon be capable of providing structural information about protein as good as X-Ray crystallography, and is performed in solution. Today it can yield protein structures to 2.2 Å (X-Rays typically give resolutions as high as 0.6 to 1.3 Å). In the laboratory of Biochemistry and Protein Science, at the University of Malta, we use X-Ray crystallography to determine protein structure, with crystallisation conditions determined in our laboratory applied and subjected to X-Ray diffraction at the University of Leeds, UK in collaboration with Dr Chi Trinh. We have determined the structures of several superoxide dismutase enzymes and mutants (to a minimum of 1.7 Å) and are currently working to solve the structures of others, in-

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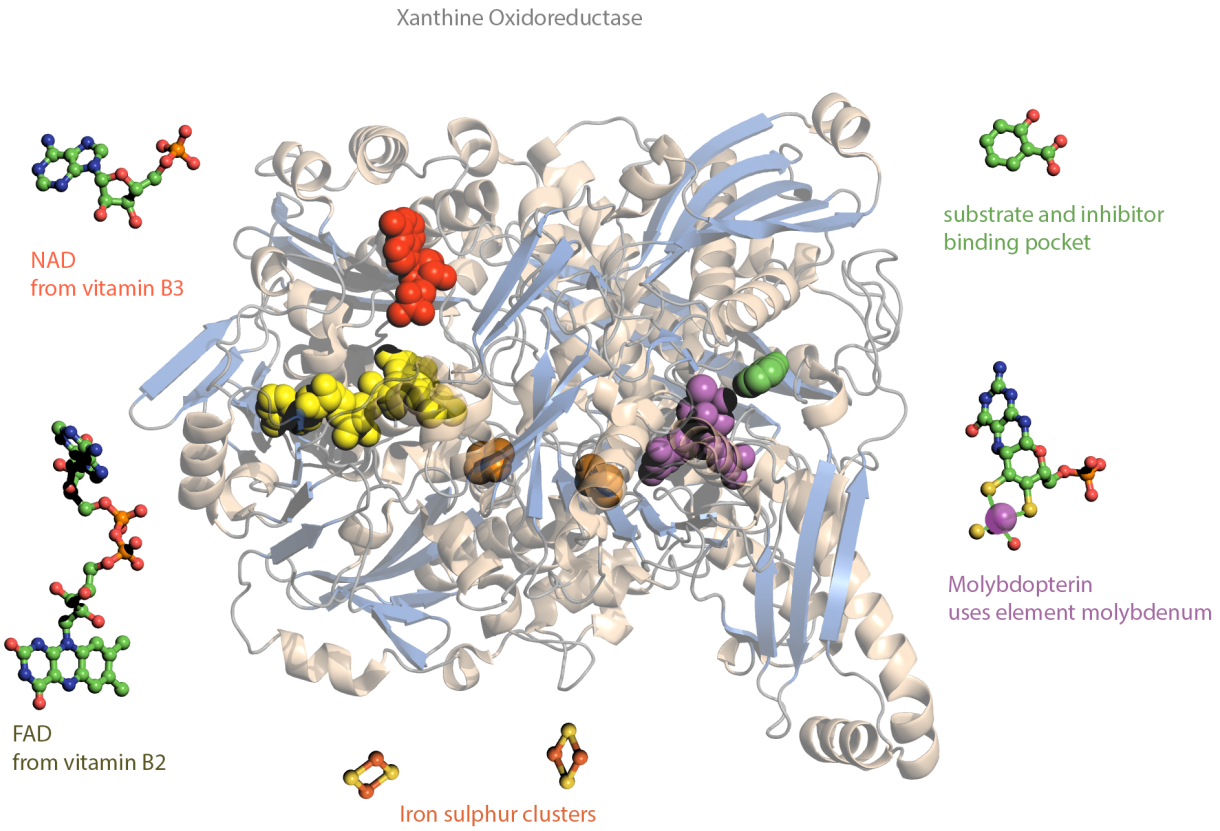

Figure 1: Bovine Xanthine Oxidoreductase. Only one of two identical protein subunits is shown, in cartoon representation with blue beta sheets (arrows) and cream alpha helices (spirals). Cofactors and substrates are shown surrounding the protein in ball-and-stick representation and their corresponding positions of binding within the protein as atomic spheres. NAD is in orange, FAD is in yellow, iron-sulphur clusters are brown, the molybdopterin is purple and xanthine (substrate) in green. Together, the protein forms a scaffold for the cofactors which form an electron transport chain from one side of the protein (substrate) to the other (FAD/NAD). The figure was created using the PyMOL molecular Graphics System (Schrödinger LLC, 2010).

cluding human XOR. Structures such as these help us to understand how the protein functions, and will help to design chemicals to be used pharmaceutically as modifiers of enzyme activity. X-Ray structures usually provide us with a quite static picture of the protein, and it is best combined with other techniques in order to obtain a detailed idea of how the protein functions.

2 Molecular Dynamics Simulations of Biomolecules

Molecular Dynamics Simulations are applied in the investigation of numerous dynamic properties and processes by scientists in a variety of fields that include structural biochemistry, enzymology, biophysics, molecular biology, biotechnology and pharmaceutical chemistry. Molecular Dynamics Simulations allow the researcher to study the thermodynamic and time-dependent (kinetic) properties of biomolecules such as proteins. This provides an understanding of numerous dynamic aspects of biomolecular structure, recognition, and function (Adcock & McCammon, 2006) The techniques involving Molecular Dynamics Simulations involve Langevin's or Newton's equations of motion, as well as a particular molecular bond structure, parametrized force fields, and an initial conformation of atomic positions, together with the velocities that are necessary to generate the atomic dynamics in a molecular system. Molecular Dynamics Simulations have a limited function when used in isolation. The trajectory of Molecular Dynamics (i.e., the progress of a simulated structure correlated to time) usually generates data related only to the level of atomic positions, velocities and single-point energies. Researchers are usually interested in obtaining macroscopic properties. The latter requires the application of statistical mechanics, which combines microscopic simulations together with macroscopic observables. Statistical mechanics provide the mathematical expressions associating the distributions

and motions of atoms to macroscopic observables including free energy, pressure and heat capacity (Callen, 1985; McQuarrie, Salvaterra, De Blas, Routes & Mahler, 1976). Molecular Dynamics Simulation programs include AMBER, CHARMM, NAMD and POLY-MD.

Kinetic rate constants of ligand-receptor interactions are essential in enzymology (Bar-Even et al., 2011) and drug discovery (Copeland, Pompliano & Meek, 2006), as they provide a good indication of drug efficacy (Copeland et al., 2006). Thus, the prediction and optimisation of these parameters is an important challenge in medicinal chemistry (Copeland, 2016). Even though these values may be measured experimentally, an accurate computational prediction would result in a useful alternative in cases where the experiment is either expensive or difficult to perform. Additionally, advances in computational power, have allowed simulations to be carried out in significantly less time. This provides a great potential for methods that require vast amounts of computational power.

Predicting the interaction between an enzyme and its substrate and other ligands via Molecular Dynamics Simulations is essential to fully understand the mechanism of the enzyme. Predicting hydrogen bonding in an enzyme is crucial for analysing the structure and function of this type of biological molecule, especially in terms of enzyme catalysis. Molecular Dynamics Simulations provide information on the molecule that is not observable in the data obtained via X-ray crystallography experiments alone. With this knowledge it is then possible to design new chemicals based upon the binding requirements discovered to inhibit or enhance the biological activity of the protein. In many cases it may be adventitious to modify the structure of an existing, known effector molecule (enhancer or inhibitor) to increase or decrease its activity. With computer aided rational design, a new or modified pharmaceutical may be created with better effectiveness and reduced side effects. Molecular simulations give us the power to suggest or reject such modifications prior to chemical synthesis of the compound. This saves time, effort and money.

3 Protein Structure, Molecular Dynamics, Drug Discovery - tying the knot with computational approaches

Structure-based virtual screening is a computational method employed to find small, bioactive molecules which sterically fit and interact with a protein. A library of small molecules (ligands) is "docked" to the protein's binding site in a typical "lock-and-key" fashion (Meng, Zhang, Mezei & Cui, 2011). Three things are required for this computational approach. Firstly, a protein structure is either determined experimentally (as described earlier) or modelled computationally, typically using homology modelling. In homology modelling, we use one or more known protein structures with close sequence similarity as a template to model our protein of interest. The binding site on the protein needs to be identified. Secondly, a library of small molecules must be prepared and provided to the docking algorithm. This preparation may imply many steps such as sanitisation, setting the appropriate ionization state, removing salts, etc. Thousands to millions of molecules form part of the digital library, only a fraction of which could possibly be tested physically in a laboratory. Thirdly, a docking protocol is required which defines the parameters used in the docking experiment. This includes, but is not limited to, ligand flexibility, protein side-chain flexibility, role of water molecule in the binding site, and which scoring function to use. The scoring function is of critical importance as it assesses the goodness of the fit, producing a quantitative score which can be used to rank each individual ligand. Many aspects are taken into consideration when evaluating the interaction of the protein with each ligand including steric fit, electrostatics, polar interactions and hydrogen bonding. The problem is compounded by the many possible conformations the ligand (or protein) takes on. The scoring function must evaluate each of these binding poses. Some of the major critiques of docking are the inability to calculate the free binding energy correctly (possibly because of the additive nature of most scoring functions), protein main-chain flexibility, the correct prediction of water in binding and the intensive computational resources required. In order to alleviate some of these issues, docking is sometimes used as a filtering first step before a more rigorous and computationally intensive molecular dynamics simulation. The top hits of the docking experiment are then rescored using MD. In a typical workflow, large virtual screening databases are first filtered using fast and inexpensive docking protocols. This rescoring is based on more physically realistic techniques for binding free energy estimations such as thermodynamic integration, free energy perturbation, linear interaction energy and molecular mechanics/Poisson-Boltzmann and surface area (MM/PB-SA). Overall, this provides a more accurate prediction of the binding affinity between the protein and the ligand (compared to the scoring function in docking tools). Computer-aided drug design is an active field of research, which has gained a lot of momentum in recent years - mostly driven by the decreasing productivity of the pharmaceutical industry to find new drugs.

