



BIOREFINERY FOR FOOD & FUEL & MATERIALS

Book of Abstracts

7-10 April 2013 Wageningen | The Netherlands

Ton van Boxtel and Marieke Bruins



Symposium Biorefinery for Food, Fuel and Materials 2013

Wageningen UR

Chairs:

Johan Sanders

René Wijffels

Michael O'Donohue

7 - 10 April 2013

Wageningen

The Netherlands

Symposium Biorefinery for Food, Fuel and Materials 2013

Wageningen UR

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Hof van Wageningen, Wageningen

The Netherlands

Editors: Marieke Bruins and Ton van Boxtel

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Welcome

The *Biorefinery for Food, Fuel and Materials 2013* symposium addresses the challenge of how we can use biomass from a variety of sources in an efficient and economical way. This will become possible once we are able to separate individual components from biomass in such a way as to allow them to be used in the most economical or ecological manner. In a Biobased Economy, fossil resources, which are currently used for the production of energy (including liquid fuels for transport) and chemicals, are replaced by biomass raw materials, with substitution levels being as high as 30%. To achieve this goal within the foreseeable future using currently available knowledge and technologies it is anticipated that enormous amounts of biomass will be required, thus necessitating the mobilization of significant amounts of land. Therefore, the prospect of a full blown bioeconomy is increasingly a source of concern with regard to the security of food supply.

Nevertheless, considering that the production of the 2500 kcal of digestible energy (the average per capita daily food consumption) requires 25 000 kcal of biomass and 25 000 kcal of fossil energy, there is a lot of room for improvement, increasing the efficiency of biomass resource use and diminishing energy expenditure.

In the past, the incentive to develop resource and energy efficient processes was not a priority in Europe, basically because both agricultural raw materials and energy were plentiful and quite cheap. However, this will increasingly not be the case and thus the prospect of raw material scarcity and soaring energy prices are providing impetus to develop new processes and approaches, in particular biorefineries. The biorefinery concept describes a group of integrated unit operations that can convert biomass into a wide variety of products, thus contributing to the optimal use of biomass in a world where more people will need to share the Earth's finite resources, simultaneously deriving both food and non-food products from the same biomass raw materials. This will be possible through the tight integration of the food and non-food sectors, a strategy that will ensure reduced waste and lead to closed loops. Accordingly, through the future deployment of appropriate technologies, non-food crops will provide feed for animals and perhaps food ingredients, while food crop co-products and food chain wastes will form the raw materials for non-food products, thus ensuring food security for the world's population.

Johan Sanders, Wageningen University

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AlgoSource Technologies, France

Natuurlijk vermogen.

Groene grondstof als motor van onze economie.

Essent wil samen met industrieën, overheden en kennisinstellingen een biobased economy realiseren. Dit is een economie waarin fossiele grondstoffen worden vervangen door groene grondstoffen bij de productie van o.a. medicijnen, plastics, veevoer, papier en energie.



HOUT



ALGEN



MEEST



BERENDE PLANTEN



AFVAL

Biomassa second generation

Medicijnen
vetten, farmacon

Veevoer
eiwitten, mineralen
vitamines

Papier
vezels, cellulose



Cosmetica
vitamines, vetten
geur- en kleurstoffen

Plastics
suikers, zetmeel

Biobrandstof
koolhydraten, olieën

Elektriciteit lignine

In de biobased economy van Essent worden alle elementen van groene grondstoffen optimaal gebruikt, zodat er een nieuwe economie ontstaat die duurzaam is en economisch rendabel. Ons natuurlijk vermogen is oneindig, laten we het samen optimaal benutten.



Weer een stapje
dichter bij écht duurzaam.

Essent levert.

-essent

General Information

Abstracts

This book contains the abstracts of keynote speakers, followed by the abstracts of oral presentations, and finally the poster abstracts. The posters are arranged in alphabetical order of the first author. The organising committee does not take any responsibility for scientific or typographical errors.

Oral presentations

Plenary presentations will be held in the Ir. Haak room.

The sessions will take place in the Ir. Haak room and the Dorskamp room

Posters

The posters are enlisted alphabetically in this book. They will be on display throughout the congress and can be found in the Wolfswaard room

Excursions and Greenlab Meeting

Registration on site is possible

1. AlgaePARC: Exploring scale-up challenges in microalgae mass production.

During the visit you will be shown the latest state-of-the-art algae research facility; AlgaePARC. This unique Wageningen UR centre is set-up to bridge the gap between fundamental algae research and full-scale algae production. In the fully automated processes in the different photobioreactors are compared to develop a more efficient system and optimize operational concepts. A five year research program started at AlgaePARC has been started and is presently supported by 19 companies in the food, oil, chemical and technology development sectors.

Costs: €50 p.p. (including transport)

Time: 14.00 hrs – 16.00 hrs, Wageningen

2. ACRRES: ACRRES wants energetic cooperation with you!

ACRRES focusses on sustainable energy and green raw materials. It centres on different regional chain development, biofermentation, windfarms, biodiesel production; with design and development of regional energy concepts with the focus on the efficient connection of circuits. And also optimisation of biomass use for the Biobased Economy.

Costs: €50 p.p. (including transport)

GreenLab International Matchmaking Event

As part of the BFF2013 symposium, participants can join the Greenlab International Matchmaking Event on April 10. The 'Greenlab International' meetings facilitate matchmaking on cross sectoral topics with different interesting players in the biobased economy, to give more insight into market opportunities for biobased products, to identify and invite any missing partners and suppliers and to create promising business cases, partnerships and project ideas focussing on e.g. Horizon 2020 topics. On April 11 excursions to two interesting companies are scheduled.

Costs: €100.

Time: following the BFF programme on April 10, Wageningen

Lunches and Symposium Dinner

During the symposium lunch will be served in the Terras room.

On Tuesday April 9th the symposium dinner will be served in the restaurant of Hotel de Wereld.

Sponsors

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Programme

Programme

* all posters will be displayed in the Wolfswaard room

Sunday, April 7th 2012

18.00 – 20.00 *Registration and drinks*

Monday, April 8th 2012

08.30 – 09.00 *Registration*

Plenary opening of the symposium - Ir. Haak room

09.00 – 09.10 *Opening*
Johan Sanders, Wageningen UR

09.10 – 09.20 *Word of welcome*
Aalt Dijkhuizen, Wageningen UR

09.20 – 10.00 *The Circular Economy*
Herman Wijffels, University of Utrecht

10.00 – 10.45 *From Food versus Fuel to Food and Fuel (plus Materials)*
Marcel Wubbolts, DSM

10.45 – 11.15 *Coffee break*

11.15 – 14.00 Session 1: Redesigning the business concept and sustainability
Ir. Haak room Chair: Michael O'Donohue, INRA

11.15 – 11.45 Session keynote – Roland Essel, Nova-Institut GmbH
Food-crops for Industry? Background information and surprising answers

11.45 – 12.05 *Eric van den Heuvel, Argos*
How to interweave food, feed, fuel and fibre to an integral business chain

12.05 – 12.25 *Saicha Gerbinet, Université de Liège*
Land use change: an essential aspect of Life Cycle Analysis of lignocellulosic biomass conversion processes

12.25 – 12.45 *Jan Broeze, Wageningen UR*
Demand orientation of sustainable innovations on food, fuels and materials

12.45 – 14.00 *Lunch and poster browsing – Terras room and Wolfswaard room*

11.15 – 14.00 Session 2: Integration of chains*Dorskamp room Chair: Ludo Diels, VITO***11.15 – 11.45 Session keynote – Ludo Diels, VITO**

Process Intensification in Biotechnology: towards an Integrated Biorefinery

11.45 – 12.05 *Marinella Tsakalova, National Technical University of Athens*

On the systematic synthesis screening and integration of real-life biorefineries

12.05 – 12.25 *Annemarije Kooijman, University of Twente*

Assessment of coinciding and diverging requirements on the biorefinery system design following from new connections in biorefinery value chains

12.25 – 12.45 *Jeroen Tideman, Bioclear B.V.*

Towards on-shore aquaculture in the Eemshaven

12.45 – 14.00 Lunch and poster browsing – *Terras room and Wolfswaard room***14.00 – 17.00 Session 3: Biochemical conversion and separation I***Ir. Haak room Chair: Jean Francois Jenck, AlgoSource Technologies***14.00 – 14.30 Session keynote – Robert Bakker, Wageningen UR**

Developing Pretreatment and Fractionation for Integrated Biorefineries

14.30 – 14.50 *Joachim Venus, ATB Potsdam*

Pilot plant for the scale-up of continuous mode lactic acid fermentation

14.50 – 15.10 *Mieke van Eerten-Jansen, Wageningen UR*

Bioelectrochemical production of caproate and caprylate from acetate

15.10 – 15.30 *Jue Wang, Wageningen UR*

New driving forces for dry fractionation

15.30 – 16.00 Coffee break

16.00 – 16.20 *Edgar Suarez Garcia, Bodec Process Technology*

Bodec's valorization approach: The Primordial Soup Concept

16.20 – 16.40 *Ana Lopez Contreras, Wageningen UR*

Seaweed biorefinery: production of fuels and chemicals from native North Sea seaweed species

