european chitin society

NEWSLETTER

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No. 39

Message from the President

At the end of sixteenth century a Japanese boat with an embassy of *daimio* Hideyoshi arrived to Seville with some gifts and proposals for an agreement with Philip the Second, king of Spain and Portugal. At the time Spain and Japan were neighbor empires in Philippines and therefore both countries share common, not always friendly interests. The members of the Japanese boat eventually settled in Seville´s surrounding area and one can find there even today some descendants of that crew bearing the family name "Japón" (Spanish for "Japan").

We will meet soon in Seville in the joint EUCHIS-SIAQ Meeting and after that the next one will be held in Japan. Let me think that this exchange of venues Seville-Osaka is a late devolution of that old Hideyoshi embassy but with a different, more peaceful and positive content: scientific and technological exchange.

Angeles Heras Caballero.

EUCHIS President

Editorial

It is our pleasure to distribute another Newsletter just a few days ahead of the 13th EUCHIS Conference, 2017. This year we celebrate the 25th anniversary of EUCHIS. In a brief retrospective, the foundation of EUCHIS is commemorated and some its activities since then are summarized (page 3).

A life cycle assessment of chitosan is of interest not only for producers and agencies. A first, very interesting publication dealing with this issue appeared recently. The abstract is reprinted in this Newsletter (page 5) and reading of the full article is highly recommended to everyone and interested in economic and environmental aspects of chitosan production.

Following earlier calls for submission of abstracts of PhD, just one contribution has been sent to the editorial office (page 6).

The survey of EUCHIS member's publications is continued (page 9), covering the period November 2016 – April 2017. We found 79 citations, including reviews, conference abstracts, articles, and patents, reflecting ~ 1.2 % of ~ 6200 citations in ScienceFinder®, dealing with chitin and/or chitosan between January and April, 2017.

Enjoy reading ! See you all in Sevilla !

Svetlana Bratskaya, Secretary
Martin G. Peter, Assistant Secretary

25 Years EUCHIS

A brief history

The European Chitin Society was founded on March 27, 1992, in Villeurbanne, France, and registered on April 12 in the Prefecture du Rhone, Dossier No. 1/31151. A photo, showing 12 of the 13 members of the foundation assembly, was taken in a coffe break during the constitutional sessions.

Left to right:
Front: Riccardo Muzzarelli (Italy), Maria Terbojevich (Italy), Charles Jeuniaux (Belgium), Alain Domard (France), Marie-Madeleine Giraud-Guille (France), Vassilis Bouriotis (Greece)
Back: Kjell Vårum (Norway), Martin Peter (Germany), Jacek Dutkiewicz (Poland), Gerhard Goffinet (Belgium), George Roberts (United Kingdom), Klaus-Dieter Spindler (Germany)
Not seen in this photo is Henryk Struszczyk (Poland)

The first Board meeting took place during the 1st Conference on Chitin Enzymology, Senigallia, Italy, in May 1993 where Alain Domard was elected as the first president. The first General Assembly was held at the 1st EUCHIS conference in Brest, France, 1995. Alain Domard was reelected for a second period of two years. He was followed by Riccardo Muzzarelli (Italy, 1997 - 1999), Martin Peter (Germany, 1999 - 2004), Kjell Vårum (Norway, 2004 - 2009), and Sevda Senel (Turkey, 2009 - 2013). Since then, the President is Angeles Heras (Spain).

EUCHIS' Honorary President was Charles Jeuniaux (deceased 1999). Olav Smidsrød was awarded the honorary presidency in 2004.

So far, twelve EUCHIS conferences were organized, usually on a two-year schedule, as listed below. Proceedings of nine conferences have been published so far as a series of "Advances in Chitin Science". Thirty eight Newsletters were issued. From Newsletter No. 15 these are available open access on the website of EUCHIS.