4 Limitations of MD-based methods

The main force fields that are currently being employed for biomolecular simulations include AMBER (Asensio & Jimenez-Barbero, 1995), CHARMM (MacKerell et

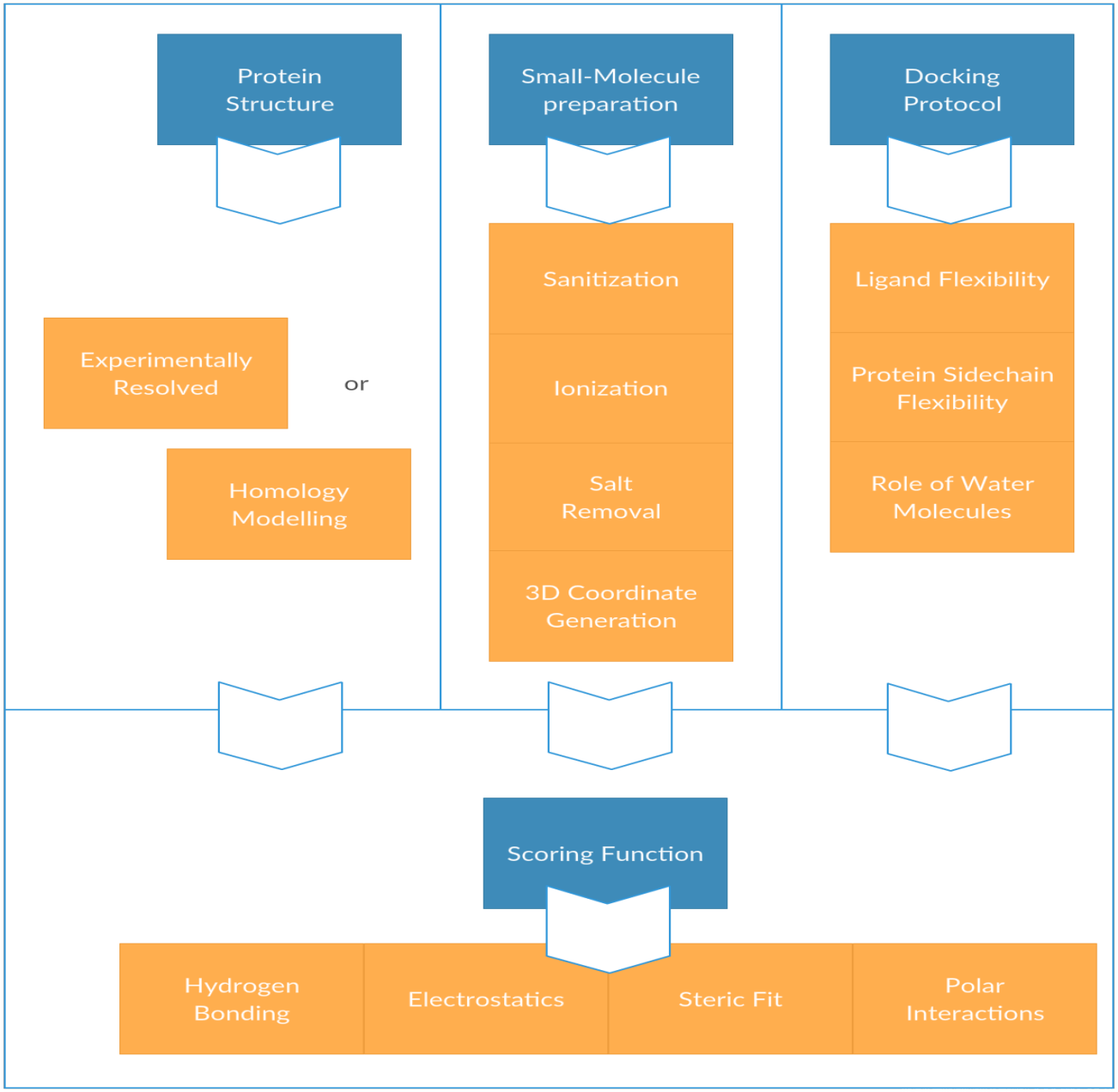

Figure 2: Components of a computational protein-ligand docking experiment. The goodness-of-fit of different small-molecules in a protein's pocket is assessed by means of a scoring function. The top-ranked results may serve as input to more computationally exhaustive techniques, such as Molecular Dynamics.

al., 1998), and OPLS (Jorgensen & Tirado-Rives, 1988). Although, extended parametrisation for amino acids, nucleic acids, lipids, carbohydrates, and several ionic species has been included in the parent force fields in recent years, the variability of small molecules (i.e., ligands) still poses a challenge to condensed-phase force fields. Thus, the user must carry out specific parametrisation. The latter is a time-consuming and an error-prone procedure, and has lead to the development of some general force field sets such as GAFF57 for AMBER, and CGenFF58 for CHARMM, together with specific parametrisation toolkits. Several challenges must be overcome to further increase the importance of MD-based methods on drug design. The molecular mechanics force fields that are presently available partially or fully neglect charge transfer and polarisation effects, as well as many electronic-based interactions. The current limits of force field and MD-based methods allow certain

target families, such as metalloproteins, to be studied with limited accuracy (De Vivo, Masetti, Bottegoni & Cavalli, 2016).

Conclusion

It is the combination of computational approaches that encompass techniques such as molecular dynamics simulations and docking, together with the interpretation of related experimental structural data, which is essential to provide a comprehensive understanding of the motions in proteins and their assemblies. Information on the latter is crucial when synthesising improved biomolecules and designing new drugs.

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Meeting Report

Neuropathology and Neuropharmacology of Monoaminergic Systems

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Abstract. The third EU COST Action CM1103 "Structure-based drug design for diagnosis and treatment of neurological diseases: dissecting and modulating complex function in the monoaminergic systems of the brain" Annual Conference entitled "Neuropathology and Neuropharmacology of Monoaminergic Systems" was hosted by the University of Bordeaux, France on 8-10 October 2014. The conference, organized by Prof. De Deurwaerdére, was supported by COST (European Cooperation in Science and Technology) and LABEX (LABEX Brain, University of Bordeaux). The program took the form of a three-day meeting, comprising a series of French and international invited talks and breakout sessions designed to identify key gaps in current knowledge and potential future research questions. The aims of this Conference were two-fold: 1. To identify the current state-of-the-art in the understanding of the pathological mechanisms that contribute to different neuropsychiatric disorders, and to what extent, monoamines a multi-target drugs and/or other interventions might prevent these changes. 2. To identify specific areas of research where information is sparse but which are likely to yield data that will impact on future strategies to treat neurodegenerative disorders.

Meeting report

COST Action CM1103 (http://www.cost.eu/domains_actions/cmst/Actions/CM1103) *Structure-Based Drug Design For Diagnosis And Treatment of Neurological Diseases: Dissecting and Modulating Complex Function in the Monoaminergic Systems of the Brain* was established to stimulate an interdisciplinary approach to the task of understanding the molecular basis of neurological and psychiatric disorders. Medicinal chemistry now has many computational tools to aid in the design of novel drugs targeting either one or several proteins. Biological insights are essential to evaluate the efficacy of these novel drugs and to propose also new targets and approaches. Commonalities amongst the brain monoamine neurotransmitters, dopamine (DA), noradrenaline (NA), adrenaline (A) serotonin (5-HT) or histamine, are evident as these systems of neurotransmission are all involved in the pathophysiology of all major neuropsychiatric disorders and brain affections, such as mood disorders, schizophrenia, autism-spectrum disorders, Parkinson's disease (PD), Alzheimer disease, epilepsy, ischemia and dementias. Indeed, the efficacy of numerous medicines against the above-mentioned pathologies has been related at least in part to an action of these chemical drugs on the monoaminergic systems. The collaborative work in the action is a permanent interaction between bottom-up and top-down analyses towards understanding the neuropathology and neuropharmacology of monoaminergic systems. The extended abstracts in these Proceedings are a selection from the contributions to the third annual meeting, held in Bordeaux, of groups actively working towards these goals.

Chemical developments come from existing molecules and their modification either via classical or new ways of synthesis, and computational modeling. Reported at this conference are compounds for single targets such as acetylcholinesterase or aldo-ketoreductase by Magdalena Majekova (Slovak Academy of Sciences, Bratislava), or families of compounds such as benzothiazoles by Kamil Musilek (Kadir Has University, Istanbul) or quinolines by José Marco-Contelles (Consejo Superior de Investigaciones Científicas, Madrid) synthesized and studied with the goal of treating Alzheimer disease, Parkinson's disease or stroke by Mercedes Unzeta (Universitat Autonoma de Barcelona). A series of multi-target compounds has been also designed with action on specific pairs (or multiples) of targets. Ligands care-

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fully designed to bind to multiple targets are another accepted strategy in tackling the complex neurodegenerative diseases Cavalli et al., 2008; Geldenhuys and der Schyf, 2013 but producing the best combinations to explore phenotypic results is not a trivial task Prati, Uliassi and Bolognesi, 2014. With large numbers of compounds tested and published in databases, one starting point comes from data-mining existing knowledge. Theoretical prediction of pharmaceutical targets for new compounds is possible using a probabilistic method to build a model of any compound from the ChEMBL dataset. Then, using the circular fingerprint descriptors, a cheminformatic method, developed to explore known off-target interactions of known compounds, was applied to identify which of the new compounds that should bind to the desired targets. This and confirmation of the predicted efficacies are included in this topic.

Reuptake transporters are targets to modulate amine levels in the synaptic cleft, but, unfortunately, crystal structures are not available yet. Nonetheless, homology models of the transporters in conjunction with mutational studies are beginning to define the molecular determinants of binding to these proteins. Structure-based drug design together with good pharmacological data provides the basis for designs combining the features needed for each target into one molecule. These molecular determinants predict quite nicely the behaviour of some compounds (amphetamine, tyramine, cocaine, DA, 5-HT; Yelekci and Connaly, this conference) toward the dopamine transporter in models *in vitro* or *in* vivo Navailles and De Deurwaerdère, 2011.

Drug design and computational studies are rendered easier by the crystal structures that identify the interaction of the protein with a ligand. Structures for the enzymes that degrade the monoamine neurotransmitters (MAO and COMT) are available for computational exploration of molecular determinants of binding. Inhibition of monoamine oxidases (MAO A and MAO B) by several irreversible inhibitors and a few new, well-tolerated, reversible inhibitors used over the last 30 years, results in increased levels of brain amines Youdim and Bakhle, 2006. Docking and molecular dynamic studies of the inhibitor in the active site are now standard tools for medicinal chemists aiming to improve inhibitor binding or decide which part of a molecule may be changed without loss of affinity Samadi et al., 2012. Using structure-based techniques, complete theoretical searches for new lead compounds are also possible Vilar, Ferino, Quezada, Santana and Friedman, 2012. Finally, using these models, it is possible to address the selectivity of series of new compounds toward MAO. More recently, the crystal structure of glutamatergic AMPA receptors has been obtained, leading to conceive new series of compounds targeting the GluR1 subunit of the AMPA receptor hopefully as efficiently as antinociceptive compounds as described by Stefania Butini (University of Siena).

Pharmacological evaluation is still a necessary step to determine the accurate efficacy of compounds and to evaluate the selectivity towards other targets. Several MAOI are not selective for MAO and display good affinities for other targets such as lysine-specific demethylases or cytochrome P450 Binda et al., 2010; Massimo Valoti (University of Siena); Thomas Malcomson (University of St Andrews). Lysine-specific demethylases are involved in epigenetic, cytochrome oxidase are involved in the metabolism of xenobiotics. These other targets together may participate in the behavioural effects of these compounds in animal models and in humans. Indeed, as detailed by Marco Bortolato (University of Kansas) gene deletion of MAOA gives surprising results compared to pharmacological compounds Finberg, 2014. Apart from the longitudinal and developmental dimensions inherent to gene deletion, Keith Tipton (Trinity College, Dublin) showed that the differences suggest that the biological effects of MAOI do not only result from their interaction with MAO Finberg, 2014.