16.40 – 17.00 *Sebastien Jubeau, AlgoSource Technologies*

Biorefinery concept applied to microalgae

14.00 – 17.00 Session 4: Chemicals, Fuels and Materials I

Dorskamp room Chair.: Axel Kraft, Fraunhofer UMSICHT

14.00 – 14.30 Session keynote – Axel Kraft, Fraunhofer UMSICHT

Alternatives to fossil jetfuel and related challenges

14.30 – 14.50 *Janneke Krooneman, University of Groningen*

BioSyn: Production of green chemicals from industrial waste streams

14.50 – 15.10 *Corneels Schabort, North-West University*

Food, fuel and bio-plastics from Sweetstem Sorghum

15.10 – 15.30 *Theresa Cesário, IBB - Instituto Superior Técnico*

Up-grading of wheat straw lignocellulosic hydrolysates to bioplastics

15.30 – 16.00 Coffee break

16.00 – 16.20 *Klaas Breitzkreuz, Fraunhofer*

New pathway to fuels or fuel additives starting from short-chain alcohols

16.20 – 16.40 *Chuan Wang, Institute of Chemical and Engineering Sciences*

Dehydration of lactic acid and 3-hydroxypropionic acid to acrylic acid

16.40 – 17.00 *Kars Gökhan, Selçuk University*

Aminolevulinic acid and biohydrogen production from agro-industrial by-products in a biorefinery concept

Tuesday, April 9th, 2013

- 09.00 – 09.45 Keynote – New Perspective for Biorefinery of Cereals
Ir. Haak room Joël Abécassis, INRA
- 09.45 – 10.15 Potentialities of Microalgae in a Context of Environmental Biorefineries
Ir. Haak room Jean-Philippe Steyer, Greenstars
- 10.15 – 10.45 Coffee break
- 10.45 – 14.00 Session 5: Biochemical conversion and separation II**
Ir. Haak room Chair: Elinor Scott, Wageningen UR
- 10.45 – 11.15 Session keynote - Elinor Scott, Wageningen UR**
Integration of chemistry into agro-food chains to achieve a win-win situation
- 11.15 – 11.35 *Rebecca Pfützenreuter, RWTH Aachen University*
Biogenic Isosorbide tert-Butyl Ethers (ITBE)-Continuous Production and Reaction Kinetics
- 11.35 – 11.55 *Nikolaos Mavroudis, Northumbria University*
Ion exclusion chromatography a potentially valuable tool for fractionation of complex fluids: Juice de-acidification as a case study
- 11.55 – 12.15 *Dries Vandamme, KU Leuven Kulak*
Design and evaluation of flocculation based harvesting processes for microalgae biomass production
- 12.15 – 12.35 *Sandra Hinz, Dyadic Netherlands*
Functional analysis of the Myceliophthora thermophila C1 xylanase machinery
- 12.35 – 14.00 Lunch and poster browsing – *Terras room and Wolfswaard room*

- 10.45 – 14.00** **Session 6: Multi-resource and multi-product biorefineries**
Dorskamp room *Chair: Joël Abécassis, INRA*
- 10.45 – 11.15** **Session keynote – Sara Giarola, Imperial College London**
 A supply chain approach to biorefinery sustainability assessment
- 11.15 – 11.35 *Jaap van Hal, ECN*
 Fractionation of sustainable biomass from land and sea. Technology development at ECN
- 11.35 – 11.55 *Michel Eppink, Wageningen UR*
 Biorefinery of Microalgae: production of high value products, bulk chemicals and biofuels
- 11.55 – 12.15 *Christophe Luguel, IAR Cluster*
 Open innovation platforms on sugars, fatty acids and proteins: developing the high value products biorefineries
- 12.15 – 12.35 *Heleen de Wever, VITO*
 Biorefinery concepts from earth to space
- 12.35 – 14.00 Lunch and poster browsing – *Terras room and Wolfswaard room*
- 14.00 – 17.00** **Session 7: New food and feed ingredients from biorefinery**
Ir. Haak room *Chair: Lilia Ahrné, SIK*
- 14.00 – 14.30** **Session keynote – Atze Jan van der Goot, Wageningen UR**
 Biorefinery concepts aimed at healthy and sustainable food products
- 14.30 – 14.50 *Pascalie Pelgrom, Wageningen UR*
 Dry fractionation for production of functional pea protein concentrates
- 14.50 – 15.10 *Arwa Mustafa, Lund University*
 Bioactive food ingredients from agricultural by-products using green technologies
- 15:10 – 16:00 Short intermezzo with drinks
- 16.00 – 16.20 *Bart Smit, NIZO Food Research*
 RubisCo: from Refinery to product Functionality
- 16.20 – 16.40 *Ai Ling Ho, University of Reading*
 Autohydrolysis processing of oil palm by-product: xylooligosaccharides production and prebiotic potential
- 16.40 – 17.00 *Luc Marchal, Nantes University*
 Algorefinery of microalgal biomass - mild treatments integration for pigments and proteins fractionation from *Chlorella vulgaris*
- Evening **Symposium dinner – Hotel de Wereld**

- 14.00 – 17.00 Session 8: Chemicals, Fuels and Materials II**
Dorskamp room Chair: Hong Qi, Alberta Agriculture and Rural Development
- 14.00 – 14.30 Session keynote – Hong Qi, Alberta Agriculture and Rural Development**
 Growing Bioeconomy - Alberta Activities and Capacities
- 14.30 – 14.50 *Peter Flippo, Arizona Chemical*
 How to develop a Profitable and Sustainable Biorefinery? - Success by maximizing value and being resource efficient
- 14.50 – 15.10 *Johan van Groenestijn, TNO*
 Biomass pretreatment using superheated steam as a starting point for biorefining
- 15:10 – 16:00 Short intermezzo with drinks
- 16.00 – 16.20 *Ischa Lamot, Wageningen UR*
 3-Hydroxybutyrate production by genetically engineered *escherichia coli*
- 16.20 – 16.40 *Chahinaz Aouf, INRA*
 The use of biosourced phenolic compounds as a sustainable alternative to bisphenol A in the field of materials
- 16.40 – 17.00 *Benjamin Brehmer, Evonik Industries AG*
 What is in store for bio-based polyamides?
- Evening **Symposium dinner – Hotel de Wereld**

Wednesday, April 10th 2013

09.00 – 09.30 **Keynote - BRIDGE PPP - Biobased and Renewable Industries for Development and Growth in Europe**

Ir. Haak room *Ward Mosmuller, Biobased Industries Consortium*

09.30 – 10.15 **Keynote - Designing Sustainable Biofuel Systems: Using our Heads to Product Food and Fuel from Cellulosic Biomass**

Ir. Haak room *Bruce Dale, Michigan State University*

10.15 – 10.45 Coffee break

10.45 – 12.15 **Session 9: Small scale biorefineries**

Ir. Haak room *Chair: Marieke Bruins, Wageningen UR*

10.45 – 11.15 **Session keynote – Marieke Bruins, Wageningen UR**
Small beats large

11.15 – 11.35 *Göрге Deerberg, Fraunhofer*

A decentral approach for sustainable supply with raw materials and energy -
Fraunhofer Innovation Cluster »Bioenergy«

11.35 – 11.55 *Devrim Murat Yazan, University of Twente*

A comparative supply chain sustainability evaluation of mobile pyrolysis
plants and pyrolysis-based bio-refineries

11.55 – 12.15 *Ellen Slegers, Wageningen UR*

Small versus large scale microalgae production

10.45 – 12.15 Session 10: Valorisation of knowledge and technology

Dorskamp room Chair.: Wim Soetaert, Ghent Univerity

10.45 – 11.15 Session keynote – Wim Soetaert, Ghent Univerity

Bio Base Europe: Open innovation and education center for a sustainable biobased economy

11.15 – 11.35 *Harmen Willemse, NEN*

Standardization: codifying and disseminating state of the art technology and best practices to the market

11.35 – 11.55 *Farid Chemat, SQPOV, INRA-Université d'Avignon*

Green extraction of natural products as tools for biorefinery

11.55 – 12.15 *Hilke Bos-Brouwers, Wageningen UR*

Integrated valorisation of biomass

12.15 – 12.30 Closure of the symposium

Ir. Haak room *René Wijffels, Wageningen UR*

12.30 Lunch – *Terras room*

14.00 Excursions to ACRRES or AlgaePARC, or participation in 'Greenlabs' meetings

Keynote lectures

Ir. Haak room

Monday April 8th

The Circular Economy

Wijffels, Herman

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Since 2009, Herman Wijffels (1942) is professor of Sustainability and Societal Change at the Utrecht Sustainability Institute(USI) of Utrecht University.

Wijffels studied economics at Tilburg University. He worked at the Dutch Ministry of Agriculture & Fishery and subsequently as secretary general of the Dutch Christian Employers Association (NCW). In 1981 he joined Rabobank as an executive director and was appointed Chair of the Executive Board in 1986. In 1999 followed his appointment as Chairman of the Social-Economic Council (SER). From 2006 to 2008 Wijffels was executive director at the World Bank in Washington DC.

Herman Wijffels also served on a number of Boards in both the public and private sector, such as president of the Association 'Natuurmonumenten', the Board of Tilburg University and the Supervisory Boards of the Rijksmuseum, Utrecht University Medical Centre and TNO, as well as the corporate Boards of DSM and SHV.

