EUROPEAN *C*HITIN SOCIETY *N*EWSLETTER

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- Editorial
- A word from the President of EUCHIS
- "On the nature of fungi" – M.H. Braconnot
- Prof. Angeles Heras Caballero
- Prof. George Roberts
- Prof. Saburo Minami
- Prof. Huei Chen

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EDITORIAL

Dear Society Members,

I am pleased to present you with the new issue of the European Chitin Society bulletin. Last year we celebrated the 200th anniversary of the discovery of chitin by Henry Braconnot. This discovery was one of several important discoveries that the XIX century brought, one that opened a gate to the scientific world of chitin and its derivatives. This word became an important part of life for many scientists, joining us together in a "chitin" family.

To celebrate this Anniversary, we invited well known scientists to contribute in this issue of ChitinNewsLetter with their impressions on the importance of H. Braconnot's discovery. You will find also an abstract of original H.Braconnot work on fungal chitin prepared by Prof. Jacques Desbrieres. Original paper (we got it thanks to Prof. George Roberts) was extremely long (39 pages) and was written in French, so we decided to prepare a short version of the text, and focus only on the description of chitin or "fungine" as Braconnot named this substance. Everybody who would like to get an original paper may contact me or Prof. J. Desbrieres.

At the end of these editorial remarks I would like to apologise to Prof. Angeles Heras Caballero that I gave her name incorrectly in a previous issue of ChitinNewsLetter.
We had also some changes in the numbering of volumes of Advances in Chitin Science. The last one (edited after the conference in St. Petersburg) is VOLUME XIII, ISBN 978-5-4253-0133-8. (http://ecs-11.chitin.ru/Editions.htm), Proceedings of the 10-th International Conference of the European Chitin Society, SAINT-PETERSBURG, RUSSIA, 2011.

Malgorzata M. Jaworska
Secretary

Message from the President

Dear members of EUCHIS family,

After a long break, we are back again with this issue dedicated to H. Braconnot who is the real pioneer of the chitin world. It was indeed a great pleasure to celebrate the 200th anniversary of chitin's discovery with our members at the EUCHIS'13 meeting which was held in St. Petersburg during May 20-24, 2011.

Herewith, I take the opportunity to thank to those who have contributed to this issue.

I want to inform you that the preparations of our coming EUCHIS meeting which will be held between 5-8 May 2013 in Porto, Portugal, is going quite well , and we look forward to receiving your contributions both before the meeting and during the meeting to achieve a successful meeting. Please do not hesitate to get back to me or any of our board members with your suggestions to make EUCHIS'13 more attractive and fruitful.

Wishing you all the best in good health and peace,

Sevda Şenel

„Laboratory in Nancy" by Olga Boroday,
(canvas, oil, 50 x 70, 2011)

thanks to Prof. Valery Varlamov, Center "Bioengineering" RAS

RECHERCHES ANALYTIQUES

Sur la nature des Champignons ;

PAR M. HENRI BRACONNOT,

Professeur d'Histoire naturelle, directeur du Jardin des Plantes, et membre de l'Académie des sciences de Nancy.

(Lu à la Société des sciences, lettres, agriculture et arts, de Nancy, le 15 novembre 1810).

La famille des champignons renferme, comme on le sait, une quantité prodigieuse d'espèces fort différentes des autres productions vivantes, et remarquables par la singularité de leurs formes, et par la consistance tendre et charnue dont la plupart sont douées.

Ces êtres qui semblent évidemment former le dernier chaînon qui unit le règne végétal au règne animal, ont vivement excité l'attention des botanistes modernes, et on

doit à leur sagacité la méthode claire et précise qu'ils ont su répandre dans leurs classifications : mais si cette étude a fait de grands progrès dans ces derniers tems, la chimie n'avait encore rien appris de bien satisfaisant sur la nature intime des espèces qui composent cette branche intéressante de l'histoire naturelle. On doit cependant aux travaux de M. Bouillon-Lagrange l'analyse de trois espèces vivaces : le *Boletus laricis*, le *Boletus igniarius*, et le *Lycoperdon tuber*. MM. Fourcroy et Vauquelin ont aussi fait connaître la nature de l'*Uredo segetum*, ou la nielle du froment, tels sont les seules espèces qui ont été examinées jusqu'ici avec soin.