Neuropharmacological explorations of the mechanism of action of current drugs are a big challenge in neurobiology in order to identify the targets, ameliorate the phenotype, and limit the side effects. Even if these drugs are currently used in clinic, their mechanism of action is often misunderstood, not only for MAOI. L-DOPA is the gold standard medication in Parkinson's disease but the numerous motor and non-motor side effects occurring after years of treatment lead to conceive other therapeutic strategies and to focus on its mechanism of action Meissner et al., 2011. Numerous strategies are developed to find new chemical compounds able to stimulate the DA D2 and D3 receptors, the primary mechanism thought to underline the efficacy of L-DOPA. The chemical development of new compounds unmasks new pharmacological concepts and the development of compounds will integrate these new concepts such as biased signaling or heterodimerization of receptors, as detailed by Holger Stark (Heinrich Heine University). Abdelhamid Benazzouz (Université de Bordeaux) emphasized the importance of the deep brain stimulation that also permits to highlight unpredicted targets such as D5 receptors, 5-HT_{2C} receptors in the control of DA neurons De Deurwaerdère, Lagière, Bosc and Navailles, 2013 or to highlight the involvement of 5-HT and NA neurons in the control of dopamine-mediated function Di Matteo, Di Giovanni, Pierucci and Esposito, 2008; Navailles, Di Giovanni and De Deurwaerdère, 2014; Navailles, Milan et al., 2014. Finally, Philippe De Deurwaerdère (Université de Bordeaux) presented evidence that the

Figure 1: Some members of EU COST Action CM1103 "Structure-based drug design for diagnosis and treatment of neurological diseases: dissecting and modulating complex function in the monoaminergic systems of the brain" at the Annual Conference "Neuropathology and Neuropharmacology of Monoaminergic Systems" hosted by the University of Bordeaux, France on 8–10 October 2014.

chronic use of L-DOPA could affect the activity of MAO enzymes in vivo showing that the different topics developed in the action are interconnected.

Understanding the interactions of neurotransmission systems is an important step for the optimization of therapeutic strategies. The plurality of targets that potentially bind antipsychotics or antiparkinsonian, antidepressant drugs favours the need to develop multitarget compounds. Because neurobiological systems of neurotransmitters establish close relationships Chesselet, 1984, it is likely that a pharmacological action toward one system will more or less directly affect the other one. This is highlighted perhaps by famous associations and links between some neurotransmitter systems such as the 5-HT/DA interaction Di Giovanni, Di Matteo, Pierucci and Esposito, 2008; Di Matteo et al., 2008, the glutamate/DA interaction M. Carlsson and Carlsson, 1990, the 5-HT/GABA interaction Soubrié, 2010 and so one. The role of monoamines still remains unresolved even in pathologies such as depression or anxiety where the connection between monoamines and the diseases has been known for years. In epilepsy, although monoamines were thought to control the excitability of hippocampal cells via a lowering influence on depolarizing current, Giuseppe Di Giovanni (University of Malta and Cardiff University), found that 5-HT_{2C} receptors do not modify the electrophysiological responses in the model of maximal dentate activation (MDA) of temporal lobe epilepsy (TLE) while they undergo to a cellular redistribution in the hippocampus of epileptic rats Orban et al., 2014. Di Giovanni also showed that in the pilocarpine-model of TLE, the activation of the 5-HT_{2C} receptors surprisingly induce a powerful anti-epileptic effect, probably mediated by interacting with GABA and glutamate. These findings highlight a strong model-dependence of the 5-HT_{2C} receptor effects which is especially true for TLE epilepsy.

Moreover, the physiopathology of numerous pathologies is still misunderstood. Nela Pivac (Rudjer Boskovic Institute, Zagreb) presented some data concerning the post-traumatic stress disorder, a pathology that probably involves monoamines, but devoid of efficient treatment.

A better understanding of the relationships of chemical systems in the brain relies on the availability of good chemical compounds for research and diagnosis and good models to address the efficacy of compounds and the physiopathology of brain diseases. These models are sometimes classical as those that have been developed in rodents to study numerous neuropsychiatric diseases or less classical as the use of crayfish to study the neurobiological bases of anxiety as underlined by Pascal Fossat (Université de Bordeaux) Fossat, Bacqué-Cazenave, De Deurwaerdère, Delbecque and Cattaert, 2014.

Acknowledgements

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Conference proceedings

INTERDISCIPLINARY CHEMICAL APPROACHES FOR NEUROPATHOLOGY CM1103

"4th Neuroscience Day University of Malta"

22nd Tuesday	Speaker	Topic // Title
14.00-14.30	UoM - **Richard Muscat** MT COST - **Janet Mifsud** UoM-St Andrews, UK- **Rona Ramsay** UoM - **Giuseppe Di Giovanni**	Welcome // Introduction
Session 1	**Chair: Rona Ramsay**	
14.30-15.00	Di Giovanni Giuseppe (MT, WG4)	GPCRs modulation of extrasynaptic GABAARs
15.00-15.30	Peter Gmeiner (DE, guest speaker)	GPCR ligands probing structure and controlling function
15.30-16.00	Tipton Keith (CR, WG4)	From magic bullet to scatter-gun; is there a viable alternative?
16.00-16.30	Mavri Janez (CR, WG4)	How Enzymes Work? QM/MM Simulation of MAO
Session 2	**Chair: Mück-šeler Dorotea**	
17.00-17.30	Crespi Francesco (IT, guest speaker)	Non-invasive analysis of brain penetration of chemicals: concomitant Near-Infrared Spectroscopy [NIRS] and pharmacokinetic - pharmacodynamic [PK/PD] study
17.30-18.00	Magdalena Majekova (SK, WG4)	Bioactivity parameters of indole - type compounds and their possible relevance to treatment of neurological diseases

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23rd Wednesday	Speaker	Topic // Title
Session 3	**Chairs: Giuseppe Di Giovanni/Richard Muscat**	4th Neuroscience @ University of Malta
9.00-9.30	Valentino Mario (MT, No COST)	Two-photon imaging of cortical microvessels and astrocytic interactions in live mouse brain
9.30-10.00	Ruben Cauchi (MT, No COST)	Modelling Spinal Muscular Atrophy in Drosophila: a Fruitful Approach?
10.00-10.30	Neville Vassallo (MT, No COST)	Lipid Membranes - a new target for neurodegeneration
10.30-11.00	Zammit Christian (MT, No COST)	Immature axons: a new therapeutic target for neonatal white matter ischaemia?
Session 4	**Chair: Marco-Contelles José**	
11.30-12.00	Simic Goran (HR, WG4)	The necessity of reliable biomarkers for monitoring potential treatments in Alzheimer's disease
12.00-12.30	Roberto Di Maio (USA, NoCOST)	Muscarinic stimulation elicits abnormal GABA-ergic differentiation in Mouse-derived stem cells
12.30-13.00	Philippe De Deurwaerdére (FR, WG3)	5-HT2C receptors: a G-protein coupled receptor involved in opposite and distributed controls in basal ganglia
13.00-13.30	Mauro Pessia, (IT, guest speaker)	Potassium channels as target of CNS disorders
Session 5	**Chair: Di Giovanni/Muscat**	4th Neuroscience @ University of Malta
14.30-14.50	Massimo Pierucci (MT, No COST)	Nicotine Addiction and Lateral Habenula
14.50-15.10	Gabriella Andrina Mifsud(MT, No COST)	Oligodendrocyte pathophysiology and treatment strategies in ischemia
15.10-15.30	Stephanie Ghio and Michelle Briffa. (MT, No COST)	Amyloid neurodegeneration: from electrophysiology to flies
15.50-16.10	Frau Robert (IT, WG4)	Positive allosteric modulation of GABA-B receptors: a novel therapeutic approach for schizophrenia
16.10-16.30	Esteban Gerard (ES, WG1)	'Effect of new MTDL hybrids based on donepezil, pyridyl and indolyl moieties on Monoaminergic and Cholinergic systems: An HPLC metabolic approach'.

24th Thursday	Speaker	Topic // Title
Session 6	**Chair: Valoti Massimo**	
9.00-9.30	Musilek Kamil (CZ WG2)	Design, synthesis and evaluation of modulators counteracting ABAD Aβinteraction
9.30-10.00	Unzeta Mercedes (ES WG3)	In vivo and in vitro biological assessment of ASS234, a novel Donepezil-indolpropargylamine, as a multifunctional molecule with a potential therapeutic profile for Alzheimer's disease
10.00-10.30	Mück-šeler Dorotea (HR WG4)	Serotonergic receptors, the new targets in the treatment of Alzheimer's disease

10.30-11.00	Marco-Contelles José (ES WG2)	The Revisited MAO Inhibition by N-(Furan-2-ylmethyl)-N-prop-2-yn-1-amine Derivatives as Potential Drugs for the Treatment of Alzheimer's Disease
Session 7	**Chair: Dr Maria Carreiras**	
11.30-12.00	Najat Aourz (BE, WG3)	Sst2 and sst3 - but not GHS-R1a- receptors are involved in the anticonvulsant effects of cortistatin-14
12.00-12.30	Stark Holger (DE, WG1, 2)	Bioisosteric Replacement in Dopamine D2-like Receptor Agonists
12.30-13.00	Carreiras Maria (PT, WG1,2)	Synthesis, pharmacological assessment, and molecular modeling of AChE/BuChE inhibitors: effect against amyloid-β
13.00-13.30	Marcello Leopoldo (IT, guest speaker)	Recent Advances in the Study of 5HT7 Receptor Pharmacology: Focus on the Selective Agonist LP-211
Session 8	**Chair: K. Tipton**	
14.30-15.00	Ponimaskin Evgeni (DE, No COST)	Interplay between serotonin receptors 5-HT1A and 5-HT7 in regulation of receptor functions in the brain
15.00-15.30	Nikolic Katarina (RS WG1)	Pharmacophore Modeling of Novel Non-imidazole Histamine H3 Receptor Ligands with Inhibitory Histamine N-Methyltransferase Activity
15.30-16.00	Valoti Massimo (IT, WG3)	CYP-dependent metabolism and vascular effects of ASS234, a novel multitarget-directed ligand
Session 10	**Chair: Simic Goran**	
16.30-17.00	Vianello Robert (HR WG1)	Recent progress in understanding the catalytic activity of monoamine oxidases
17.00-17.30	Yelekci Kemal (TR WG1)	In silico design of novel and selective neuronal nitric oxide synthase (nNOS) inhibitors
17.30-18.00	Butini Stefania (IT, Guest Speaker)	Novel Tools for Disease Modifyng anti-Alzheimer's Drugs: hChEs and b-Amyloid Aggregation Inhibitors

ABSTRACTS

P1.1
MODULATION OF EXTRASYNAPITIC GABAA RECEPTORS BY G-PROTEIN-COUPLED RECEPTORS

Giuseppe Di Giovanni[1,2] **Adam Errington**[1], **Vincenzo Crunelli**[1]
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$GABA_A$ receptors ($GABA_ARs$), the main inhibitory neurotransmitter-gated ion channels in the central nervous system, are finely tuned by other neurotransmitters and endogenous ligands. The regulation of synaptic $GABA_ARs$ ($sGABA_ARs$) by G protein-coupled receptors (GPCRs) has been well characterized and is known to occur either through the conventional activation of second-messenger signalling cascades by G proteins or directly by protein-protein coupling. In contrast, research on the modulation of extrasynaptic $GABA_AR$ (eGABAARs) is still in its infancy and it remains to be determined whether both of the above mechanisms are capable of controlling $eGABA_AR$ function. In this talk, I will summarize the available literature on $eGABA_AR$ modulation by GPCRs, including $GABA_B$, dopamine (DA) and serotonin (5-HT) 2A/2C ($5–HT_{2A/2C}$). Although at present these GPCRs−$eGABA_ARs$ cross-talks have been investigated in a limited number of brain areas (i.e. thalamus, cerebellum, hippocampus, striatum), it is already evident that $eGABA_ARs$ show nucleus and neuronal type-selective regulation by $GPCR_s$ that differs from that of $sGABA_ARs$. This distinct regulation of $eGABA_ARs$ versus $sGABA_ARs$ by GPCRs provides mechanisms for receptor adaptation in response to a variety of physiological stimuli and under different pathophysiological conditions. Further research will advance our understanding of $eGABA_ARs$ and GPCR signalling and offer novel targets for the treatment of many neurological and neuropsychiatric disorders where abnormalities in $eGABA_ARs$ have been suggested to exist.