From Food versus Fuel to Food and Fuel (plus Materials)

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Royal DSM N.V. is active in health, nutrition and materials. We connect our unique competences in Life Sciences and Materials Sciences to drive economic prosperity, environmental progress and social advances to create sustainable value. It is DSM's mission to create brighter lives for people today and generations to come. Sustainability therein reflects that we need to do more with less or in other words attain higher resource efficiency.

Doing more with less becomes particularly relevant once we embrace the notion that by 2050, we will need to feed 9 billion people and have to do that with a single planet, with the same amount of arable land and using sustainable energy sources that meet our growing demands. Innovative solutions, based on truly sustainable feedstocks, are pivotal to make this happen and it will require the concerted involvement of business systems that in many cases are not connected currently. Partnerships along and across value chains thus are part of the solution.

DSM is active in the biobased economy and is developing sustainable processes that are not in competition with the food chain for biofuels – biogas, cellulosic bioethanol and biodiesel – and biobased materials based on renewable building blocks such as succinic acid. These examples, in the context of an integrated biorefinery concept, will be presented.

Tuesday April 9th

New Perspective for Biorefinery of Cereals

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Cereals cover a world surface of 700 million ha (50% of arable land) and ensure a production of 2 200 million tons of grain and of about 800 million tons of straw. They are vital resources for many human activities. Their different applications have led to the structuring of multiple chains: food chains (milling, baking,...), feed industries, starch industry, and more recently biofuels. In the near future, also a protein industry may arise. All these sectors mobilize a range of different economic actors who are seeking to maximize their economic activities by adjusting their business models to the ability to produce or to add value. Such a sector-oriented optimisation leads to striving for maximal production of the key product of each chain whatever the opportunities are for by-products. Accordingly, this may result in our societies to a conflict of interests between cereal sub-sectors (as for example between food and fuel) which in turn reinforce competition between these sectors.

Even more one should take into account population growth, rising food related health problems, change of climate, pressure on fossil fuels, etc. A global solution is hard to reach, since one is dealing with balances between offers and demands by market prices. Such a balance is subjected to fluctuations inherent to agricultural production, which can cause highly significant price distortions and local difficulties. Regulations are set in place, however, not always easy to follow due to this dynamics. The alternative to this approach is to review the organization of the full cereal production to end-use system with the aim to satisfy each of these supply chains as well as possible at a global scale. This asks for a more holistic approach applied to the complexity of the full cereal system. In which one should understand major resources, operations and end-product opportunities.

The search for new ways of organizing cereal chains forces us to first reconsider the overall cereal production, a rich and diverse source of as renewable natural resources for (functional) fractions, ingredients or molecules. The production has to be adjusted and optimized according to the needs for final end-products and societal choices. Those should be better known (step 2). In this sense, the new concept of biorefinery appears as a major lever to meet such expectations. It should be re-adjusted and/or innovated accordingly (step 3). According to this holistic view, a new global approach should simultaneously focus on:

- Characterize plant resources as structural assemblies with specific compositions and properties defined by their end-use properties.
- Develop a new process engineering approach to improve the efficiency of dissociation, full usage of harvested biomass and of energy usage; the aim is to optimize the overall performance in terms of end-use value of all potential end-products rather than a single unit operation.

Finally, as part of a generic sustainability approach, cereal production and biorefinery processes must also remain compatible with local territories and the social-economic needs of people depending on the cereal system. Their cultural heritage should be taken into account. In practice, this means a search for innovative and competitive small-scale production units for a wider variety of grains and diversity of end-products.

Potentialities of Microalgae in a Context of Environmental Biorefineries

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Micro-algae offer a potential for innovation in energy, chemistry, human and animal nutrition, cosmetics because of their intrinsic richness. Indeed, micro-algae are known for their extraordinary composition: including protein, fat, fiber, vitamins, minerals and pigments. They still offer a field of exploration to develop innovative products, natural and functional.

In the context of global strategic challenges involving the development of biorefineries, micro-algae could be a solution for the future and bring major economic development in ten years. Research initiatives and investments in the field of micro-algae for energy purposes have increased considerably. 3rd generation biofuels can be produced using industrial CO₂ or substrate from sewage waters, following a recycling strategy and a management of waste due to human activity. Furthermore, the production can be performed on non-arable land. In addition, micro-algae are able to accumulate as much as half their weight in lipids, one of the primary matters for producing biofuels, which leads to potentially high productivity levels.

This presentation will highlight different key scientific challenges still to be addressed over the complete production chain and that could lead to a viable economic and environmental model.

Wednesday April 10th

BRIDGE PPP - Biobased and Renewable Industries for Development and Growth in Europe

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The bioeconomy presents formidable potential for Europe and its people. It can propel Europeans on the path of reindustrialization and sustainable growth, reversing the investment trend currently going to other regions of the world, and breathing new life into the whole economy. It offers prosperity across all EU regions, creating new and retaining millions of jobs. Farmers are to play a pivotal role to enable the bioeconomy to deliver locally sourced and produced materials. With sustainability at the heart of the bioeconomy, growth is decoupled from resource depletion and environmental impact. Instead, it boosts the EU's ability to transition more rapidly to a low carbon and resource efficient society. And finally, it enables the EU to lead and compete in a global bioeconomy market valued at €200 billion by 2020.

BRIDGE (Biobased and Renewables Industries for Development and Growth in Europe) is a Public Private Partnership between the Biobased Industries Consortium (BIC) and the European Commission that is currently in the making. Inspired by the vision and the potential of the bioeconomy for Europe, the PPP will lay down the foundations of this new economy, investing in research, the creation of entirely new value chains, and flagship initiatives to deliver a fully functional economy, based on the principles of excellence, sustainability, smart and inclusive growth. This public private partnership is a BRIDGE to a more prosperous future.

Designing Sustainable Biofuel Systems: Using our Heads to Product Food and Fuel from Cellulosic Biomass

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A high rate of energy consumption (about 4 kilowatts per capita) is required for human beings to obtain good levels of health, education and wealth. Currently, most energy consumption is based on non-renewable fossil fuels. Thus nearly the entire basis of the current health, education and material prosperity enjoyed by the developed world is not renewable. That prosperity will disappear when the underlying power consumption disappears. In this light, renewable energy is not just a "nice idea". Renewable energy is absolutely required if more human beings, now and in the future, are to achieve health, wealth and learning.

Energy consumption provides three primary services for humankind: heat, electricity (light) and liquid fuels for mobility. While we have many sources of renewable electricity and heat, only cellulosic biomass can provide large scale, low cost, low greenhouse gas liquid fuels. The cellulosic biofuels industry is just beginning to emerge. Our objective should be to make sure that this new industry is sustainable; that it meets the triple bottom line of people, planet and profit.

This presentation focuses on what may be the key barrier in many peoples' minds to sustainable biofuel production, the apparent conflict between food production and biofuels. Actually, if we think carefully and act creatively, there are many ways to integrate food production with biofuel production to their mutual benefit. Food production can increase with increased biofuel production, and many positive environmental services can be achieved simultaneously.

This presentation will discuss and describe some of these approaches for integrating biofuel production and food production. Thoughtfully designed biofuel systems based on perennial grasses and trees, in particular, can simultaneously provide very large environmental services including lower greenhouse gas production, carbon sequestration, enhanced water quality, greater available water quantity, more fertile soil and enhanced biodiversity. Thus sustainable cellulosic biofuel production is more limited by our intelligence and imagination than by hard resource or technical constraints. Therefore, let's use our heads.

Oral presentations

Session 1

Redesigning the business concept and sustainability

Monday, April 8th 2013

Ir. Haak room

Food-crops for Industry? Background information and surprising answers

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Triggered by the public discussion during the food crisis in 2008, politics and industry gave a too simplified answer to the potential food versus industry conflict: Industry should switch to non-food crops as soon as possible. From our point of view, the question of food versus non-food crops for industry is itself oversimplified and misleading. The real questions and conflicts are different, both uses compete for land.

This means, an appropriate way of finding an answer would include asking whether there are – in the country or region – free agricultural areas left, which are not necessary for food and animal feed production, domestic use or export? In most countries and regions, there is arable land left for potential biomass production for industrial uses – material or/and energy. In this case, the real question is: “How can we use these free areas as a sustainable feedstock for industry with the highest resource efficiency, the highest climate protection and the lowest impact on food competition?”

The aim of the presentation is, to provide a fact-based, more realistic and adequate view on the use of food crops in bio-based industries – apart from the often very emotional discussion. And the presentation also discusses how the additional biomass demand could be covered and how much arable land is still available.