Je me propose dans ce Mémoire d'exposer l'analyse des champignons annuels comme étant les plus nombreux, et les seuls destinés par la nature à la nourriture de l'homme et des animaux ; mais avant de passer à cet examen des espèces en particulier, je vais faire connaître la nature de la substance qui forme le corps ou la base charnue insoluble du champignon, et que je désignerai sous le nom de fungine.

De la Fungine.

Je propose de nommer ainsi cette substance tendre et charnue des champignons, parce qu'elle me paraît d'une nature particulière ; d'ailleurs son seul aspect, ses propriétés externes, son abondance dans une famille nombreuse de corps organisés, et l'emploi qu'on en fait comme aliment, me semblent assez justifier cette dénomination pour la distinguer des autres matériaux immédiats des plantes. Cette substance de quelques champignons qu'elle provienne, séparée de tout corps étranger par l'eau bouillante aiguisée d'un peu d'alcali, est, plus ou moins blanche, molasse, fade, insipide, peu élastique en la comparant au gluten, et se divisant assez bien entre les dents ; dans cet état il n'est pas douteux qu'elle ne puisse servir avec beaucoup d'avantage, et dans plusieurs circonstances, à la nourriture de l'homme, malgré l'assertion de plusieurs médecins, qui regardent généralement les champignons de quelque manière qu'on les prépare, comme mal-sains et peu nourrissans ; assertion complettement démentie par les habitans des campagnes qui dans certains

"ON THE NATURE OF FUNGI"

M. Henri Braconnot

Annales de Chimie, Tome LXXIX, pp. 265-304 (1810)

Abstract by Prof. Jaques Desbrieres, Université de PAU et des Pays de l'Adour (France)

Chitin was discovered in 1810 by Henri Braconnot who has isolated an impure residue from cellular walls of fungi. He named "fungine" this substance which composes the body or the insoluble plump base of the fungus.
In this paper he presented different experiments to characterize the nature of this substance. From its separation using boiling water added with alkali a whitish, flabby, tasteless and a slightly elastic substance comparable to gluten was obtained. Mirbel has carried out microscope observations and he observed that fungous substance was constituted by a continuous cellular tissue, which explains why it is very water permeable and soaked as a sponge.

The roasted fungine burns and gives off a toasted bread odor. If it was exposed to the flame of a candle, it went up in flames with swiftness and a white ash was obtained.
Braconnot presented a very accurate analysis of the different components of this substance. When 38 grams of dried fungine were distilled in a glass retort, melting occurs at the beginning and then 19 grams were obtained: 8 grams of an empyreumatical brown thick oil and 11 grams of an aqueous amber liquor. This latter was composed of "sub ammonium acetate" soiled with a few oil. As a consequence fungine seems to contain more hydrogen and nitrogen than wood. 10 grams of coal stayed in the retort which inner surface was embedded of a metallic shiny coating with strong adhesive properties. These are reduced in 5 grams of ash blended with sand grains when they were burnt in a capsule. This ash was dissolved by nitric acid without a strong effervescence, producing a sulfur hydrogen odor. Insoluble residue weighted 1.7 g and was composed of quartz sand. From analysis tests (reaction with different chemicals such as potassium nitrate, ammoniac..) it was concluded that the ash was made up of lime phosphate.

Further investigation was carried out using alkali or acids. Alkali has a very slight action on fungine and its behavior was different from woody materials. However if a concentrated solution of this substance in potassium hydroxide was boiled, fungine partially "melts" and a soapy liquor was obtained from which acids separated in a fleecy deposit.
Finally 30 grams of dried fungine were distilled with 188 g of 29° nitric acid. First fungine turned yellow, softened and considerably swelled; the reaction was sudden. Prussic acid and calcium oxalate were found.

Fungine in water was spontaneously decomposed. Fade gluten odor was in a first time smelt and then animal matter in decomposition. Supernatant, after three months, does not contain any free acid nor ammoniac but a viscous part was precipitated by lead acetate. This viscous matter had properties of mucus. This decomposing fungine became black as lead sulphide when lead oxide was added. As a consequence, it seemed that sulfur was one of the major components of fungine, which will be denied later.