KEY WORDS: Absence epilepsy, metabotropic receptors, monoamines, phosphorylation, tonic $GABA_A$ inhibition.

P1.2
GPCR LIGANDS PROBING STRUCTURE AND CONTROLLING FUNCTION

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GPCRs constitute a large superfamily of target proteins (nearly 800 different human genes encode for GPCRs) and each of them can adopt functionally distinct conformations. The first X-ray crystal structures of druggable GPCRs in complex with ligands provide a basis for the investigation of molecular determinants responsible for affinity and selectivity of ligands. Moreover, the structures of different activity states of GPCRs allow us to identify molecular interactions discriminating between inverse agonists, antagonists and agonists. These fundamental results also contribute to the rational discovery of drugs selectively binding to particular conformational states. Thus, there is growing evidence that homo- and heterodimers effect and diversify G-protein coupling. Besides this, the concept of functional selectivity (biased signaling) owing to ligand-specific GPCR conformations has been corroborated. Although GPCR-binding drugs could be evolved for a number of target GPCRs, the rational development of drugs with beneficial selectivity patterns between structurally related GPCRs and functionally relevant GPCR conformations, controlling intrinsic activity profiles, requires a better understanding for GPCR ligand interactions. We have developed GPCR ligands as molecular probes for structural investigations and structure-function relationship studies. Probing the molecular determinants of GPCR function, we designed functionally selective dopamine D2 receptor agonists that are able to differentiate between the activation of two relevant G-proteins, G_o and G_i.

KEY WORDS: GPCR, molecular probe, functional selectivity.

P1.3
FROM MAGIC BULLET TO SCATTERGUN: IS THERE A VIABLE ALTERNATIVE?

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It is over 100 years since the Ehrlich concept of the magic bullet, but, leaving aside monoclonal antibodies,

very few drugs have achieved the selectivity and specificity that he had hoped for. In many cases multi-targeted drugs have proven advantageous. However, the complexities of drug actions and interactions in the tissues make it difficult to envisage likely responses without time-consuming experimentation and testing. Systems biological approaches may help to shorten the time and expense of drug development and assessment. The approach described here involves deconstruction of the putative drug molecule into component structures that can then be used to predict its metabolic fate in the tissues and the metabolic products that might influence its actions. Extensions also allow the possibility of predicting receptor interactions and groups on the molecule that may impede such interactions, which may then assist rational drug design. Finally, *in silico* approaches to investigate tissue and species differences in the metabolism of drugs will be outlined.

KEY WORDS: *in silico* drug development, drug metabolism, systems biology.

P1.4
HOW ENZYMES WORK? QM/MM STIMULATION OF MAO

Janez Mavri[1,2], Matej Repič[1], Rok Borštnar[1], Miha Purg[1], Fernanda Duarte[3], S.C. Lynn Kamerlin[3] and Robert Vianello[4]
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Understanding of biological processes at the molecular level is one of the greatest challenges in biomedical research and the key to understanding how biomolecules, biomolecular systems, cells, and ultimately, living organisms function. Molecular dynamics simulations of hydrated enzymes provide rate constants for enzymatic reactions.

In this talk I will give an overview of this simulation of hydrated enzymes. The choice of initial state, effectively polarized vs. polarizable force fields, proper treatment of long-range electrostatics, protonation states of ionizable residues and associated pKa values, inclusion of explicit water molecules and necessity for hierarchical treatment of enzymes will be discussed. We will touch the ideas behind treatment of chemically reactive systems using QM/MM approach and quantization of the nuclear motion allowing for treatment of tunneling. As a case study I will use monoamine oxidase B (MAO B), an enzyme that catalytically decomposes dopamine and to a lesser extent serotonin. For this enzyme we suggested the mechanism that is consistent with all available experimental data and we performed a series of biomolecular simulations.

KEY WORDS: Biomolecular simulation, hydrated enzymes, electrostatics, QM/MM, MAO B, enzyme, dopamine, serotonin.

P1.5
NON INVASIVE ANALYSIS OF BRAIN PENETRATION OF CHEMICALS: CONCOMITANT NEAR-INFRARED SPECTROSCOPY [NIRS] AND PHARMACODYNAMIC [PK/PD] STUDY

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Near-infrared spectroscopy (NIRS) selectively monitors non-invasively the absorption spectra of the oxygenation - deoxygenation states of haemoglobin (HbO_2/Hb, respectively). These measurements and the total haemoglobin concentration (HbO_2 + Hb) considered as total blood volume are indicative of the state of vascular activity, the level of oxygen saturation, and therefore the state of the metabolism in the living tissue. This study proposes that changes in brain metabolism measured by NIRS are a useful index of brain penetration and therefore brain activity of chemical entities. Compounds from different chemical classes were selected on the basis of their known brain penetration and pharmacokinetic profile. In particular, two NK1-SSRI receptor antagonists (GSK135... and GSK189...) having similar molecular characteristics and two glycine-1 transporter inhibitors (GSK270... and GSK267...) were chosen based on in vitro high or low rat brain penetration (B/B) ratio, respectively. It appears that treatment with GSK135 (B/B ratio: 2.70:1) modifies the NIRS parameters while GSK189 (B/B ratio: 0.22:1) does not significantly alter HbO_2 - Hb levels when comparing to vehicle treated rats. Similar results are obtained using GSK270 or GSK267 (brain concentration 1hr post treatment: 388 or 13ng/g, respectively).

These results indicate a direct relationship between brain penetration (and possibly efficacy) of drugs and brain metabolism. Thus, they support that *in vivo* non-invasive NIRS contributes to assess brain penetration of chemicals, i.e. significant changes in

NIRS parameters could be related to brain exposure, or vice versa the lack of significant changes in NIRS HbO_2/Hb could be indicative of low brain exposure and indeed low efficacy.

KEY WORDS: *in vivo* non-invasive NIRS, HbO2/Hb, rat brain, blood brain barrier.

P1.6
BIOACTIVITY PARAMETERS OF INDOLE-TYPE COMPOUNDS AND THEIR RELEVANCE TO TREATMENT OF NEUROLOGICAL DISEASES

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Compounds with indolic moiety are known for their manifold potential in biological activities. In the perspective of an intervention against neurological diseases several activities are coming into focus, as the ability to prevent oxidation stress, the preservation of the monoamine neurotransmitter signal (e.g. by the inhibition of MAO enzymes), the anti-inflammation properties, etc. The summary of our recent knowledge in this field is the goal of the presentation.

The hexahydropyridoindoles derived from their parent structure stobadine ((-)-cis-2,8-dimethyl-2,3,4,4a,5,9b-hexahydro-1H-pyrido[4,3-b]indole) exhibited neuroprotective properties manifested in hypoxia-reoxygenation treated brain tissues and slices (in vitro) and ischemia/reperfusion brain (in vivo). From the compounds studied, the derivatives with R8 methoxy substitution exceeded others in the antioxidant and neuroprotective activities. Studies in vivo established anxiolytic effect for the methoxy derivative SMe1EC2 (±)-8-methoxy-1,3,4,4a,5,9b-hexahydro-pyrido[4,3-b]indole-2-carboxylic acid ethyl ester. The derivative SMe1EC2 was found to protect the hippocampus of rats exposed to trimethyltin (a model of Alzheimer-like neurodegenerative disorder) from cell death and damage. For further study, the elaborated model of MAO-B inhibition based on 2v5z complex with safinamide with YAMBER3 force field was used. The key interactions for methoxy substituted derivatives were determined.

The derivatives of 1-indole acetic acid were found to be efficient inhibitors of aldo-keto reductases (AKR). The role of the AKR enzymes in the development of neurodegenerative disorders and a possible intervention via AKR inhibition are brought for discussion.

KEY WORDS: Indole-type compounds, MAO-B inhibition, molecular modeling, trimethyltin, neuroprotection.

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P2.1
TWO- PHOTON IMAGING OF CORTICAL MICROVESSELS AND ASTROCYTIC INTERACTIONS IN LIVE MOUSE BRAIN

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In vivo imaging with two-photon microscopy is becoming an indispensable technique to investigate cellular and subcellular phenomenon in living tissues including the central nervous system. This microscopy enables us to image the dynamics of molecules, morphology, and excitability with minimal invasion to tissues and with unsurpassed spatial and temporal resolution. Two-photon microscopy provides a number of advantages that aid the study of the mechanisms underlying neurovascular coupling and cerebrovascular disease in animal models, including: (i) the resolution needed to visualize single cortical vessels and their surrounding cells; (ii) penetration depths of 250μm through a PoRTS (polished and reinforced thin skull) window and 500μm with dura-removed craniotomies, and even deeper imaging with longer excitation wavelengths; (iii) reduced photodamage and photobleaching; (iv) high-speed user-defined line scans for near-simultaneous measurement of RBC velocity, lumen diameter, and local cellular activity; (v) longitudinal imaging over several months; and (vi) the ability to image vascular dynamics deep in the cortex of awake mice. This two-photon imaging method allows extremely high spatial and temporal resolution for studying pathological

mechanisms that underlie ischemic injury.
We will provide examples on how we apply these techniques to the study of local blood flow regulation and vascular pathologies such as small-scale stroke including abnormal changes in calcium cell signalling, vascular dysfunction following photothrombosis, and inflammation.

KEY WORDS: Two-photon microscopy, cranial window, neurovascular coupling, cerebrovascular disease, vascular dynamics, photothrombosis, calcium signalling.

P2.2
MODELLING SPINAL MUSCULAR ATROPHY IN DROSOPHILA: A FRUITFUL APPROACH?

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Spinal muscular atrophy (SMA) is the most common genetic killer of new-borns. The cause of this devastating neuromuscular disorder has been pinned on very low levels of the survival motor neuron (SMN) protein. SMN partners with the Gemin proteins to form a highly ordered complex. The best-characterised function of the SMN-Gemin complex involves assembly of the basic units that form the spliceosome or the chief editor of RNA messenger molecules that instruct cells how to fabricate proteins. Flies have a minimalistic complex that is amenable to genetic manipulation. We describe the phenotypes resulting from disruption of the *Drosophila* SMN complex. Our findings inform on the molecular pathway that might be negatively impacted in SMA.

KEY WORDS: Spinal muscular atrophy, *Drosophila*, survival motor neuron, SMN-Gemin complex, gemins, motor neuron degeneration.

P2.3
LIPID MEMBRANES- A NEW TARGET FOR NEURODEGENERATION

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Alzheimer's disease (AD) and Parkinson's disease (PD) are neurodegenerative disorders characterised by the misfolding of proteins into soluble prefibrillar aggregates. In our work, we have demonstrated that amyloid aggregates of recombinant amyloid-β(1-42) peptide, tau-441 and α-synuclein proteins, robustly compromised the membrane integrity of model liposomes. Interestingly, such liposome permeabilisation mimicked that of the pore-forming bacterial peptides gramicidin. Also, we screened 11 natural polyphenolic compounds, 8 synthetic N'-benzylidene-benzohydrazide compounds and black tea extract for protection against membrane damage by the amyloid aggregates. We therefore identified a select group of potent inhibitory compounds which include baicalein, morin, nordihydroguaiaretic acid and black tea extract. Since mitochondria are intimately involved in the pathophysiological cascades of both AD and PD, we further explored the interaction of soluble amyloid aggregates with mitochondrial membranes. Here, we made use of two *in vitro* model systems, namely: (i) lipid vesicles with defined membrane compositions that mimic those of mitochondrial membranes, and (ii) respiring mitochondria isolated from neuronal SH-SY5Y cells. Briefly, it was found that aggregates, but not monomers, induced a robust permeabilisation of mitochondrial-like vesicles, and triggered cytochrome c release from isolated mitochondrial organelles. Importantly, the effect on mitochondria was shown to be dependent upon cardiolipin, an anionic phospholipid unique to mitochondria and a well-known key player in mitochondrial apoptosis. Thus, we propose a generic mechanism of thrilling mitochondria in which soluble amyloid aggregates have the intrinsic capacity to permeabilise mitochondrial membranes, without the need of any other protein.