EUCHIS Conference	Year	Proceedings
1st: Brest, France	**1995**	Advances, Vol. 1
2nd: Lyon, France (joint with 7th ICCC)	**1997**	Advances, Vol. 2
3rd: Potsdam, Germany	**1999**	Advances, Vol. 4
4th: Senigallia, Italy	**2001**	
5th: Trondheim, Norway	**2002**	Advances, Vol. 6
6th: Poznan, Poland	**2004**	Advances, Vol. 8
7th: Montpellier, France France (joint with 10th ICCC)	**2006**	Advances, Vol. 9
8th: Antalya, Turkey	**2007**	Advances, Vol. 10
9th: San Servolo, Italy	**2009**	Advances, Vol. 11
10th: St. Petersburg, Russia	**2011**	Advances, Vol. 13
11th: Porto, Portugal	**2013**	In preparation
12th: Münster, Germany (joint with 13th ICCC)	**2015**	In preparation
13th: Sevilla, Spain (joint with 8th SIAQ)	**2017**	

In a personal retrospective, 25 years of EUCHIS meant 25 years of professional as well as personal rewards. It was always a great experience to meet colleagues coming from all fields of research and application, to learn about the fascinating properties of chitosan polymers and oligomers, in material sciences, biomedicine, pharmaceutical, agricultural and biochemical aspects. The number of publications is growing at an unforeseen speed, making it nearly impossible to follow new developments just by reading the literature. Thus, EUCHIS is the ideal and a necessary platform to keep up-to-date in chitinology.

Martin Peter

Life Cycle Assessment of Chitosan

Munoz, I., Rodriguez, C., Gillet, D. & Moerschbacher, B. M.
Life cycle assessment of chitosan production in India and Europe.
Int. J. Life Cycle Assess. **2017,** Ahead of Print; DOI 10.1007/s11367-017-1290-2

Purpose: The aim of this article is to present the first life cycle assessment of chitosan prodn. based on data from two real producers located in India and Europe. The goal of the life cycle assessment (LCA) was to understand the main hot spots in the two supply chains, which are substantially different in terms of raw materials and prodn. locations. *Methods*: The LCA is based on consequential modeling principles, whereby allocation is avoided by means of substitution, and market mixes include only flexible, i.e. non-constrained suppliers. The product system is cradle to gate and includes the prodn. of raw materials, namely waste shells from snow crab and shrimp in Canada and India, resp., the processing of these in China and India and the manuf. of chitosan in Europe and India. Primary data for chitin and chitosan prodn. were obtained from the actual producers, whereas raw material acquisition as well as waste management activities were based on literature sources. The effects of indirect land use change (iLUC) were also included. Impact assessment was carried out at midpoint level by means of the recommended methods in the International Life Cycle Data (ILCD) handbook. *Results and discussion*: In the Indian supply chain, the prodn. of chems. (HCl and NaOH) appears as an important hot spot. The use of shrimp shells as raw material affects the market for animal feed, resulting in a credit in many impact indicators, esp. in water use. The use of protein waste as fertilizer is also an important source of greenhouse-gas and ammonia emissions. In the European supply chain, energy use is the key driver for environmental impacts, namely heat prodn. based on coal in China and electricity prodn. in China and Europe. The use of crab shells as raw material avoids the composting process they would be otherwise subject to, leading to a saving in composting emissions, esp. ammonia. In the Indian supply chain, the effect of iLUC is relevant, whereas in the European one, it is negligible. *Conclusions*: Even though we assessed two products from the same family, the results show that they have very different environmental profiles, reflecting their substantially different supply chains in terms of raw material (shrimp shells vs. crab shells), prodn. locations (locally produced vs. a global supply chain involving three continents) and the different applications (general-purpose chitosan vs. chitosan for the medical sector).

Acknowledgement: The authors thank the EU's Seventh Framework Programme for research, technological development and demonstration for its financial assistance through the Nano3bio project (grant agreement number 613931).

Abstract of PhD Thesis

Quantitative mass spectrometry as a powerful tool for analysing chitosan-modifying enzymes and how they define the bioactivities of chitosans