A new Age was growing

Christian Jürgensen Thomsen (1788-1865) was a Danish archaeologist whose major contribution was the division of the history of mankind based on the technology and use of some materials: the names Stone Age, Bronze Age and Iron Age are now universal and quite familiar chronological references.

Curiously enough he had an exact contemporary, named Henry Braconnot (1780-1865) who first put in the market a new material which deserves the honour of naming a new age, the Plastic Age, as it represents very accurately our industrial times.

Chitin effectively was found in mushrooms as early as 1811, as it is well known, and it is the earliest known polysaccharide, cellulose being found in 1838; a few years later Braconnot produced xyloïdine, combining nitric acid with starch or wood fibers, paving thus the way to the plastic explosives, so unfortunately popular and widespread in our not so peaceful times.

Chitin, the earliest (bio)polymer and its derivatives stand thus at the very threshold of modern technology and apparently (and hopefully) they have not yet developed all their possibilities: it is up to us, subscribers and readers of *Chitin Newsletter* to go on exploring and unthreading them, two hundred years after they were born claiming a place among other more traditional materials.

One should refrain from asserting that a research topic has been so thoroughly explored that it does not need any further studies, as new approaches are always emerging and they cast new light on unknown or neglected parts of the old topic.

In the case of chitin one of the new approaches is obviously the nano-approach which is opening in front of our eyes a fascinating and unknown landscape. Nevertheless research on chitin has been already living long with us so as we can say that it is ripe enough to endure the test of applicability. Imitating Karl Marx we can say that we have been trying to explain chitin for too long and now it is time to make a use of it. Having explored chitin along the XIX and XX centuries, let it be employed in the XXI century to improve our food, medicines, clothes or cosmetics.

The undersigned is quite aware that this unduly fortunate polynomial R+D+I, that stands for “research, development and innovation” is neither a divine commandment nor a epistemological finding, but rather a clumsy model trying to explain the complexities of producing new knowledge, and using it; yes, this is a well known debate in research policy, but could not at least some of us, chitin lovers or pretenders, put now our main focus on the application of this two centuries old biopolymer, I mean, change the emphasis from the R to the D?

Taking this not so new way we just would be following in Muzzarelli´s, Domard ‘s, Roberts´s, Peter´s and other respected colleagues´ footsteps.

Angeles Heras Caballero
Universidad Complutense de Madrid (Spain)

The importance of Braconnot's discovery

Last year we celebrated the 200th anniversary of the discovery of chitin by Henri Braconnot. While cotton and flax had been cultivated for use as fibres from early on in the history of mankind, the history of the chemistry of cellulose only began around 1837-42 with the studies of Anselme Payen and others, including Braconnot, hence chitin was discovered almost 30 years before its close and more widely known relative, cellulose. Despite being discovered later cellulose has been an established industrial material for almost 150 years, its commercial exploitation starting very soon after its discovery; for example cellulose nitrate was being produced commercially by the 1860s and cellulose triacetate before the beginning of the 20th century. So why did the "younger sibling", cellulose, become so dominant while the earlier-discovered chitin lapsed into chemical obscurity for about 150 years?

Among the numerous possible contributing factors are the following:

1. Cellulose occurs in more convenient quantities – you have to peel an awful lot of prawns to get an equivalent amount of chitin to the cellulose you get from chopping down one tree.

2. Cellulose is more easily isolated from other components present in the source material than is chitin.

3. Purified cellulose ("α-cellulose") is a homopolymer with only a single monomer residue along the chain whereas purified chitin and chitosan are very frequently co-polymers of the two monomer residues.

4. Cellulose is more easily derivatised than are chitin and chitosan due to the greater inter-chain hydrogen bonding systems of the latter two polymers.

The first two factors are major contributors to the higher cost of chitin and chitosan compared with cellulose, while the second two must be major factors in hindering the development of industrial applications of chitosan in particular. It is difficult to develop an industrial process where there is the potential of batch-to-batch variation in what could be an important structural factor of one of the main starting materials.