KEY WORDS: Alzheimer disease, Parkinson disease, amyloid aggregate, amyloid-β, tau-441, α-synuclein, mitochondria, baicalein, morin, nordihydroguaiaretic acid.

P2.4
IMMATURE AXONS: A NEW THERAPEUTIC TARGET FOR NEONATAL WHITE MATTER ISCHAEMIA?

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Brain injury in the premature infant, especially in very low birth weight infants, is a problem of major importance in our society. Recent advances in neonatal intensive care have dramatically increased the survival rate of such infants. Premature infants are at a great risk of developing cerebral palsy together with cognitive, attentional, behavioural, and socialization deficits that significantly impair their quality of life. Cerebral white matter injury is increasingly recognised as a common form of perinatal brain injury that predisposes to such neurological defects. Extensive studies point

to the premyelinating oligodendrocyte to be the key cellular target involved in neonatal cerebral white matter injury, due to a series of maturation-dependent events. However, the premyelinating oligodendrocyte must not be regarded as the sole target.

By imaging GFP-M expression in neonatal mice optic nerves, we found highly selective injury to ischaemia of the small-diameter fluorescent axons that corresponded to the larger pre-myelinated axons. These axons, after having initiated diameter expansion and expression of functional voltage-gated calcium channels, are exquisitely sensitive to ischemic injury. Moreover, pharmacological treatment with a combination of glutamate receptor blockers and voltage-gated calcium channel blockers offered a high degree of protection following an ischaemic insult. This elevated susceptibility of early maturing axons to ischemic injury may significantly contribute to selective white matter pathology and places these axons alongside pre-oligodendrocytes, previously regarded as the most ischemia-sensitive element within immature white matter. Therefore, future therapeutic strategies must include protection to both of these white matter elements.

KEY WORDS: Perinatal brain injury, white matter ischaemia, large pre-myelinated axons, optic nerve, voltage-gated calcium channels.

P2.5
THE NEED FOR RELIABLE BIOMARKERS FOR MONITORING POTENTIAL TREATMENTS IN ALZHEIMERS DISEASE

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The search for predictive biomarkers for Alzheimer's disease (AD) is of high priority in neurodegenerative disease research underlined by the lack of significant progress in identifying new treatments for the past 12 years. Despite major efforts and considerable investments, the treatments approved for AD are only palliative. They include cholinesterase inhibitors (donepezil, galantamine, and rivastigmine) that act on the cholinergic deficit, and the NMDA receptor antagonist, memantine, which has neuroprotective effects. These agents are generally considered to have marginal efficacy. As it can be logically assumed that a late therapeutic intervention would be less efficient than an early one, development of biomarkers for AD both to diagnose the disease early and to follow-up its progression, remains a major challenge. Currently, it is comprised of 6 main approaches: 1) behavioral assessment, including measurement of cognitive status using various neuropsychological scales (MMSE, ADAS-Cog, etc.); 2) changes in brain structure (mainly volume of the cerebral cortex, particularly entorhinal cortex and hippocampus); 3) alterations in brain metabolism (most notably within the default mode network) by using FDG-PET; 4) measurement of β-amyloid load within the brain by PIB-PET; 5) cerebrospinal fluid (CSF) biomarker profiles (the three main CSF biomarkers of AD being β-amyloid, total tau, and phosphorylated forms of tau proteins); and 6) post-mortem confirmation of characteristic AD histopathology. In my talk I will attempt to describe new developments within each of these biomarker approaches, analyzing their pathological specificity, early diagnostic sensitivity, and correlation with AD progression. Finally, I will argue that, despite numerous publications and recommendation criteria, the predictive usefulness of these various biomarker approaches, individually or collectively, has yet to be established.

KEY WORDS: Alzheimer's disease, biomarkers, default mode network, DTI, fMRI, PET.

P2.6
5-HT2C RECEPTORS: A G-PROTEIN COUPLED RECEPTOR INVOLVED IN OPPOSITE AND DISTRIBUTED CONTROLS IN BASAL GANGLIA

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5-HT_{2C} receptors, one of the seven-transmembrane G-protein coupled receptors for serotonin (5-HT), is a potential therapeutic target of numerous diseases, such as Parkinson's disease and schizophrenia, that involve combined dysfunctions of dopamine (DA) transmission and basal ganglia, a group of subcortical structures involved in motor behaviours. 5-HT_{2C} receptors, present in the whole basal ganglia, would exert tonic, phasic and constitutive controls, the latter being independent of the presence of 5-HT. Using appropriate 5-HT_{2C} receptor pharmacological tools (agonists for phasic, antagonists for tonic, inverse agonists for constitutive control), we have addressed in rats the organisation of these different controls on a motor behaviour, the purposeless orofacial movements, on the expression of the proto-oncogene c-Fos, a marker of change of

neuronal activity, and on the electrophysiological responses of neurons located in the output structures of basal ganglia, namely the entopeduncular nucleus (EPN) or the substantia nigra *pars reticulata*.

Both 5-HT_{2C} agonists and inverse agonists increased abnormal orofacial movements via 5-HT_{2C} receptors. c-Fos imaging studies indicated that different 5-HT_{2C} controls are expressed in the input structures of the basal ganglia, the striatum and the subthalamic nucleus. In addition, agonists and inverse agonists altered neuronal activity in the output structures which could be associated with the emergence of orofacial movements. 5-HT_{2C} controls are influenced by the level of DA transmission. Indeed, DA neurons lesion potentiated behavioural and electrophysiological responses induced by a 5-HT_{2C} agonist by acting in the EPN. The stimulation of D2 receptors enhanced oral dyskinesia and electrophysiological responses of the cortico-subthalamonigral pathway; these effects were suppressed by selective 5-HT_{2C} antagonists. This work illustrates the complexity of the controls exerted by 5-HT_{2C} receptors and their outcome with respect to central DA transmission. A better understanding of the controls in these regions would permit to apprehend possible treatments using $5-HT$ and/or 5-HT_{2C} agents.

KEY WORDS: Serotonin 2C receptor, basal ganglia, dopamine, dyskinesia, parkinson's disease, entopeduncular nucleus, subthalamic nucleus, substantia nigra pars reticulata, striatum.

P2.7
POTASSIUM CHANNELS AS A TARGET OF CNS DISORDERS

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K^+ channels are critical for neuronal excitability and they are essential effectors of neurotransmitter-mediated signaling. They are distinguished by being the largest and most diverse class of ion channels, being encoded by more than 70 genes. In the past decades several types of human diseases have been associated to dysfunction of K^+ channels, resulting from mutations in their encoding genes. Indeed, K^+ channels defects underlie a number of distinct forms of epilepsies that have been named "K^+ *channelepsies*". Also different types of ataxias have been associated with altered K^+ channels function. In particular we have shown that episodic ataxia type 1 (EA1), a K^+ channelopathy, which manifests with short attacks of cerebellar ataxia, is caused by *loss-of-function* mutations in Kv1.1 (KCNA1) channels. The direct and indirect involvement of K^+ channels in a number of psychiatric disorders including autism spectrum disorders (ASDs), schizophrenia, and mental retardation has been reported. ASDs are characterized by impaired ability to properly implement environmental stimuli that are essential to achieve a state of cultural and social inter-relationships. The main features of this disease are marked impairments of verbal and non-verbal communication with restricted and repetitive behaviors. We have performed the genetic analysis of individuals affected by autism and epilepsy and identified new heterozygous point mutations in the KCNJ10 gene that encodes the inwardly-rectifying K^+ channel Kir4.1, expressed predominantly, but not exclusively, in astrocytes. Functionally, the mutated channels exhibited a phenotype consistent with *gain-of-function* defects. These new findings highlight the emerging role of inwardly-rectifying K^+ channels and astrocyte dysfunction in autism spectrum disorders associated with epilepsy.

KEY WORDS: Potassium channels, mutation, epilepsy, ataxia, K^+ channelopathy, ASD, astrocyte dysfunction, inward- rectifying K^+ channels.

P2.8
EFFECT OF ACUTE AND REPEATED NICOTINE ADMINISTRATION ON THE ELECTRICAL ACTIVITY OF THE LATERAL HABENULAR NEURONS IN RATS

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Tobacco smoking represents a well-known risk factor for health that still accounts for a high number of deaths. So far, existing smoking cessation therapies have not been proven very successful at quitting this habit and a better undrstanding of the neurobiology of tobacco dependence is still needed. Nicotine is the neuroactive compound contained in tobacco that is responsible for its rewarding and reinforcing properties by acting on the midbrain dopaminergic system. The lateral habenula (LHb) is an epithalamic structure involved in pain, stress, depression and in encoding

aversive stimuli. This structure is known to indirectly inhibit the DA system through the activation of the RMTg, a GABA-ergic area located at the back of the VTA. The RMTg receives a strong glutamatergic imput from the LHb and is activated by the systemic injection of nicotine in rats. Thus the LHb might represent a possible target for the action of nicotine. Our data shows that systemic administration of nicotine dose-dependently increases the activity of single LHb neurons recorded extracellularly in vivo in rats, particularly at high doses. Following two weeks of nicotine chronic treatment, this response is drastically decreased while after 1 day of withdrawal only low doses of nicotine are again able to significantly increase the firing activity of the LHb neurons compared to the control group. These evidences strongly suggest that the LHb might play an important role in mediating the effects of nicotine on the midbrain DA system thus participating to the mechanism of addiction to this drug.

KEY WORDS: Drug of addiction, extracellular recording, serotonin, dopamine.

P2.9 OLIGODENDROCYTE PATHOPHYSIOLOGY AND TREATMENT STRATEGIES IN ISCHEMIA

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Oligodendroglia, the myelin-forming cells of the CNS, form a functional unit with axons and play a crucial role in axonal integrity. An episode of hypoxia-ischemia causes rapid and severe damage to these particularly vulnerable cells via the overactivation of glutamate and ATP receptors (excitotoxicity), oxidative stress and mitochondrial disruption. Oligodendrocytes appear to be more vulnerable to HI than other CNS glia, and in certain brain regions and stages of development, more vulnerable than neurons, due to the possession of numerous features, which predispose them to injury. The cardinal effect of oligodendrocyte pathology is demyelination and dysmyelination, and has profound effects on axonal function, transport, structure, metabolism and survival. The oligodendrocyte is a primary ischemic target, in adult-onset stroke and especially in periventricular leukomalacia, and should therefore also be considered a primary therapeutic target. Further emphasis is required on therapeutic strategies targeting oligodendroglia, myelin and their receptors, as these have the potential to significantly attenuate white-matter injury in hypoxia-ischemia.

KEY WORDS: Excitotoxicity, hypoxia-ischemia, oligodendrocyte, oxidative stress, stroke.

P2.10 AMYLOID NEURODEGENERATION: FROM ELECTROPHYSIOLOGY TO FLIES

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Alzheimer's, Parkinson's and Motor Neuron disease are characterized by the deposition of abnormally aggregated forms of Aβ1-42, α-synuclein and TDP-43, respectively. An intriguing possibility that is being investigated, is the possibility of pore formation in mitochondrial membranes by aggregates of these proteins. Such pores can have deleterious consequences on the electrical physiology of a neuron.