How to interweave food, feed, fuel and fibre to an integral business chain

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Many organisations and studies have concluded biomass and biofuels to be fundamental for reaching to a low carbon economy. This is causing great controversy. 'Food' and 'fuel' are in various debates positioned as fierce opponents. But does that bring the desired future closer? Shouldn't we focus on how to interweave food and fuel, as well as feed, fibre and e.g. Pharma-products into a balanced and integrated business chain to fulfill the needs of our future society in a sustainable way? In the pre-oil age, when fossil resources, societies managed to fulfill their needs from what was abundantly available from agriculture and forests. The emergence of abundantly and at low costs cheap availability of fossil resources allowed fulfilling most of the energy and material needs from these fossil resources. One could argue that this 'allowed' the agricultural sector to prioritise and optimize, even monocultivate, towards food production. It goes without saying that the innovation and scientific focus have made tremendous achievements in both productivity improvement as well as in production volumes. To quote an FAO representative: "To date we globally produce sufficient food to feed more than 10 billion people". Facing climate change, the end of 'cheap oil era', increasing scarcity on a wide range of resources, a growing world population (of which still a too large group is below the poverty line and without proper access to food) – it is undisputable that today's global society is challenged to redesign its economic system in such a way that it remains affordable, climate neutral and resource secure and that respects socio-economic and environmental circumstances in a sustainable way. During the presentation the perspectives are sketched of one of the largest independent oil companies in north West Europe that operates as front runner in the development and usage of sustainable and 'best- in class' biofuels blends. The presentation will highlight how those activities support the development of biofuels with high carbon mitigation potential and limited land utilization performance will simultaneously strengthening the production of other agricultural and forestial products needed in a sustainable society.

Land use change: an essential aspect of Life Cycle Analysis of lignocellulosic biomass conversion processes

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The biomass is seen as a promising way to substitute fossil fuels. In a social Perspective, the lignocellulosic biomass conversion processes are interesting in that they imply less competition with food crops for land and water than first generation technologies. Moreover, lignocellulose is abundant in cheap and non-food materials extracted from plants such as wood and energy crops. So, the farmers can diversify their earnings. Nevertheless, the environmental impact of lignocellulosic biomass conversion should be accurately quantified. The Life Cycle Assessment (LCA) methodology should be used. Indeed, LCA deals with the environmental aspects and potential impacts associated with all the stages of a product's life from raw material extraction through materials processing, manufacture, distribution, use, and end-of-life. In this type of environmental assessment the energy and material fluxes for the entire life-cycle are surveyed and analysed with special attention to possible environmental hazards or human health problems. LCA also can be used to compare the environmental impact of different products or processes. So the LCA is the most suitable methodology to quantify the environmental impact of lignocellulosic biomass conversion pathways. The gasification process has been studied in details. Indeed, this process converts biomass into a combustible gas, called syngas, in the presence of a suitable oxidant. The syngas can substitute the fossil combustible in their two main applications: fuels and building blocks for the chemical industry. Since the syngas can be used directly to produce electricity and/or heat. Nevertheless, it also can be converted into a large range of products, such as diesel, via a Fischer-Tropsch process, or methanol, used for producing dimethyl ether, both of which can serve as fuels in engines. Syngas can also be used to produce ethylene and propylene, two building blocks for the chemical industry. Within this context, this study focuses on the state-of-the art of the LCA about gasification processes. This latter underlines the need of new methodological development in view of taking into account the land cover and the land use change impacts. Two mechanisms exist for land use change: direct Land Use Change (dLUC) and indirect Land Use Change (iLUC). dLUC occurs when new lands are taken into production and feedstock for studied product purposes displaces a prior land use. Whereas iLUC occurs when this previous land use remains necessary and so has to be implemented on a new land. The first steps in this methodological development are presented in our study. First, the concerned impact categories at each analyse level are detailed. Then the need to choose a land state as reference is underscored such as the necessity to determine the considered time period.

Demand orientation of sustainable innovations on food, fuels and materials

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Because of the difficult competitive power of biorefinery processes, broadly agreed contribution to sustainable development is critical for successful practical development of these processes. Practical development of biorefinery processes and chains therefore should not only be demand-oriented but also adequately address needs of other (societal) stakeholders. A large set of sustainability criteria exists, with lots of mutual contradictions. For example bioenergy development based on imported wood is at odds with sustainable forest and nature conservation, and from an eco-efficiency perspective intensive animal husbandry is more sustainable than organic farming. We need higher awareness of the differences in interpretation of the term 'sustainability' so that sustainable biorefinery initiatives can be more effectively communicated. Inevitably, initiators of new processes use a limited set of these criteria in dialogues with stakeholders. Adequate proactive positioning to market and stakeholders' preferences is essential for successful development, as is illustrated by practical successes and failures that will be presented at the symposium. Some examples:

- In regional initiatives, neighbouring citizens mostly are not directly involved. Their natural reaction (raising objections because of local nuisance) can be prevented through creating added value for them, for instance through share ownership or offering (residual) heat at low prices.
- Increasing crop valorisation effectiveness (e.g. through biorefinery) generally will be aimed at producing half-products. The biorefinery actor will be the main interested party to communicate this with the consumers; thus besides the buyer of the half-product also the consumers need to be convinced. At the symposium, the authors will further elaborate the broad definition of 'sustainable development'. Next, success and fail factors of various examples of practical business initiatives with biorefinery character will be discussed, with focus on interpretation of sustainability by different stakeholders.

Session 2

Integration of chains

Monday, April 8th 2013

Dorskamp room

Process Intensification in Biotechnology: towards an Integrated Biorefinery

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The strong growth of the world population and accompanying social development leads to pressure on the climate, scarcity of resources, environmental problems and reduction in biodiversity. One of the possible human mitigation responses is the development of a bio-based economy inspired by the use of renewable resources (biomass) and the full fractionation and transformation of it into food/feed – materials/chemicals – energy in the so-called cascade-approach.

Biorefineries, already existing for some decades now, were mostly focused on the food production chain. Nowadays, however, we are moving towards an integrated biorefinery in which either all compounds present in the biomass will be valorised as much as possible, or new biomass streams (including biowaste) will be tackled. New challenges exist in the disclosure of lignocellulosic biomass (second generation) in order to valorise not only the food components but also the straw, fibres, leaves, etc. Disclosure of these rather persistent molecules in an environmentally-friendly and sustainable way is still an important challenge. On the other hand, the extraction of several high-added value compounds (e.g., polyphenols) can be a success by using rather mild disclosure methods in order to keep its glycosilated forms intact and active. Although great expectations exist and a lot of development is going on at lab and pilot scale, still many problems need to be solved. It is known that between 50 and 70% of all the costs of a biorefined product are linked to its downstream processing.

The integration of fermentation or bioconversion processes with subsequent separation or purification processes will become the challenge of the coming years in order to allow a real breakthrough of the integrated biorefinery of the future. The integrated recovery of the produced molecules or the (in-situ) removal of inhibiting chemicals from the process will keep them under certain threshold limits leading to more efficient conversion processes or reduction in feedback inhibitions. The economical value of this integrated approach lies in the increase in conversion efficiency (less substrate that is not used) at higher rates and less energy consumption in the further downstream processing. Removal of organic acids by electrodialysis and related concepts will allow to produce acids without concomitant production of huge amounts of waste products (such as gypsum). Recovery of volatile alcohols by integrated pervaporation systems will reduce toxicity in the fermentation broth and lead to a first concentrated product flow. Other technologies such as membrane distillation, membrane extraction, adsorption or crystallization have also large potential in these integrated systems. An important discussion in these integrated systems concerns the need for a first (membrane) separation of the cells from the broth allowing a cleaner process to proceed for the further separation of the produced molecules. In fact, such membrane-based fermentors can probably become standard technology in the future. Not only fermentation (whole-cell based) processes can be integrated, but also enzyme reactors are under development. The immobilization of enzymes on beads or membranes can save on enzyme cost and improve the quality of the produced new molecules. Challenges and problems of these integrated conversion-separation processes will be discussed in detail leading to new insights in future biorefineries including (semi-) continuous processes compared to batch processes.

Keywords: process intensification, in situ product recovery, pervaporation, fermentation, electrodialysis, enzyme immobilization

On the systematic synthesis screening and integration of real-life biorefineries

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Biomass technologies have been inundated with new ideas and innovations the industrial world and the labs posing new challenges to chemical engineering. Individual bio-based paths, production of a single chemical product or fuel have gradually taken a significant position in markets. Given the plethora of degrees of freedom related to different feedstocks, paths and portfolio of products, the area seems to be chaotic. Opposing to the vast amount of options and possibilities, systems technology has offer powerful methods to optimize designs and systematize improvements of biomass processing systems. Systematic screening leads to a holistic approach of the problem far from the applications that focus on individual processes or plants. This paper introduces a methodology to address the systematic integration of biorefineries supporting decisions for selection and the integration of processing paths. The implementation takes the form of a synthesis model that is generalized with process and intermediates as appropriate to the case. For the estimation of cost the method uses regression models based on the thermodynamics of the process. The investment cost is quickly and roughly calculated based on energy losses resulting from the difference of the Lower Heating Value (LHV) of the inlet and outlet flows of a process. The methodology is applied to 82 different chemistries that lead to the production of more than 80 intermediate and final products. The biomass is decomposed to three main streams of C5 sugars, C6 sugars and lignin. Those intermediates are the precursors for bio-based products and fuels such as polyamides, PVC, polypropylene, ethanol, aromatic polyols, resins, vanillin etc. that can be their petrochemical counterparts. The model produces a rank of products identifying the optimal integrated flows. The optimization model results to an optimal solution of coproduction of xylonic acid polyamide and lignin based castor oil. An impressive observation coming out from the results is that high market price of the product does not offer tops in the ranking lists. Uncertainty analysis is presented additionally to highlight the sensitivity against variation such as market demands and prices, impact of the supply chain – transport cost. The case study of acetone and ammonia production has been used for illustration where integration of enzymatic fermentations and thermochemical manufacture of biomass are used for the production of desirable chemicals at the market demand. The case study additionally shows the switch towards the utilization of municipal solid waste when the biomass availability is limited and the cost of transportation high.