PhD Thesis

Fachbereich Biologie

Stefan Cord-Landwehr

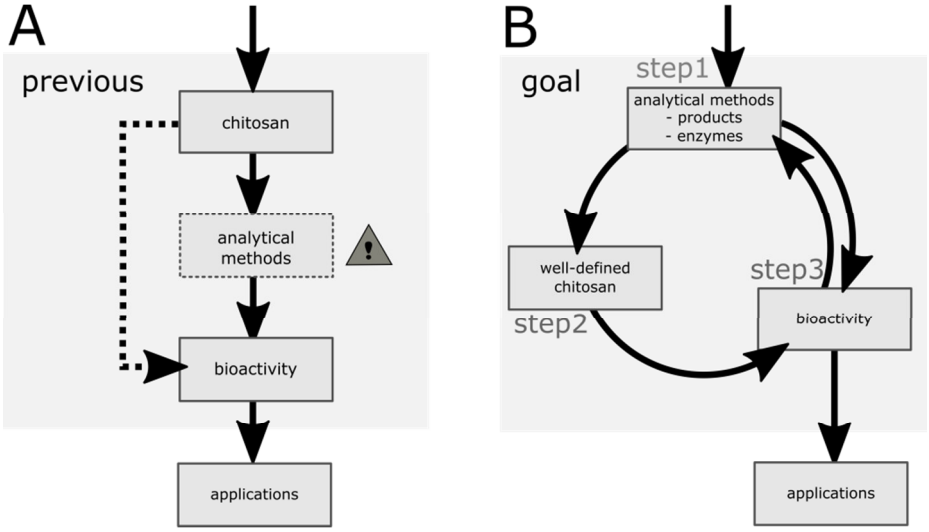
Universität Münster, 2016

Supervisor: Prof. Dr. Bruno Moerschbacher
Co-Supervisor: Prof. Dr. Henk Schols

Abstract

Chitosans are a group of highly promising biomolecules, which consist of β(1-4) linked glucosamine (GlcN; D) and *N*-acetylglucosamine (GlcNAc; A) units. They have a wide variety of biological activities and therefore many potential applications in e.g. agriculture or medicine. It is known that their biological activities depend on the degree of polymerization (DP) and acetylation (DA). Additionally the pattern of acetylation (PA) likely influences these properties. However, the lack of well-defined partially acetylated chitosan oligomers (paCOS) and the time- and material-consuming and sometimes poorly used analytical methods are some of the main obstacles to understanding their structure-function relationship and therefore the application of chitosans (Scheme A).

The studies presented in this doctoral thesis have the goal of improving each part of the process required for understanding and using chitosans, which will thereby reinforce the connections between these parts, allowing us to gain more information about each aspect (Scheme B). To achieve these goals, the first step (Scheme B) is to improve and develop methods to analyse the chitosan products themselves; through this, we can learn more about the chitin- and chitosan-modifying enzymes (CCME) used to produce them. The second step is to produce novel and well-defined chitosans, and then perform in the third step bioactivity tests on these compounds. In turn, this offers information about the natural functions of the initial enzymes used to produce the paCOS products (return to step 1, Scheme B), as well as potential applications for chitosan.



To address the first step (Scheme B), a hydrophilic interaction liquid chromatography-mass spectrometric (HILIC-MS) method developed to analyse plant cell wall carbohydrates was optimised to analyse the fully acetylated chitin, the fully deacetylated and partially acetylated chitosan oligosaccharides within one experiment. Quantitative analysis of paCOS including their DP, DA, and PA was achieved by (I) [$^{2}H_3$]*N*-acetylation of free amino groups in the paCOS, (II) spiking the samples with [$^{13}C_2$; $^{2}H_3$]acetylated chitin oligomers as internal standards, (III) analysing the mixture by HILIC-ESI-MS, and (IV) pattern determination using HILIC-ESI-MS/MS with paCOS containing an ^{18}O label at the reducing end. The main outcome of this quantitative HILIC-ESI-MSn analysis is the fast, efficient and precise determining of subsite and substrate specificity of chitin- and chitosan-modifying enzymes (CCME) while only requiring a small amount of produced oligosaccharides compared to the previous used SEC-NMR method. Using ChiB from *Serratia marcescens*, the best-described chitinase, as benchmark, the newly developed quantitative HILIC-ESI-MS method was verified as having only small variations compared to the results obtained with the common SEC-NMR method. Furthermore, the increased sensitivity of the method enabled the monitoring of the subsite specificity at different time points and for chitosans with different DAs. Additionally the new quantitative analysis HILIC-ESI-MS method allows for investigation of the interplay between the neighbouring subsites (-2 and -1, +1 and +2); in contrast, the SEC-NMR method only allows one subsite isolated from the neighbouring units to be studied.