Electrophysiology studies are performed using a lipid bilayer workstation, which allows detailed electrophysiological characterisation upon incubation of amyloid aggregates with mitochondrial membranes. Electrical currents at the level of a single channel are recorded, and changes in membrane permeability can be correlated to toxic channel activity. The potential of natural polyphenols and bioactive extracts to block amyloid pores will be assessed, thereby preventing disruption of neuronal ion homeostasis.

Currently there are no drugs or clear-cut pathogenic mechanisms that do more than improve the symptoms associated with these diseases. Identification of compounds that lead to a marked and consistent recovery, will be a great asset to developing new therapeutic approaches.

Drosophila models of neurodegenerative disease have been successfully used in whole-genome screens aimed at identifying genetic modifiers, which can lead to the discovery of drug targets. The disease fly models are being generated by the overexpression of the respective human transgene in the wild-type fly brain.

A graded dose of a select group of test drugs are being tested and adult flies monitored for survival and climbing ability using well-established protocols. Data will be analysed to determine whether the drug-supplemented diet markedly, and consistently ameliorates the phenotypic defects intrinsic to the disease fly models.

KEY WORDS: Amyloid, neurodegeneration, Drosophila, mitochondria, aggregates, drugs.

P2.11
POSITIVE ALLOSTERIC MODULATION OF GABA-B RECEPTORS: A NOVEL THERAPEUTIC APPROACH FOR SCHIZOPHRENIA?

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Preclinical and clinical investigations have suggested that Gamma-amino-butyric acid $(GABA)_B$ receptors may play a key role in the pathophysiology of psychiatric disorders. We previously reported that baclofen, the prototypical $GABA_B$ agonist, exerts antipsychotic-like properties in two well-validated rodent models of schizophrenia, the prepulse inhibition (PPI) deficits produced by dizocilpine (MK-801) and the genetically low PPI displayed by DBA/2 mice. However, the adverse side effects elicit by Baclofen, point to develop alternative therapeutic tools for regulating $GABA_B$ in schizophrenia.

Thus, we investigated the impact of a new allosteric enhancers of $GABA_B$, *rac-BHFF* (RAC), on the MK-801 mediated-PPI disruption in Sprague-Dawley (SD) rats and C57/BL mice, two of the most used rodent species in PPI with high baseline of PPI and susceptibility to the NMDA receptor manipulations. Furthermore, we evaluated the properties of RAC in ameliorating the naturally low PPI performance displayed by DBA/2J, in comparison with the positive control antipsychotic, clozapine. RAC did not produce any effects on PPI per se and dose-dependently counteracted the PPI impairments produced by MK-801 in both SD and C57. Notably, dissimilar to Baclofen, these effects were not accompanied with significant alterations of startle parameters. Moreover, RAC was able to restore PPI deficits in DBA/2J, akin to the atypical antipsychotic clozapine.

Our data strengthen previous evidence of $GABA_B$ receptors as an important biological target for the modulation of PPI and suggest a new potential therapeutic application in neuropsychiatric disorders related to sensorimotor gating dysfunctions, without exerting the side effects share by the putative $GABA_B$ agonists.

KEY WORDS: $GABA_B$, PAM, pre-pulse inhibition, sensorimotor gating, schizophrenia, NMDA receptors.

P2.12
EFFECT OF NEW MULTITARGET-DIRECTED LIGANDS BASED ON DONEPEZIL, PYRIDYL AND INDOLYL HYBRIDS ON MONOAMINERGIC AND CHOLINERGIC SYSTEMS: AN HPLC METABOLIC APPROACH

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The multifactorial nature of Alzheimer's disease (AD) has prompted the search for new Multitarget-Directed Ligands (MTDL) able to simultaneously bind both cholinesterases and monoamine oxidases. We have developed and assessed novel series of MTDLs based on Donepezil-Indolyl hybrids [**MBA98F1** (IC_{50}); AChE= 0.19μM; BuChE= 0.83μM; MAO A= 5.5nM; MAO B= 0.15μM], Donepezil-Pyridyl hybrids [**MBA115** (IC_{50}); AChE= 1.4nM; BuChE= 0.51μM; MAO A= 53.3μM; MAO B= 10.2μM] or α-Aminonitriles hybrids [DHP6 (IC_{50}); AChE= 1.8μM; BuChE= 1.6μM; MAO A= 6.2μM; MAO B= 10.2μM; with metal-chelating properties] for their potential pharmacological use in AD.

The effect of the MAO A-selective inhibitors clorgyline and the multipotent **ASS234** on the monoaminergic system, was also evaluated on human neuroblastoma SHSY-5Y and undifferentiated pC12 cell lines. High activity levels of MAO A were determined in both cell lines; this activity was fully inhibited after treating the cells with 1μM of clorgyline or **ASS234** for 24 hours.

Both inhibitors were able to modulate the levels of monoamines by HPLC after treatments. Levels of serotonin (5-HT) and 3-metoxytryptamine (3-MT) were significantly increased while those of dopamine, DOPAC, 5HIIA and homovanillic acid (HVA) decreased. The levels of noradrenaline and L-DOPA remained unaltered. These results suggest that novel multipotent inhibitors herein presented deserve further investigation for their potential pharmacological treatment of Alzheimer's disease.

KEY WORDS: Multipotent drugs, Fe/Cu Chelators, ChE/MAO inhibitors, monoamines, Alzheimer's disease.

P3.1
DESIGN, SYNTHESIS AND EVALUATION OF MODULATORS COUNTERACTING ABAD-AβINTERACTION

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Although the aetiology of AD is still unknown, the build-up of amyloid β-peptide (Aβ) is considered to play a central role in the pathogenesis of the disease. It is well established that the intracellular accumulation of Aβis associated with AD and increasing evidence suggests that mitochondria may be an important target for intracellular Aβto exert its neurotoxic effects.

Amyloid-binding alcohol dehydrogenase (ABAD) is to date the most characterized Aβ-binding intracellular protein. Direct interaction of this mitochondrial enzyme with Aβwas confirmed by many different methods. Aβbinding to ABAD triggers a series of events leading to mitochondrial dysfunction characteristic for AD. Thus this interaction may represent a novel target for treatment strategy against AD.

The benzothiazole urea analogues related to known immunosuppressant frentizole was synthesized and in vitro evaluated for its capability to inhibit interaction between ABAD and Aβ. Several prepared compounds showed ability to inhibit ABAD *in vitro*. These promising compounds are going to be further tested on living cells.

KEY WORDS: Mitochondria, ABAD, β-amyloid, inhibitor, benzothiazole.

P3.2
In vivo AND *in vitro* BIOLOGICAL ASSESSMENT OF ASS234, A NOVEL DONEPEZILINDOLPROPARGYLAMINE, AS A MULTIFUNCTIONAL MOLECULE WITH A POTENTIAL THERAPEUTIC PROFILE FOR ALZHEIMER'S DISEASE

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A key pathological hallmark of AD is amyloid beta (Aβ aggregation and deposition. Growing evidence suggest that the neurotoxicity of these peptide is related to the formation of toxic oligomeric aggregates. Thus, a deeply investigated therapeutic strategy comes at present from blocking the formation of these species to non-toxic aggregates. Nevertheless, clinical trials evaluating anti-Aβ drugs are not giving conclusive results and brain penetration of these molecules is also an important challenge to be solved. The multifactorial nature of Alzheimer's disease (AD) supports the most current innovative therapeutic approach, which proposes that single molecules acting on multiple targets might be more suitable for the treatment. Thus, molecules possessing a rich pharmacology are of great interest. In this context, we recently identified ASS234, a new multipotent drug showing an interesting inhibitory profile towards cholinesterase and monoamine oxidase enzymes, possessing also a significant anti-Aβ aggregation activity. In this work, we explore more in detail its anti-Aβ activity and show that ASS234 reduces Aβ_{1-42} aggregation more efficiently than that of Aβ_{1-42}, as well as completely blocks the AChE-induced Aβ_{1-42} and Aβ_{1-42} aggregation. We also describe that ASS234 is able to limit the Aβ_{1-42}-mediated cytotoxicity, by preventing the activation of the mitochondrial pathway of apoptosis. Moreover, we demonstrate a significant ability of ASS234 to reduce oxidative stress and the finding of its capability to cross the blood-brain barrier. Overall, our results demonstrate that ASS234 is able to bind to multiple targets and suggest that it might be considered for therapeutic development against AD.

KEY WORDS: Alzheimer's disease therapy, amyloid cholinesterase inhibitors, multi-target directed ligand, neuroprotection, propargylamines.

P3.3
SEROTONERGIC RECEPTORS: THE NEW TARGETS IN THE TREATMENT OF ALZHEIMERS DISEASE

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Serotonergic neurotransmission is implicated in the modulation of many physiological (sleep, sexuality, appetite), behavioural (aggression, mood) and cognitive (learning, memory) functions, which change in aging related disorders. *In vivo* and *in vitro* evidence suggest neuroprotective and pro-cognitive effect of serotonin in Alzheimer's disease (AD). Serotonin also plays a crucial role in the development of behavioural and psychological symptoms of dementia (BPSD) which are present in up to 90% of patients with AD.

Complex functions of the serotonergic system depend on the activity and function of its receptors, classified in seven groups from 5-HT1 to 5-HT7, which differ in terms of structure, action and location. The loss of 5-HT2, 5-HT6 and pre- or post-synaptic 5-HT1A and 5-HT1B receptors were found in patients with AD. It is unclear if these changes are primary or secondary (retrograde), due to the damage of postsynaptic target neurons in regions of the nerve endings. The activation of 5-HT3 and 5-HT4 receptors enhances the acetylcholine release and could induce the pro-cognitive effect. Since preclinical studies have shown that agonists of 5-HT4 and antagonists of 5-HT1A, 5-HT3 and 5-HT6 receptors improve cognitive functions, serotonergic receptors might represent the new pharmacological target for the treatment of AD and BPSD.

KEY WORDS: Serotonin, receptor, Alzheimer's disease, medications.

P3.4
THE REVISITED MAO INHIBITION BY N-(Furan-2-ylmethyl)-N-prop-2-yn-1-amine DERIVATIVES

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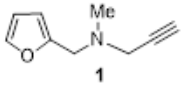

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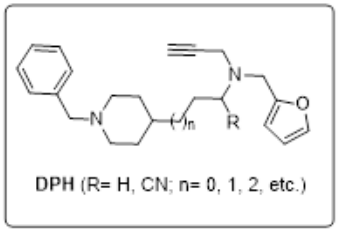

The MAO inhibition analysis of N-(furan-2-ylmethyl)-N-methylprop-2-yn-1-amine (1) has been revisited, showing that this propargylamine is a moderate, but selective, partially reversible and uncompetitive MAO B inhibitor ($IC_{50} = 5.16 \pm 0.86$ μM), whose ADMET properties predict the best profile for acting as CNS drug.

This result paves the way for the projected synthesis and biochemical analysis of new **DPH** ("Donepezil+Propagyl+Hybrid") multipotent molecules, as drugs for the potential treatment of Alzheimer's disease.

KEY WORDS: MAO enzymes, inhibitors, propargylamines, kinetics.