Assessment of coinciding and diverging requirements on the biorefinery system design following from new connections in biorefinery value chains

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The concept of biorefinery relies on the development of new connections between sectors at the resource or the product end of the refinery process. The agriculture and forestry, food and feed, chemical and energy sectors, and within those sectors, enterprises at different scales or customer groups, each have their own rationality for optimisation of their part of the value chain from feedstocks to customers. They may also have different drivers to engage in biorefinery, such as security of feedstock supply, profit margins, or the 'green' value of using biomass. The cooperation between the stakeholders in biorefinery will require understanding of coinciding and diverging preferences between them that influence the design, location and process decisions of the biorefinery system. This paper presents an analysis of coinciding and diverging preferences as characteristics of specific value chains with the objective to contribute to actor engagement and policy steering of biorefinery systems. The approach is based on the development of contrasting situations in which a biorefinery links the value chains of at least two sectors. The contrasts are built according to complexity and scale of the biorefinery. This leads to the selection of three situations for analysis: firstly large scale centralised biorefinery with high complexity integrated into existing large scale oil refinery, secondly intermediate scale biorefinery with intermediate complexity being linked to several value chains, and thirdly small scale biorefinery with relatively low complexity. For these three situations, the endogenous characteristics are assumed to influence factors of biorefinery system design through setting demands on cooperation within the value chain or sensitivity to demands from the value chain or context. The relevant factors such as location, process efficiency and product quality, are identified based on a literature review and on interviews with experts from actors along relevant value chains that are or may be involved in the three defined contrasting situations. The situation descriptions and literature based mapping of factors serve to guide reflection by these actors on drivers that underlie preferences. The sensitivity to specific characteristics of the biorefinery is structured based on the classification of biorefineries according to feedstocks, platforms, processes and products. This structured analysis and mapping of coinciding and diverging preferences for the development of biorefinery value chains and their causes forms an input for actors in the chains and policy developers to identify opportunities for steering the emerging developments in biorefinery.

Towards on-shore aquaculture in the Eemshaven

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Bioclear has performed a feasibility study, initiated by the Biorenewables Business Platform and Energy Valley, on the large-scale cultivation of microalgae. Objective was to make a step-change in the scale of microalgae cultivation to a total land area of 10 hectare using proven algae cultivation techniques in open pond systems. By this, a unique opportunity is created for hands on experience in the large-scale cultivation of micro-algae and other aquatic biomass like duckweed or Azolla. Although the farm has a pilot character with a knowledge building objective, the underlying business case should be promising enough for investors to participate from the start onwards. Target focus region of this study has been the Eemshaven region, the most northern part of Dutch mainland, due to the combination of agro with industry - the availability of industrial waste streams, space and salinated farmland. The strong link of this region with the Waddenzee makes it at the same time essential to find a proper balance between economy and ecology. Enablers for economic feasibility were found in the integration of chains. By utilizing locally available industrial waste streams as water, heat, CO₂ and nutrients cycles can be closed locally, costs can be avoided and yields improved. The initial focus on finding a customer for the substantial amounts of protein rich biomass in the feed sector did however not result in a viable business model. Economy of scale had little impact in improving the proposal. Therefore the feed-option was abandoned and in the second phase focus was given to both higher value end-applications that at the same time would reduce cost from the technology side. The solution was found in the on-shore aquaculture, the cultivation of filter feeders such as oysters and clams for which algae are the essential natural food source. On the one hand costs are reduced as expensive harvesting techniques can be removed from the equation for aquaculture. On the other hand the value of the protein rich end product for human consumption is much higher. The business case of this scenario did show economic potential. During this study the microalgae farm evolved into an on-shore aquaculture farm. The vision and proposal have already yielded support from a broad range of parties during the end meeting of the project. Amongst the enthusiastic attendants people willing to conserve the environment, people willing to create economic activity, people willing to provide technology, waste streams or knowledge. The potential is definitely there to proceed with next steps. First contacts with parties cultivating shells in Zeeland have been made, the plans have been met with enthusiasm and there seems to be a strong will for cooperation.

Session 3

Biochemical conversion and separation I

Monday, April 8th 2013

Ir. Haak room

Developing Pretreatment and Fractionation for Integrated Biorefineries

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Lignocellulose, the most abundant renewable biomass on earth, is composed mainly of cellulose, hemicellulose and lignin. Both the cellulose and hemicellulose fractions are polymers of sugars and thereby a potential source of fermentable sugars for microbial fermentation to biofuels and chemicals. Lignin can be used for the production of chemicals, combined heat and power or other purposes. In order to valorise the different composing elements of lignocellulose, a pretreatment process is required. Pretreatment generally includes a physical or chemical pretreatment process in which cellulose is made accessible for further conversion, and enzymatic hydrolysis of cellulose and hemicellulose to fermentable sugars using cellulase enzymes. In addition, pretreatment requires suitable separation or fractionation process to supply further downstream processes with biomass fractions of sufficient concentration and purity.

The selection of suitable pretreatment techniques depends strongly on the objective of the biomass pretreatment, such as desired biorefinery products, its economic assessment and environmental impact. Only a few pretreatment methods are reported as being potentially cost-effective thus far at larger scale. These include steam explosion, liquid hot water, concentrated acid hydrolysis, dilute acid and alkaline pretreatments. Obstacles in the existing pretreatment processes include insufficient separation of cellulose and lignin, formation of by-products that inhibit fermentation, high use of chemicals and energy, and considerable waste production. Looking at industrial activities for the production of ethanol, acid-based pretreatment methods are often preferred where lignin is left with the substrate and removed after the hydrolysis or even after distillation. Research topics for these processes include minimization of sugar loss, increase of solids concentration and higher ethanol concentrations after fermentation. However, when lignin is removed from the biomass in an early stage of the process it can be recovered as a co-product with potential high added value. Another advantage is that lignin removal greatly enhances enzymatic hydrolysis, which means that delignification becomes more important.

This presentation will present a number of development strategies in pretreatment by highlighting recent pretreatment research conducted at Wageningen UR, in collaboration with research partners. Research is directed at identifying the effect of different pretreatment technologies on (optimal) separation of lignocellulosic biomass, taking into account various biomass resources (e.g. herbaceous and woody biomass, agro-food processing residues) and quality aspects of the main resulting streams. The quality of the resulting streams (cellulose, hemicellulose, lignin) from lignocellulose will highly affect its further valorization potential towards fuels, chemicals and materials. By using state-of-the-art characterisation techniques combined with application tests this potential is evaluated. Finally, a key element in the development strategy is to identify necessary processing technologies that meet both the goals of downstream biorefinery (to produce biofuels and chemical building blocks), and that can ensure the recycling of nutrients and minerals to the field that are otherwise exported.

Pilot plant for the scale-up of continuous mode lactic acid fermentation

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Renewable feedstocks (e.g. crops, lignocellulosics, green biomass, residues) are being used as raw materials for the production of microbial lactic acid. Lactic acid, its salts and esters have a wide range of potential uses and are extensively used in diverse fields. The goal is to develop a fermentation process based on the substitution of expensive nutrient supplements by cheaper materials from biomass due to their main proportion of the whole process costs.

The scale-up to a technical scale of several processing steps have to be developed for transferable solutions of biotechnologies for renewables. For that purpose a multifunctional pilot plant was planned and built at the site of ATB to investigate different raw materials and products. First results of the continuous lactic acid fermentation in a 450-L-bioreactor will be presented. One of the usual ways to keep the biomass inside of the system for increasing the overall productivity is the cell retention with hollow fibre membranes. In comparison to the process without cell recycle (e.g. chemostat) there is a triple up to four time's higher productivity of lactic acid.

Depending on the further processing of the lactic acid the separation of impurities after fermentation is a major process cost too. Therefore an optimization is necessary to find a balance between the substitution of expensive nutrients and the limitation of interfering or undesirable components of natural raw materials respectively. Exploitation of high quality L(+)- and D(-) lactic acid for the production of biopolymers is one of the recent applications. Conventional processes for down-streaming are based on precipitation steps that generate large amounts of chemical effluents. Consequently the environmental impact of traditional processes can be reduced by using alternative technologies, such as electrodialysis with monopolar and bipolar membranes.

Bioelectrochemical production of caproate and caprylate from acetate

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The use of mixed cultures to convert waste biomass into medium chain fatty acids, precursors for renewable fuels or chemicals, is a promising route. To convert waste biomass into medium chain fatty acids, an external electron donor in the form of hydrogen or ethanol needs to be added. This study investigated whether the cathode of a bioelectrochemical system can be used as the electron donor for the conversion of acetate into medium chain fatty acids. We show that medium chain fatty acids were produced in a bioelectrochemical system at -0.9 V vs. NHE cathode potential, without addition of an external mediator. Caproate, butyrate and smaller fractions of caprylate were the main products formed from acetate. In-situ produced hydrogen was likely involved as an electron donor for the reduction of acetate. Electron and carbon balances revealed that 45% of the electrons in electric current and acetate, and 31% of the carbon from acetate were recovered in the formed products. This study showed for the first time production of medium chain fatty acids caproate and caprylate from acetate at the cathode of bioelectrochemical systems, and offers new opportunities for application of bioelectrochemical systems.