Subsequently, studies which addressed step 2 in Scheme B, were performed with a novel chitin deacetylase (CDA) derived from the endophytic fungus *Pestalotiopsis* sp. and its products. This enzyme (PesCDA), recombinant produced in *E. coli* as MBP fusion protein, is a regio-selective CDA and generates novel and well-defined paCOS with the pattern $AA[D]_{n-3}A$. To address step 3 (Scheme B) bioactivity analyses on the products of PesCDA were performed, showing that this enzyme converts by deacetylation the elicitor-active chitin oligomer into an elicitor-inactive molecule. This inactivation of chitin oligomers supports a long-standing hypothesis for how chitin-containing fungi overcome detection by the plant cells and therefore avoid evoking the plant's immune system response. As such, this might be the natural function of PesCDA for the endophytic fungus, revealed by performing bioactivity tests with the products of this enzyme. Furthermore, a subsequently detailed analysis of PesCDA subsite specificity allowed knowledge-based combination of PesCDA with other regio-selective CDAs to produce additional well-defined paCOS with new pattern of acetylation, which can be used for bioactivity tests in the future.

The studies shown in this thesis provide several new possibilities to analyse chitin and chitosan oligosaccharides with a focus on quantification to enable a precise characterisation of chitin and chitosan-modifying enzymes and their chitosan products. Based on this, various chitin- and chitosan-modifying enzymes can be characterised, and several well-defined chitosans can be produced which can be used now for bioactivity assays to decipher the structure-function relationships of chitosans. The future studies should not only be oxidative burst studies, as shown for the PesCDA products with rice cell suspension cultures, but should be more significant for the biotechnological usage, such as by testing chitosan's effects in plant promotion or plant strengthening. By interconnecting each part of the chitosan studies, we will be able to obtain more refined knowledge about chitosans and the enzymes used to produce them, which will therefore enable more and more applications.

Publications

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General Assembly, Sevilla, June 2, 2017

Agenda

1. Report of the President
2. Report of the Secretary
3. Financial report 2015 and 2016
4. Member Statistics
5. Election of new Board members
6. Forthcoming Conferences
7. Prix Braconnot
8. Varia

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November 2016 – April 2017

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Patents

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Events

- **EUCHIS/ SIAQ 2017**, Seville, Spain, **May 31st – June 3rd, 2017**
  organized under the patronages of the European Chitin Society (EUCHIS) and the Iberoamerican Chitin Society (SIAQ),
  URL: http://chitin2017.com

- **19th EUROCARB,** Barcelona, Spain, **July 2nd-6th, 2017,** hosted by the Institut Químic de Sarrià, University Ramon Llull, under the auspices of the European Carbohydrate Organization,
  URL: http://www.eurocarb2017.com/

- **5th EPNOE International Polysaccharide Conference**, Jena, Germany, 20–24 August 2017,
  URL: http://www.epnoe2017.de/

- **4th International Cellulose Conference** (ICC2017), Fukuoka, Japan, October 18th – 20th, and **3rd Symposium on Bacterial Nano Cellulose** (BNC2017), October 16th – 17th, 2017,
  URL: http://www.c-linkage.co.jp/icc2017/index.html

- **XXIII Conference on "New Aspects of the Chemistry and Applications of Chitin and its Derivatives"**, Wałbrzych, Zamek Książ, Poland, 20 – 22 September, 2017
  URL: http://www.ptchit.lodz.pl/en344,walbrzych_2017.html

- **14th International Chitin and Chitosan Conference (14th ICCC) & 12th Asia-Pacific Chitin and Chitosan Symposium (12th APCCS), joint with 32nd Japanese Chitin and Chitosan Symposium,** Kansai University (Senriyama Campus), Suita, OSAKA, JAPAN, August 27-30, 2018 (see page 15)

First Circular

14th International Chitin and Chitosan Conference (14th ICCC) & 12th Asia-Pacific Chitin and Chitosan Symposium (12th APCCS) (Joint with 32nd Japanese Chitin and Chitosan Symposium)

Kansai University (Senriyama Campus)
Suita, OSAKA, JAPAN
August 27-30, 2018

SCHEDULE

- Call for papers: October, 2017
- Title and abstract submission by: March, 2018
- Notification of acceptance: April, 2018
- Early bird registration by: May, 2018
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