P3.5
SST2 AND SST3 - BUT NOT GSHR-RECEPTORS ARE INVOLVED IN THE ANTICONVULSANT EFFECTS PF CORTISTATIN-14

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Anticonvulsant and antiepileptic actions of somatostatin-14 have already widely been studied and are thus well known. For the related neuropeptide cortistatin-14 however, only one paper reports on its anticonvulsant effects. Somatostatin-14 and cortistatin-14 are structurally related peptides and have high affinities for the five somatostatin receptor subtypes (sst1-sst5). Despite these homologies, cortistatin-14 seems to act also on other receptors and it has been suggested that the ghrelin receptor (GHSR) may fulfill such a role. Here, we aim to unveil which receptors are involved in the anticonvulsant effects of cortistatin-14 by using in vivo microdialysis and telemetry-based

electrocorticography (ECoG) in rats and mice.
In rats, the involvement of sst2 and sst3 receptors was studied by administering cortistatin-14 (0.1μM - 1μM - 10μM) intrahippocampally, in the presence and absence of sst2 and sst3 receptor antagonists. Seizures were evoked by intrahippocampal pilocarpine perfusion (12mM, 40min) and seizure severity was assessed using a behavioural scoring system and ECoG. Intrahippocampal administration of 1μM and 10μM cortistatin -14 in rats showed clear anticonvulsant actions against pilocarpine-induced seizures. Furthermore, we showed that cortistatin -14 (1μM) - mediated anticonvulsant actions were reversed in the presence of 0.1μM cyanamid, a selective sst2 antagonist or 0.1μM SST3-ODN8, a selective sst3 antagonist. Intrahippocampal perfusion of these antagonists alone did not affect the pilocarpine-induced seizure severity per se.
The involvement of GHSR was tested by administering an anticonvulsant dose (1μM) of cortistatin -14 in both GHSR knock-out (KO) and wild-type (WT) mice. Seizures were evoked by intrahippocampal pilocarpine perfusion (12mM, 40min), and ECoG was used to assess seizure severity, by means of seizure duration. In these mice, both genotype - and treatment dependent alterations in seizure severity were observed by means of two-way ANOVA. Indeed, our results showed that the seizure duration in WT animals was significantly higher, when compared to their KO littermates, and that the seizure duration in the CST- treated animals was significantly lower when compared to the animals receiving only pilocarpine.
In conclusion, our results show that cortistatin -14 prevents seizures in a focal pilocarpine model and that selective sst2 or sst3 receptor antagonism abolishes these anticonvulsant actions in rats. Our findings also demonstrate that the anticonvulsant actions of cortistatin -14 in mice are not mediated via the GHSR receptor.

KEY WORDS: Somatostatin, cortistatin, epilepsy, seizures, pilocarpine, ghrelin.

P3.6
BIOISOSTERIC REPLACEMENTS FOR OPTIMIZED DOPAMINE RECEPTOR AGONISTS

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LevoDOPA and dopamine agonists have been in therapeutic use for the symptomatic treatment of Parkinson's disease for a long time. Despite the success of this medical approach, numerous unwanted side effects and an unclear receptor-crosstalk raise the need for new and improved therapeutics. Based on the early discovery of Etrabamine and established on the non-ergot dopamine agonist Pramipexole we have developed a series of tetrahydrobenzothiazole derivatives with high receptor affinity, improved receptor subtype selectivity and different efficacy profiles from agonist to antagonist properties. The 2-aminothiazole moiety of Pramipexole has generally been taken as catechol bioisosteric moiety. The replacement of the 2-amino functionality, as well as the modification of the heterocycle, led to novel classes of compounds with moderate to excellent affinity at dopamine D2 and/or D3 receptor subtypes. The synthesis has been performed by reductive amination of cyclohexa-1,4-dione monoketal, followed by deprotection and heterocyclic ring formations by different procedures. Deamination in 2-position could be performed by diazonium formation under reductive conditions.

Some of these derivatives displayed up 400fold binding preference for D3 over D2 receptors, whereas in a functional assay on $[^{35}S]GTP_{\alpha}S$ the binding the preference was less pronounced. Particular compounds showed an impressive biased signaling. The pharmacological *in vivo* profile was assessed for selected compounds in a Parkinsonian model, on 6-OHDA lesioned rats with intraperitoneal (i.p.) and *per os* (p.o.) administration, showing a good potential for further development not only for Parkinson's disease but also for erectile dysfunction.

KEY WORDS: Affinity, biased signaling, D2 receptor, D3 receptor, efficacy, pramipexole.

P3.7
SYNTHESIS, PHARMACOLOGICAL ASSESSMENT AND MOLECULAR MODELING OF ACETYLCOLINESTERASE/BUTYRYLCHOLINESTERASE INHIBORS: EFFECT AGAINST AMYLOID-BETA - INDUCED NEUROTOXICITY

Daniel Silva[1,2], **Mourad Chioua**[2], **Abdelouahid Samadi**[2], **Paula Agostinho**[3,4], **Pedro Garção**[3,4], **Rocio Lajarin-Cuesta**[5], **Cristobal de los Rios**[5], **Isabel Iriepa**[6], **Ignacio Moraleda**[6], **Laura Gonzalez-Lafuente**[5], **Eduarda Mendes**[1], **Concepción Pérez**[7], **Maria Isabel Rodriguez-Franco**[7], **José Marco-Contelles**[2], **and M. Carmo Carreiras**[1]
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Synthesis, molecular modeling, and pharmacological analysis of phenoxyalkylamino-4-phenylnicotinates (**2-7**), phenoxyalkoxybenzylidenemalononitriles (**12-13**), pyridonepezils (**14-18**), quinolinodonepezils (**19-21**), and pyrazolo[3,4-b]quinolines (**35-37**) will be summarized in this talk. The most potent and selective EeAChE inhibitor was ethyl 6-(2-(1-benzylpiperidin-4-yl)ethylamino)-5-cyano-2-methyl-4-phenylnicotinate (**16**) [IC50 (EeAChE)= 0.0167 ± 0.0002 μM], which exhibits the same inhibitory potency as donepezil against hAChE. The most potent and selective hAChE inhibitor was ethyl 6-(4-(1-benzylpiperidin-4-yl)butylamino)-5-cyano-2-methyl-4-phenylnicotinate (**18**) [IC50 (hAChE)= 0.25 ± 0.02 μM]. Pyridonepezils showed to be selective and moderately potent against hAChE inhibition, whereas quinolinodonepezils showed to be poor hAChE inhibitors. Compounds **2, 7, 13, 17, 18, 35** and **36** significantly prevented the decrease in cell viability caused by $A\beta_{1-42}$. All compounds were effective in preventing the enhancement of AChE activity induced by $A\beta_{1-42}$. Compounds **2-7** caused a significant reduction whereas pyridonepezils **16-18** also showed some activity. The pyrazolo[3,4-b]quinolines **36** and **38** also prevented the upregulation of AChE induced by $A\beta_{1-42}$. Compounds **2, 7, 12, 13, 17, 18** and **36** may act as antagonists of VSCC since they significantly prevented the Ca^{2+} influx evoked by KCl depolarization. Docking studies show that compounds **16** and **18** adopted different orientations and conformations inside the active-site gorges of hAChE and hBuChE. The structural and energetic features of the **16**-AChE and **18**-AChE complexes compared to the **16**-BuChE and **18**-BuChE complexes account for a higher affinity of the ligand toward AChE. Compounds **2, 7, 17, 18** and **36** are attractive multipotent molecules acting in different key pharmacological targets. They may accomplish a potential disease-modifying role in the treatment of Alzheimer's disease.

KEY WORDS: Alzheimer's disease, pyridonepezils, AChE/BuChE inhibitors, Aβpeptide, neuroprotection, Ca^{2+} dyshomeostasis.

P3.8
RECENT ADVANCES IN THE STUDY OF 5-HT7 RECEPTOR PHARMACOLOGY: FOCUS ON THE SELECTIVE AGONIST LP- 211

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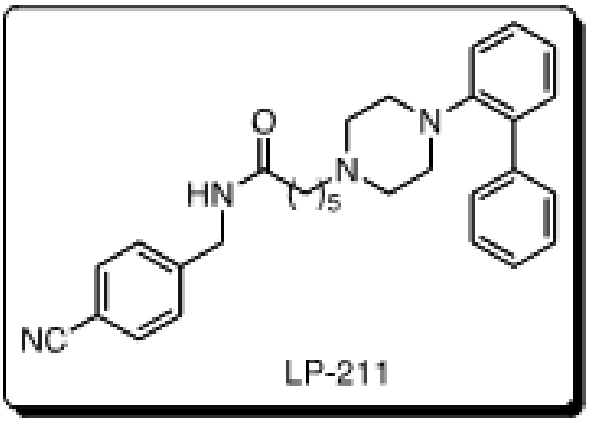
Twenty years after the 5 HT7 receptor was first cloned, there is a large amount of data available in terms of the pathophysiology of this serotonin receptor. Medicinal chemistry efforts have resulted in the identification of 5 HT_7 receptor selective agonists and antagonists. While 5 HT_7 receptor antagonists have been proposed as antidepressant drugs, the possible therapeutic applications of selective activation of 5 HT_7 receptor are emerging in recent years after various selective agonists became available. This lecture will illustrate the process that led to the identification of various selective 5 HT_7 receptor agonists in our laboratory, following structure-activity relationship studies on "long-chain" arylpiperazine derivatives. The studies culminated with the discovery of LP-211, a brain-penetrant selective 5 HT_7 receptor agonist.



Ki [nM].

r5-HT_7	h5-HT_7	h5-HT_{1A}	h5-HT_{1B}
0.58	15	379	215
h5-HT_{1E}	h5-HT_{2A}	h5-HT_{2B}	r5-HT_{2C}
> 10000	626	67	91
h5-HT_3	h5-HT_{5A}	h5-HT_6	
> 10000	178	1571	

Recent studies conducted with LP-211, have suggested that selective activation of 5 HT_7 receptors may represent a novel strategy in the therapy of Fragile-X syndrome; the most common form of inherited intellectual disability and autistic spectrum disorders. Moreover, treatment of murine striatal and cortical

neuronal cultures with LP-211 significantly enhances neurite outgrowth, suggesting the involvement of 5 HT_7 receptor in shaping central nervous system connectivity, which may be intimately linked to psychiatric and neurodevelopmental disorders.

KEY WORDS: Serotonin 5-HT_7 receptor, arylpiperazine, central nervous system.

P3.9
ROLE OF HETEROOLIGOMERIZATION BETWEEN SEROTONIN RECEPTORS 5-HT1A AND 5-HT7 IN REGULATION OF RECEPTOR FUNCTIONS

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Serotonin receptors 5-HT_{1A} and 5-HT_7 are highly co-expressed in brain regions implicated in depression. However, their functional interaction has not been established. In the present study we show that 5-HT_{1A} and 5-HT_7 receptors form heterodimers both in vitro and in vivo. Resonance energy transfer-based assays revealed that, in addition to heterodimers, homodimers composed either by 5-HT_{1A} or 5-HT_7 receptors together with monomers co-exist in cells. The highest affinity to form the complex was obtained for the 5-HT_7-5-HT_7 homodimers, followed by the 5-HT_7-5-HT_{1A} heterodimers and 5-HT_{1A}-5-HT_{1A} homodimers. Functionally, heterodimerization decreases 5-HT_{1A} receptor-mediated activation of Gi-protein without affecting 5-HT_7 receptor-mediated signalling. Moreover, heterodimerization markedly decreases the ability of the 5-HT_{1A} receptor to activate G-protein gated inwardly rectifying potassium channels in a heterologous system. The inhibitory effect on such channels was also preserved in hippocampal neurons, demonstrating a physiological relevance of heteromerization in vivo. In addition, heterodimerization is critically involved in initiation of the serotonin-mediated 5-HT1A receptor internalization and also enhances the ability of the 5-HT1A receptor to activate the mitogen-activated protein kinases. Finally, we found that production of 5-HT_7 receptors in hippocampus continuously decreases during postnatal development, indicating that the relative concentration of 5-HT_{1A}-5-HT_7 heterodimers and, consequently, their functional importance undergoes pronounced developmental changes.