So while there is a wide range of cellulose-based products in the market place – cellulose organic and inorganic esters, cellulose ethers, regenerated cellulose in the form of fibres and film – the same is not true for chitin and chitosan. Obviously there are commercial applications of chitin and chitosan but they are limited in number. The biggest use of chitin is as a feed stock for the production of **D**-glucosamine which is sold as a food supplement, frequently in admixture with chondroitin sulphate, and that for chitosan is as a cholesterol-reducing agent under trade names such as "Fat Busters" or "Fat Magnets". Another area of commercial activity is the use of chitosan in wound dressings. Perhaps the haemostatic wound dressings produced by HemCon Medical Technologies Inc. are the most successful of these products to date, both in terms of income generation and public awareness; newspaper headlines such as "Shrimps help heal wounds" are fairly sure to have an impact on the public consciousness.

Although a wide range of chitin and chitosan derivatives have been reported in the scientific literature, very few appear to have made it through to commercial exploitation, despite the considerable number of patents involving chitosan that are filed each year. One such success, if only for a limited time, was the use of hydroxypropyl chitosan in a range of hair products marketed by Wella AG. However it can be argued that apart from the occasional paper and even more occasional patent, most research on chitin and chitosan was carried out initially in the areas of biology and enzymology; the upsurge in chemical research only beginning about 35 years ago when it was kick-started by the 1^{st} ICCC in Boston in 1977. So chemical research on these polymers is still in its infancy, and it is possible that the next 35 years will see a large increase in the number and volume of chitin and chitosan derivatives produced commercially.

So if Braconnot's discovery cannot be said to have led to a chitin-based industry, at least up to the present time, how important was it? It has given scientists another polysaccharide to study, or it would be more correct to say a new family of polysaccharides in which the variability in chain composition makes them a more difficult and interesting topic of study than cellulose. Indeed the study of chitin and chitosan presents intellectual challenges not present in other polysaccharides with the possible exception of the alginates. Another appeal of chitin and chitosan lies the variety of research disciplines where chitin and chitosan have and are being studied. This is evident from the wide range of interests covered by delegates to any chitin/chitosan conference, either international or national, and makes such conferences very valuable for developing and broadening one's ideas.

Whatever the fairly limited importance of Braconnot's discovery in commercial terms may currently be, from my personal viewpoint it was an extremely important discovery. Chitin has given me a research interest for many years, taken me to many countries that I would otherwise most probably never visited and more importantly, it has led to many lasting friendships with colleagues around the world. I am sure that many of you would agree and it is for these reasons that I classify Braconnot's discovery as very important indeed.

George Roberts
UK

Chitin Science World: "Braconnot's discovery is a world treasure"

In 1811, Henri Braconnot discovered chitin in mushrooms in the laboratory of the Botanique Garden Saint Catherine, Nancy, France. The discovered chitin was a high-molecular-weight polymer with a high degree of crystallinity due to strong hydrogen bonds, particularly observed in alpha-type chitin. These properties make chitin insoluble by ordinary organic solvents and inhibit its application for industrial, pharmacological, and medical usage. Prudden et al.—a team of American medical doctors—discovered the potent ability of chitin to accelerate wound healing in 1977, 166 years after Braconnot discovery. Subsequently, many scientists, engineers, and medical doctors attempted to implement this discovery in their respective fields; however, its utility in medical remedies was limited by its insolubility. In 1985, 8 years after Prudden et al.'s discovery and 174 years after Braconnot's discovery, Unitika Co. Ltd (Kyoto, Japan) developed a pure chitin non-woven fabric as the world's first medical artificial skin, named "Beschitin W." On August 28, 1990, a cross-border rescue drama between Russia and Japan occurred. A 3-year-old Russian boy named Konstantin (Константин Игоревич Скоропышный), who lived in Sakhalin, was severely burned over 90% of his body after falling into boiling water. At that time, Sakhalin and Japan were strictly divided by the Iron Curtain of the Cold War. However, the heads of each country's ministry of foreign affairs obtained their government's permission to transport the boy from Yuzhno-Sakhalinsk Children's Hospital (Sakhalin) to Sapporo Medical University Hospital (Hokkaido, Japan) for skin transplantation. Human skin was collected from Hospitals in Tokyo, but was insufficient to completely cover the boy's lost skin; therefore, the medical team utilized the Beschitin W artificial skin. The surgery was successful and the boy lived. At age 15, he returned to Japan to attend the 16th Japan Gate Ball Cup held in Memuro city, Hokkaido, as a member of the Sakhalin team; at that time, medical doctors confirmed the health of his skin. Several processes are involved in the manufacture of chitin non-woven fabric. In Japan, Ifuku et al. developed a simple method for developing chitin nano-fibril from chitin powder using a grinding machine (Super Masscolloider, Masuko Sangyo Co., LTD, Japan); this chitin nano-fibril retained the pure and natural form, and could easily disperse in any mild acid solution. Therefore, the chitin nano-fibril can be effectively molded and developed by scientists into various suitable shapes. I believe that the chitin nanofibril will pioneer a revolution in chitin science for applications in engineering, agriculture, and medicine. It is now 201 years since Braconnot's discovery and we, the family of chitin scientists, should gather our knowledge and ideas to develop materials that will aid in improving the health of humans, animals, and the environment.