New Driving Forces For Dry Fractionation

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Current wet bio-refining techniques or waste valorization processes, e.g. purification of gluten from wheat, are carried out under harsh conditions (high temperature, addition of acids, etc.), which degrade the functionality of individual components. Besides, the processes usually require a suspension or dissolution into copious amounts of water (or other solvents), which will not end up in the final product. This does not only generate huge quantities of waste water, but also leads to large consumption of energy for dehydration. A dry fractionation method will reduce or even eliminate waste water generation, strongly reduce energy usage and maintain the functionality of product. This PhD project is part of a larger project with the ambition to develop a novel dry fractionation process for concentrated particulate streams, which relies on the use of new driving forces, such as electrostatic force. This is achieved by first tribo-electrically charging the particles and then letting them migrate under the influence of an external electrostatic field. In our study, tribo-electric charging behaviour of dry particles are experimentally analyzed, in the first instance, by using well-defined particles made from polystyrene in combination with an aluminum charging tube. The results gave insight in the dynamics of the charging process, i.e. charging behaviour of concentrated particulate streams as a function of a.o. particle size, concentration, gas velocity and humidity. These results will be further used for validation of numerical simulations. Currently, the charging behaviour and consequent separation of industrially relevant feedstock is evaluated (wheat gluten and starch, bran millings with d_p varying between 50 - 300 μm). The final aim of this project is to design and evaluate a laboratory device that can fractionate industrially relevant feedstock.

Bodec's valorization approach: The Primordial Soup Concept

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Nearly 2.5x10⁸ ton/year of side streams are produced in Europe as a result of food processing. Typically such streams are partially valorized into animal feed, composted or wasted. These side streams, however, are an interesting and abundant source of functional molecules with potential application in the several industries. Bodec Process technology is investigating several approaches for valorizing streams from common vegetable products in the Netherlands. One of such approach was denominated the "Primordial Soup (PS)" and establishes a valorization road map. As described in figure 1, the PS is formed from side streams from the food processing industry. The valorization scale starts with the separation of high value products like proteins and phenols. Proteins can be fractionated and purified from a watery phase through continuous chromatography (SMB) while phenols can be extracted using green supercritical technology. Phenols can be further purified also through SMB. Lipids can be extracted with solvents or with supercritical solvents. The remaining material is rich in fibers and lignin. Selective extraction can yield fibers like starch and cellulose. Through bio-chemical or thermophysical processing, chemical building blocks or energy can be obtained. Preliminary studies have been performed in order to investigate the feasibility of such valorization strategy. Considering an initial stream composed of tomato, cucumber, carrot and onion, four valorization scenarios were analyzed: o Complete Valorization (CV): Separation of proteins (SMB) and extraction of phenols and lipids with organic solvents. o Green Valorization (GV): Purification of proteins (SMB) and extraction of phenols and lipids with supercritical solvents (SCE). o Mild Valorization (MV): Extraction of proteins and phenols and further chromatographic fractionation (SMB). o Protein Valorization (PV): Only purification of proteins (SMB). Conservative figures regarding prices of raw materials, chemicals usage and purification yields were used. It was assumed that all generated products can be sold at low range market prices. Biochemical and thermophysical conversion were not considered in this study. It was also stated that the remaining stream can be safely disposed. The results indicate that the most interesting scenario is GV followed by PV. The least interesting scenario is CV. This is due to the high chemicals consumption during organic extraction. On the other hand, SCE offers an important advantage in terms of solvent recovery and mild processing. Both SMB and SCE ensure functionality in the final product which benefits directly the expected selling price. This pre-evaluation shows remarkable opportunities for the valorization of waste streams. Bodec is taking the lead in investigating integral and green approaches to generate value out of side streams. Further development is needed to take these studies into business opportunities. Figure 1. Bodec's valorization approach.

Seaweed biorefinery: production of fuels and chemicals from native North Sea seaweed species

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Seaweeds have been used in East Asia since ancient times as vegetables. Currently, seaweeds are worldwide used as food and as source of chemicals (i.e. thickening agents, gelling agents and phycocolloids). Annually, 7-8 million tonnes seaweeds are harvested, with an estimated total value of the products of US\$ 5-6 billion [1]. Because of the special chemical composition (wide range in sugars, polymers, etc) of seaweeds and the possibility of cultivating them at large scale in the ocean with high yields, they are potential feedstocks for production of renewable chemicals and fuels [2]. Seaweed biomass is highly suited as raw material for the co-production of chemicals, biofuels and energy via the biorefinery approach. In this project, adjusted and efficient biorefinery strategies and concepts are developed for seaweed biomass. In the proposed process, the seaweed is first dewatered and subsequently fractionated into its main components. In the second stage of the biorefinery, the main components, sugars, proteins and minerals are converted into bulk chemicals and energy carriers. The project aims to develop catalytic, enzymatic and fermentative conversion routes. A biorefinery route have been assessed for brown seaweeds (*Saccharina latissima*), which are a known source of hydrocolloids. Freshly harvested samples of *Saccharina latissima* have been pressed using different techniques (pressing, refining) in order to reduce the water content. Mannitol from this seaweed was extracted, purified and used as precursor of chemicals or as fermentation feedstock. In addition, sugar-rich fractions from different seaweeds (including *Saccharina latissima*, *Palmaria palmata* and *Ulva lactuca*), have been characterized as substrate for the production of acetone, butanol and ethanol by fermentation. Acknowledgement

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Biorefinery concept applied to microalgae

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For more than 3 billion years, the photosynthetic machinery of microalgae uses the solar energy to convert carbon dioxide, nitrates, sulfates and other 'waste-nutrients' into dioxygen and biomass. It is estimated that algae, with over 100 000 strains ranging from microscopic single cells to giant ocean seaweeds kilometers in length, constitutes roughly half of our world's biomass. Kelp (seaweed) has long been processed for phycocolloids, while some microalgae are harvested, dried and sold as food supplements. Still, the market for products extracted from microalgae is in its infancy. Most published papers state photosynthetic microalgae as a renewable feedstock for production of biofuels. However, higher value chemicals such as pigments, antioxidants, polysaccharides can be extracted, as well as proteins, often the most important fraction of the biomass. Nutritional studies have demonstrated that these algal proteins are of high quality and comparable to conventional vegetable proteins. The microalgal biomass is a source of value, from monetary, ecological and ethical perspectives. This value must be gained by a clever processing of the biomass. Very little information is available on the whole fractionation of microalgae: known activity has dealt exclusively with the extraction of one specific compound for commercial purposes. In most cases, the target compound is a high value-added molecule for cosmetics, pharmaceuticals or chemicals. There is no proven set of unit operations leading to separated oils, proteins, carbohydrates, fine products, etc., in a sustainable and economic manner. The challenge is to find an efficient process where consecutive steps can give access to the different product classes, with minimal resource destruction when going from one step to the following. We need to develop an "AlgoRefinery", a production unit separating the various components by a cascade of extractions and treatments, and to optimize their valorization at the consecutive steps. This strategy can be considered as the eco-conception of an integrated production of microalgae. AlgoSource is a company developed around three main activities: the production of functional ingredient from microalgae, the process engineering for the production and the valorization of microalgae and the market studies. Due to its knowledge and its expertise, AlgoSource is involved into two projects that will be presented, a European one (FP7 Biofat) and a French one (Algoraffinerie), whose goal is to develop the biorefinery of microalgae. A promotion process of the red microalgae *Porphyridium cruentum*, already developed and tested, will be presented. It's composed of 4 steps: one for the production of the biomass, one for the harvesting, one for the cell disruption and selective aqueous extraction and a last one for the purification of the compounds. A similar approach is in progress for the species *Chlorella vulgaris*, *Nannochloropsis oculata* and *Tetraselmis suecica*.

Session 4
Chemicals, Fuels and
Materials I

Monday, April 8th 2013

Dorskamp room

Alternatives to fossil Jetfuel and related challenges

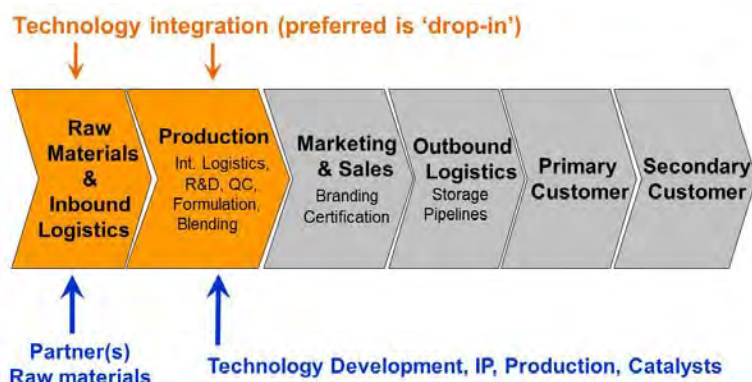
Kraft, Axel

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“Biofuels offer opportunities to developing countries to grow new livelihoods and reduce dependency on imported fossil fuels, as well as the obvious benefits to the environment and the industry. These are compelling reasons for governments to get on board”. Those were the words of Paul Steele, Director Environment of IATA (International Airport Transport Association) in December 2010. Moreover, he stated that if airlines wait until the price is right and commercial quantities are available Biofuels might never happen. For kerosene alternatives like batteries for cars are not an option.