Generally, our data suggest that the regulated and balanced ratio of homo- and heterodimerization on pre- and postsynaptic neurons may be critically involved in both, the onset as well as response to treatment of psychiatric diseases such as depression and anxiety.

KEY WORDS: G-protein coupled receptors, serotonin, hetero-oligomerization.

P3.10
PHARMACOPHORE MODELING OF NOVEL NONIMIDAZOLE HISTAMINE H3 RECEPTOR LIGANDS WITH INHIBITORY HISTAMINE N-METHYLTRANSFERASE ACTIVITY

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Dual acting compounds able to enhance histaminergic neurotransmission in the central nervous system, are a novel class of nonimidazole histamine H_3 receptor (H_3R) antagonists, that simultaneously possess strong inhibiting potency on catabolic histamine N-methyltransferase (HMT). The set of thirty-five multipotent H_3R/HMT ligands containing a piperidinoalkyl group, are a key structural feature for human H3 receptor (hH_3R) antagonism; connected by different spacer lengths to an aminoquinoline moiety, have been studied as a pharmacophoric moiety for HMT inhibiting activity, by the use of 3D-QSAR (Quantitative Structure-Activity Relationship) and pharmacophore study.

In order to better understand the crucial chemical functionalities for combined hH3R/HMT activities, 3D-QSAR pharmacophore models for hH3R antagonistic and HMT inhibiting activities were developed using Pentacle 1.06 program. Created 3D-QSAR models (hH_3R: R^2 (0.98), Q^2 (0.94), RMSE (0.171); and HMT: R^2 (0.80), Q^2 (0.60), RMSE (0.159)) showed different important DRY, TIP and related variables as essential 3D-pharmacophoric feature for both activities. 3D-Pharmacophoric features for hH_3R antagonistic activity mainly differs from the pharmacophore for HMT inhibiting activity in presence of specific lipophilic/steric components of the hH_3R pharmacophore. The H-bond accepting components of the hH3R pharmacophore, H-bond donating components of the HMT pharmacophore, and a longer optimal distance between H-bond donor and steric

hot spots were observed in the hH3R pharmacophore than in the HMT pharmacophore. Formed 3D-QSAR models were applied for design of novel piperidino-aminoquinoline hybrids, as multitarget hH3R/HMT ligands with a potential therapeutic impact in sleep-wake disorders and cognitive impairment. Designed compounds with 3D-QSAR predicted pKi(hH_3R)> 9.6 and (pKi(hH_3R)+pIC_{50}(HMT))> 16.8 were selected for further study.

KEY WORDS: Histamine H3 receptor, histamine N-methyltransferase, pharmacophore, QSAR, drug design.

P3.11 CYP-DEPENDENT METABOLISM AND VASCULAR EFFECTS OF ASS234, A NOVEL MULTITARGET DIRECTED LIGAND

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The determination of the metabolic profiles and the safety of a new drug provides information that might be used to guide further modifications of a chemical, in order to obtain favorable therapeutic properties. In this context, cytochrome P450 (CYP) plays a crucial role in metabolism and toxic action of a drug.
Several monoamine oxidase (MAO) inhibitors present a propargylamino moiety. This chemical group confers properties as irreversible inhibitors towards the MAO and could represent a potential molecular site in the formation of suicide substrates toward CYP, which could be the origin of drug-drug interactions. Furthermore, concerning the safety pharmacology, important aspects that have been highlighted are the interactions with the cardiovascular system. For these reasons the metabolic features of a new series of PF9601N derivatives, characterized by MAO and acetylcholine esterase (AChE) inhibiting properties were studied in human liver microsomes, and the vascular effects were studied in the rat aorta rings.
The compounds presented a concentration-dependent inhibition of CYP(s), however this effect resulted in a fully reversible and a competitive fashion. Furthermore the lead compound ASS234, showed an intrinsic clearance value of CLint= $1.7\mu l x min^{-1} \times mg^{-1}$ and CLint= $129.2\mu l x min^{-1} \times mg^{-1} \times mg^{-1}$ in human and rat respectively, indicating that ASS234 is a poor substrate for human CYPs.
In the vascular studies, ASS234 showed to relax phenylephrine-induced contraction at concentrations, either > 3 μM (endothelium denuded) or > 1 μM (endothelium intact rings). The vasodilating effects exhibited by ASS234, however, were at concentrations two orders of magnitude greater than those effective on AChE, and three orders greater than those effective on MAO. These preliminary *in vitro* results, suggest that ASS234 may have a safety vascular profile.

KEY WORDS: Multitarget compounds, metabolic stability, cytochrome P450, liver microsomes, aorta rings.

This work was realized in the framework of COST CMST Action CM1103 and working group D34/0003

P3.12 RECENT PROGRESS IN UNDERSTANDING THE CATALYTIC ACTIVITY OF MONOAMINE OXIDASES

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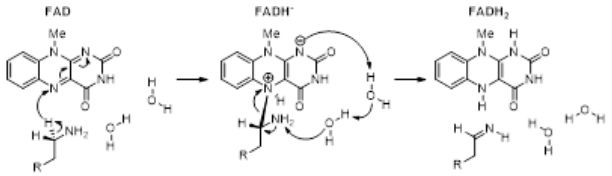
Monoamine oxidase (MAO) is a flavoenzyme responsible for regulating the level of biogenic amines in various parts of brain. Although MAO have been the central pharmacological targets in treating depression and Parkinson's disease for over 60 years, there has been no consensus in the literature about the precise molecular mechanism of its catalytic activity. On the basis of model quantum chemical calculations, we have proposed a new two-step hydride mechanism for the MAO-catalysed oxidative deamination of amines (Scheme 1). In the rate-limiting first step, the flavin N5 atom directly abstracts the hydride anion from the substrate α-carbon atom, and forms a strong covalent adduct intermediate with the thus created cationic substrate. This is subsequently followed by the deprotonation of the substrate amino group to the flavin N1 atom, facilitated with two active-site water molecules, which produces fully reduced flavin, $FADH_2$, and releases neutral imine.

This presentation discusses the significance of the mentioned flavin-substrate adduct formation, since its non-equal feasibility in both MAO isoforms has been suggested, and implied that this feature might play a crucial role in determining differences in catalytic mech-



Scheme 1. Complete two-step mechanism of MAO catalysed amine degradation.

anisms, and substrate selectivities between MAO-A and MAO-B enzymes. Also, we present some results of our preliminary all-atom QM/MM simulations within the Empirical Valence Bond theory that provide further support in favour of the hydride mechanism, particularly in the context of very recent papers by other researchers that do not necessary agree with all aspects of our mechanistic proposal.

KEY WORDS: Amine metabolism, computational chemistry, flavoenzymes, monoamine oxidases, neurodegenerative disorders.

P3.13
In silico DESIGN OF NOVEL AND SELECTIVE NEURONAL NITRIC OXIDE SYNTHASE (nNOS) INHIBTIORS

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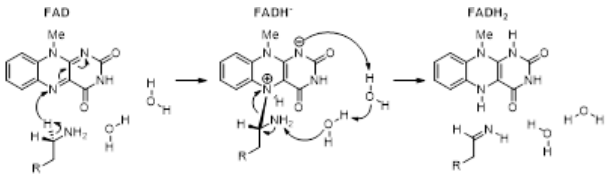
Nitric oxide gaseous free radical molecule (NO) acts as a messenger in various tissues and is responsible for different physiological functions and pathological symptoms. Nitric Oxide synthases (NOS) catalyse the oxidation of L-Arginine to a nitric oxide molecule (NO) and L-citrulline (Figure 1). Mammals contain three different NOS isozymes: Neuronal NOS (nNOS, in the brain), inducible NOS (iNOS, in macrophage cells), endothelial NOS (eNOS, the inner walls of blood vessels). Indeed, NO is a free radical gaseous molecule under normal conditions that is a highly toxic substance to our cells. In our body, it is produced locally at proper concentration and proper time. In endothelial cells, it relaxes smooth muscle causing a decreased blood pressure. Macrophage cells generate NO as an immune defence system to destroy microorganisms and pathogens.
In our brain, after a certain age and under certain pathological conditions, excessive NO is produced, causing tissue damage and oxidative stress. It also reacts with other free radicals to create specific molecular modifications. The overproduction of NO, especially by nNOS (in brain) is implicated in various diseases states such as neurodegeneration, stroke, migraine and chronic headache, Parkinson, Alzheimer, and Huntington diseases, tissue damage, hypotensive crises during septic shock, colitis, arthritis, and various kinds of inflammatory diseases. For this reason, it is important to inhibit nNOS selectively in the brain. Three isozymes show extraordinarily structural similarities hindering the selective inhibitor design. In previous literature there are many outstanding studies, however there has not yet been any drug developed which accomplishes the required affinity and selectivity. In this research, computer-modelling studies were used based on the known crystal structure of three NOS isozymes. The selected scaffolds were used hoping to increase both selectivity and potency toward the nNOS enzyme. Several hundred compounds were screened in *silico* for prioritization of lead candidates. *De novo* design method was used for the modifications of and additions to selected scaffolds within a target-binding site in order to enhance its binding affinity and selectivity to that isozyme. The best candidates showing high activity and selectivity against nNOS over eNOS and iNOS isoforms were determined.



KEY WORDS: Nitric oxide synthase, in silico design, selective nNOS inhibitors, de novo design.

P3.14
NOVEL TOOLS FOR DISEASE MODIFYING ANTI-ALZHEIMER'S DRUGS; hChEs AND b-AMYLOID INHIBITORS

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Conformational flexibility of AChE active site gorge has been a topic of intense research. We have proposed a thorough structural and bioinformatic analysis of the active site gorge of cholinesterases (ChEs), along with the identification of their fluctuations, which already drew the optimisation of our design strategy to discover extremely potent human Acetylcholinesterase and Butyrylcholinesterase (hAChE and hBuChE) bis-tacrine reversible inhibitors. Starting from these AChE and BuChE ligands, a set of potent multiple binding site homo- and hetero-bivalent inhibitors were designed, aiming to selectively interact with specific protein substructures on the surface of the enzymes

around the peripheral anionic site. Accordingly, functionalised linkers differentially spacing two tricyclic moieties were investigated as molecular yardsticks to probe the finest interactions with specific amino acid residues along ChEs gorge (hot spots). On these molecular supports, and aiming at identifying novel Alzheimer's modifying pharmacological tools, we have more recently developed bis-tacrines functionalized with a specific peptide moiety for interference with the hAChE surface sites which bind amyloid-beta (Aβ) and promote aggregation. These new high molecular weight compounds proved to be inhibitors of hAChE catalytic and non-catalytic functions (binding the catalytic and peripheral sites) can interfere with hAChE-induced Aβaggregation, Aβspontaneous aggregation, and with the Aβself-oligomerization process. Molecular modeling studies for these new ligands in complex with TcAChE confirmed the preliminary results obtained by X-ray and will be presented, highlighting how the bis-tacrine systems span the gorge, while the peptide moieties bulge outside the gorge in proximity of the peripheral site, thus explaining observed activity.

KEY WORDS: Cholinesterases, inhibitors, amyloid beta fibrils, amyloid beta oligomers, Alzheimer's disease, anti-Alzheimer's drugs.

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