I hope that the 13th International Conference on chitin and chitosan will be held at the botanical garden in Nancy with the 12th EUCHIS conference.

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Saburo Minami, DVM, Ph.D,
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In the memory of 200th Anniversary of discovery of chitin by H. Braconnot What we have learn and what we have to do

Chitin is the most abundant natural amino polysaccharide. Chitin is a high molecular weight polymer made of the repeating sugar N-Acetyl-D- glucosamine (GlcNAc). Chitin and its derivatives have many properties that make them attractive for a wide variety of applications, from foods, nutriceuticals and cosmetics to biomedicine, agriculture and the environment. Their antibacterial, anti-fungal, anti-viral properties and biocompatibility make them particularly useful for biomedical applications, such as wound dressings, surgical sutures, replacements for bone, veins, cartilage, arteries and periodontal disease treatment. Chitin's biodegradable and antifungal properties are a plus for environmental and agricultural uses. Chitin possess splendid sorption ability of transitive and especially heavy metals such as copper, zinc, nickel, cobalt, vanadium, titanium, antimony (stibium), ruthenium, strontium; and selectiveness (the ability to separate (divide) some metals: ferrum and copper, nickel and ferrum, cadmium and nickel). However, chitin is insoluble in most common solvents thus it is most often converted into derivatives or chitosan, both of which can be dissolved in either water or aqueous acids. Major uses of chitin are the production of chitosan and glucosamine.

Chitosan is a cationic (positively charged) polymer consisting units of the amino sugar D-Glucosamine. Many of its uses in cosmetics, water purification, waste recovery, food, medicine, and agriculture. The chitosan has the unique ability to attach itself to lipids or fats and bile acids form a indigestible micelle mass which the body cannot absorb. This large mass is then eliminated from the body. There are no calories since chitosan is not digestible. Chitosan is currently used in cosmetics in a variety of creams and lotions as a moisturizing agent and shampoos and hair conditioning products. Chitosan is targeted in a number of biomedical applications, including drug delivery. These applications will require a very high purity product made under strict FDA guidelines.

Glucosamine (GlcN) is usually produced by depolymerization of chitin with acid such as HCl or by enzymes. Although both GlcN and GlcNAc have been shown to be effective in relieving pain of osteoarthritis, GlcN appears to be more effective. Glucosamine is the No. 1 dietary supplement sold in the US. Sales of glucosamine are $392.4 million in 2002.

The production of chitin and chitosan from crustacean shells, a by-products of shrimp industry, is economically feasible, especially if it includes the recovery of carotenoids. Global chitin derivatives market to reach US$63 billion by the year 2015 and global chitin market to exceed 51.4 thousand metric tons by 2012 according to a report by Global Industry Analysts, Inc. Key factors driving market growth include increasing applications and surging demand for chitin and chitosan from agrochemicals and healthcare/medical end-use sectors.

The thorny issues faces chitin chitosan industry and people involved is to lobby and to convince the regulatory agents such as US FDA and EU Medicine Agency (EMEA) that chitin and their derivatives are the gifts of nature and are safety to be used as nutritional supplement and as a drug delivery system or medical aids and to pass the bill for legal uses of chitn and chitosan in biomedical and nutriceutical industries. For people involved have to define what is the chitin chitosan product in term of Mw and its distribution, DD and its distribution because they affect the functional properties mentioned above.

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