Aside from technical issues there are tough challenges to overcome: biofuels, like any alternative fuel must be competitive in price and available in quantity. Additionally, competition of raw materials with food, chemicals and materials and a limited amount of arable land to grow raw materials are major hurdles to overcome. To meet sustainability criteria waste streams, non-food material like algae or carbon mono- and carbon dioxide are preferred, but there is very limited experience in the refinery for them. Alternative kerosene production demands coupling of hitherto not linked value chains and partially new supply chains for petrochemicals, chemicals, agriculture, food, fodder and algae on a global scale (see figure below).

Uncertainty of prices for key commodities, oil and new commodities to come as well as unclear legislation will guarantee for an interesting journey ahead. Many start-ups compete already with the big petrochemical players on technology development and are permanently pushing the boundaries of existing knowledge. For the time being all alternative fuels must be blended with fossil fuels (fossil part to be at least 50%). However, start-ups are typically not aware of procedures for quality control for jetfuels. This presentation will give an overview about the technologies in the pipeline, in particular alcohol-to-jet and hydroprocessing of fats and oils, as well as the related challenges ahead.



Value chain for alternative kerosene focusing on key aspects (Source: Fraunhofer UMSICHT)

BioSyn: Production of green chemicals from industrial waste streams

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Waste streams containing large amounts of organic material are a potential source for the production of biofuels and green chemicals. These streams are for example available in the paper industry, waste management and agriculture. At the moment, industry often incurs significant costs for treatment and disposal of wet waste streams, although these generally contain a significant amount of organic material and have therefore potential for the production of green chemicals like volatile fatty acids (VFA), alcohols and bioplastics (PHA). The use of inexpensive substrates will both increase the economic viability of the conversion of biomass to green chemicals and can provide new markets for these waste streams. Within BioSyn, part of the project BioCab, the potential for biological production of green chemicals from wet organic waste is explored by using the synthesis power of natural biological processes. Organic material generally consists of both easily degradable organic components and persistent fibers such as cellulose, hemicellulose and lignin. One of the challenges in the project is therefore the pretreatment and enzymatic hydrolysis of fiber- and cell-rich streams for increasing the accessibility of fibers and large molecules for biological degradation. Production of green chemicals from biomass will be stimulated by using open mixed fermentations. These have the advantage of being robust, inexpensive and easy to manage. Within BioSyn is being explored how the production of green chemicals from open mixed fermentation technologies (on large scale) can be deployed, managed and controlled for maximum conversion of biomass to valuable chemicals and fuels. Therefore, formation of low-value chemicals and fuels such as methane must be avoided as much as possible. Finally, the separation and isolation of the produced chemicals from the organic waste is a major challenge to achieve a total concept for the industry. The project aims to develop knowledge and technology for the production of biofuels and green chemicals from waste streams using robust and cost-effective biological processes. The three-year project was launched in the spring of 2011 and since then, promising results have been obtained on lab-scale. The results are likely to be implemented on larger scale in 2013.

Food, fuel and bio-plastics from Sweetstem Sorghum

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The effects of climate change and shifting weather patterns created an increasing awareness of the need for sustainable water resources for future economic growth. Bio-based products have been criticised for excessive water consumption and for increased food prices. Sweetstem sorghum is a hardy crop that is drought resistant and can provide a large biomass crop on marginal land, with relatively little water. In this paper, the use of sweetstem sorghum biomass to produce food, biofuels and biobased plastics is presented. Sweetstem sorghum were planted and harvested at 3 and 6 months respectively by the Agricol Company near Potchefstroom. The juice extracted from the stalks of the USA1 cultivar (0.5 L/kg stalks) contained the highest concentration of sugars (440 g.L⁻¹). A maximum ethanol yield of 170 g.L⁻¹ (0.48 g.g⁻¹) was obtained from fermentation of the sugar juice. The bagasse left over after extraction of the juice was pretreated and hydrolysed in a single step using microwave irradiation. The highest reducing sugar yield of 0.82 g/g substrate (96% conversion efficiency) was obtained with microwave irradiation at 180W for 20 minutes in a 5wt% sulphuric acid solution. An ethanol yield of 0.13 g/g bagasse (conversion efficiency of 98%) was obtained after 24 hours of fermentation when using a 10:5% v/v of *S. cerevisiae* to *Z. mobilis* ATCC31821 ratio. The theory of using lignin obtained from plant material for the production of a bio-polymer was investigated by using lignin as biomass source and dirty glycerine from a biodiesel production process to produce biodegradable polyurethane foam. It was found that the optimal conditions for the production of a polyol with desirable characteristics were a reaction temperature of 160°C, a reaction time of 1.5 hours, a solvent to lignin ratio of 9:1 and a pH of 8 for the mixture. A maximum yield of 0.4 g polyol per gram of lignin used was obtained. Polyols with a hydroxyl number of 307.93±38.07 mg KOH/g were produced and with these production conditions the polyols produced a light coloured, quite soft and brittle polyurethane foam with a density of 107.78±14.72 kg/m³. The process used produced 1.85±0.15 g foam per g polyol used for the production. A biodegradability test showed that the produced foam is biodegradable and could thus be used to potentially replace petroleum based polyurethane foam products. If the USA1 cultivar is used as an example, approximately 3000 kg of food grain, 6000 L of juice, 13000 L of ethanol and 700 kg of biodegradable polyurethane can be produced per hectare planted on marginal land. This makes a convincing case for using sweet sorghum as a crop for feedstock to a bio-refinery to produce food, fuel and bio-chemicals.

Up-grading of wheat straw lignocellulosic hydrolysates to bioplastics

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Agricultural lignocellulosic residues such as wheat or rice straw are abundant feedstocks that have low economic value and are normally used as cattle feed. They are, however, a potential source of mono- and disaccharides and thus can be upgraded, namely in the production of polyhydroxyalkanoates (PHAs). Polyhydroxyalkanoates (PHAs) are biodegradable polyesters with properties similar to some of the conventional plastics. They accumulate in the cytoplasm of bacterial cells as reserve materials under unbalanced cultivation conditions, e.g. lack of nitrogen or phosphorus and excess of carbon. In this work, wheat straw hydrolysates were prepared by Biorefinery.de GmbH by pretreating the biomass using the AFEX process followed by enzymatic hydrolysis and a subsequent concentration step. *Burkholderia sacchari* DSM 17165 was the selected strain due to its ability to convert both glucose and xylose (the main sugars in the straw hydrolysate) into considerable amounts of poly-3-hydroxybutyrate (P(3HB)). Growth and biopolymer production were studied using the real hydrolysate and also a mix of sugars simulating the hydrolysate composition (control) in 2 L stirred-tank reactors (STR) operating in the fed-batch mode. A polymer concentration of 84 g/L was reached corresponding to an accumulation in the cells of 68 % g P(3HB)/g CDW. Polymer yield and productivity was 0.22 g P(3HB)/g sugar and 1.6 g L⁻¹h⁻¹, respectively. These values were very similar to those obtained in the control cultivations. P(3HB-co-4HB) copolymers exhibit attractive thermal and mechanical properties since the presence of the 4HB monomer reduces the melting temperature, the polymer crystallinity and provides higher flexibility, thus facilitating plastic processing. These copolymers find numerous applications in the medical and pharmaceutical fields. Fed-batch strategies for the production of P(3HB-co-4HB) on glucose and gamma-butyrolactone were developed, indicating that this strain is able to accumulate the copolymer at various 4HB molar %s. Lastly, production of P(3HB-co-4HB) using wheat straw hydrolysates as major carbon source has been showing rather promising results. This study revealed that upon processing of wheat straw, an hydrolysate rich in monomeric sugars is obtained which can be upgraded into valuable bioplastics, ie, the biorefinery concept was applied to microbial bioplastics production.

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New pathway to fuels or fuel additives starting from short-chain alcohols

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A new catalytic gas-phase process developed by Fraunhofer UMSICHT allows coupling of short-chain alcohols and ketones to larger hydrocarbon molecules containing only one atom of oxygen. The main by-product is water. Starting off from fermentation based alcohols and acetone made e.g. by fermentation of carbon-monoxide rich off-gas or by ABE fermentation processes are particularly attractive. After an additional oxygen-removing step, for example hydrotreatment, molecules identical to fossil fuels are yielded. The first step comprises the self-condensation of short-chain alcohols yielding a mixture of linear and branched longer-chain alcohols. Typical process conditions range from 300 to 400 °C at a pressure of up to 50 bars. In a second step the obtained mixture of alcohols is blended with acetone and converted in a similar gas phase reaction to a mixture of branched and linear secondary alcohols and ketones. This condensation step also works with a typical aqueous ABE fermentation broth. Separation of reaction water and additional removal of oxygen, e.g. by hydrotreatment, will not be discussed in the presentation, since it is considered state-of-the-art. Depending on raw materials and process conditions a tailor-made product distribution of hydrocarbons can thus be realized. The resulting mixture of products can be employed as fuel additive, as a neat fuel or as a blending component for either diesel or kerosene. The presented overall process scheme offers an attractive alternative to other competing processes producing long-chain hydrocarbons like Fischer-Tropsch or hydrotreatment of fats and oils.

Dehydration of lactic acid and 3-hydroxypropionic acid to acrylic acid

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Biomass has attracted intense attentions recently as an alternative energy source and chemical feedstock due to the depletion of fossil fuels and rising concern over environmental protection. Chemicals manufactured from petroleum feedstocks suffer high and volatile prices. For example, acrylic acid - a widely used commodity chemical in the manufacture of paint additives, adhesives, textiles and super-absorbent materials is mainly produced from petrochemical industry nowadays by partial oxidation of propylene. The general drawback of this route is the total dependence on the nonrenewable fossil resource. Therefore, finding an alternative and sustainable route of acrylic acid production would be of great importance. The recent development of modern bio-engineering together with conventional chemical process gives a new potential for acrylic acid production from biorenewable sources. In our work, dehydration of LA and 3-HP to acrylic acid is studied on solid acid catalyst. The reactions are performed in a fixed bed reactor and aqueous solutions of LA or 3-HP are used as feedstock. The key issue of the proposed dehydration pathways is to develop a high efficient and robust catalyst system. It is found that Si-Al based zeolites have better catalytic performance than other types of materials, such as pure silica zeolite, metal oxides and inorganic salts. The superiority of Si-Al based zeolites is attributed to its Lewis acid site, suitable pore size and large surface area. Interestingly, the Bronsted acid sites behave differently in the feedstocks of LA and 3-HP. For LA, high density of Bronsted acid sites gives very low selectivity of AA due to the undesired decarboxylation reaction. As a result, unfavored acetaldehyde is the main product. However, when 3-HP is used as a substrate, high yield of AA is obtained on strong Bronsted acid zeolite. Such interesting behavior suggests that the different position of OH group in the LA and 3-HP plays an important role in the undesired decarboxylation reaction. Alkali bases modified Si-Al zeolites are also developed with significant improvement in the selectivity of acrylic acid for the dehydration of LA. In general, 3-HP dehydration gives higher yield of acrylic acid on solid acid catalyst. However, the production of 3-HP from bio-mass fermentation is much costly than lactic acid, so each reaction pathway has its own advantages and disadvantages.

Aminolevulinic acid and biohydrogen production from agro-industrial by-products in a biorefinery concept

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The potential of biohydrogen and 5-aminolevulinic acid productions by *Rhodobacter sphaeroides* O.U.001 were investigated in a biorefinery concept. Waste barley and sugar beet molasses were used as substrate after pretreatments in photofermentation. Waste barley hydrolysate and molasses were analyzed in terms of their total simple sugar, organic acid, ammonium, element and total phenol contents. Various growth media having different sugar content (5 g/L - 11 g/L) were prepared using waste barley hydrolysate and molasses to produce biohydrogen and aminolevulinic acid. Increased sugar concentrations resulted in higher cell density and hydrogen accumulation. Accordingly, highest cell density (OD₆₆₀: 1.78) and hydrogen production (0.4 L H₂/L culture) were observed in 11 g/L sugar containing medium when using waste barley hydrolysate. 67.4 µM 5-ALA production was obtained in the same culture conditions. In the case of using sugar beet molasses (8 g/L), higher cell density (OD₆₆₀: 5.0), higher hydrogen accumulation (0.48 L H₂/L) and higher ALA production (188 µM) have been recorded. These results showed that waste barley and sugar beet molasses can be used as substrate for *R. sphaeroides* for biohydrogen and aminolevulinic acid production within a biorefinery concept.

Session 5
Biochemical conversion
and separation II

Tuesday, April 9th 2013

Ir. Haak room

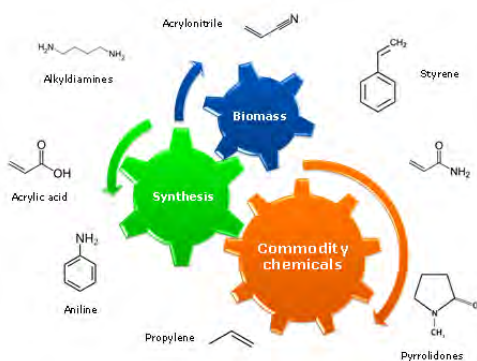
Integration of chemistry into agro-food chains to achieve a win-win situation

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Industry is exploring the use of biomass in the production of chemicals by fermentation and chemical technologies. Most approaches are compartmentalised: someone isolates the raw material another focuses on a transformation and another examines downstream processing. There is a lot of effort into the transformation of biomass derived molecules using (bio)catalysts but they do not consider how to obtain the starting material from heterogeneous streams with large amounts of water that needs to be removed at high energy and economic cost. Transformations that facilitate upstream and downstream processing will lead to more effective production routes.

Here we describe new approaches that allows (raw material) molecules to be more readily isolated and transformations that facilitate product isolation. We describe how heterogeneous aqueous (waste) streams can be used in microbial processes to produce well defined, readily isolated compounds (a number of polymeric species can be considered) and their conversion to an array of industrial chemicals. For example, cyanophycin (produced by cyanobacteria) is a polymer consisting of a poly(aspartic acid) backbone with arginine side chains that can be accumulated in cells using an "aqueous soup" of various organic compounds and amino acids. The cells can be isolated and the polymer removed in a simple procedure. Selective hydrolysis of the polymer allows formation of arginine leaving the backbone intact. Arginine can be used in immobilised enzyme conversions leading to 1,4-butanediamine, a co-monomer in nylon-4,6. As well as this aspartic acid can be used as a raw material for a number of chemicals traditionally produced from fossil resources. The use of (bio)chemical conversions (in general terms) can also lead to changes in chemical and physical behaviour thereby allowing in situ product formation and recovery here we describe such a process using electrodialysis.



Can biomass rest streams lead to a win-win for the agro-food and chemical industries, where there is an improvement in its current use while still supplying raw materials for the chemical industry? The use of non-essential amino acids as chemical feedstocks and the chemistry behind it will be described and to what extent the techno-economic and ecological aspects can compete with fossil based processes.

Biogenic Isosorbide tert-Butyl Ethers (ITBE)-Continuous Production and Reaction Kinetics

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The demand of energy and oil derived products is steadily increasing but fossil fuels are depleting. Hence, efficient approaches to convert biomass as an alternative source for those products are currently one main focus in research. Isosorbide is a versatile dihydro sugar alcohol that can be derived from starch as well as from the abundant biopolymer cellulose by hydrolytic hydrogenation and dehydration.[1-4] It is a most promising platform chemical for the production of biopolymers.[1] But also novel surfactants, high-boiling green solvents and biogenic fuel blends can be produced, especially from isosorbide ether derivatives. Instead of the conventional routes such as the Williamson etherification using alkyl halides, our work focuses on the heterogeneously catalyzed etherification of isosorbide by acidic ion exchange resins such as Amberlyst-15. We used two different approaches for the production of isosorbide tert-butyl ethers (ITBE): the condensation of isosorbide with tert-butanol avoiding by-products other than water and its addition to isobutene.[5] The latter approach was studied in autoclave reactions under pressure as well as under ambient conditions. In contrast to the established production of MTBE/ETBE from liquid substrates, ITBE production requires addition of a solvent since isosorbide is a solid at ambient conditions. Due to the different configurations of the hydroxyl groups in isosorbide three different products can be obtained: the diether as well as two different monoethers. Various reaction parameters for both pathways have been optimized. The best results were obtained by sparging isobutene through an isosorbide solution at room temperature. Almost full conversion of isosorbide was achieved with 65 % yield of the diether using dimethyl carbonate as solvent. Therefore, we focused on the isobutene approach for further studies. To facilitate the development of an ITBE production process, we investigated the reaction kinetics by determining reaction constants as well as activation energies. Furthermore, with regard to potential large scale applications such as fuel additives or green solvents we developed a lab-sized reaction set-up for the continuous production of ITBE.

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Ion exclusion chromatography a potentially valuable tool for fractionation of complex fluids: Juice de-acidification as a case study

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The need of separating fraction(s) rich in a specific compound/ingredient is a common theme in both food and nutritional sciences. In food science the emphasis is more on the development of a cost effective industrial scale separation process that yields desired fraction specifications. In nutritional science the emphasis is more on obtaining fractions rich in specific compounds from given sources (e.g. fruits, vegetables, botanicals) for delineating the role of those compounds on disease prevention. Ion Exclusion Chromatography, IEC, has applications from laboratory to industrial scale separations. Coupling IEC with advanced design chemical engineering approaches for industrial chromatography has the potential to be a valuable tool for fulfilling the needs of food scientists and nutritionists. In this study the basic mechanisms of IEC are presented and apple, orange & beetroot juice de-acidification is investigated at 2 elution speeds aiming to report on the feasibility of IEC for partial de-acidification in batch mode and the potential of complete separation of targeted acids from sugars in high yield and purity using Simulating Moving Bed, SMB, design theory. When IEC is practiced on batch mode our results shown complete separation of key acids for each commodity to be feasible only at 2.5ml/min flow rate. Following the Mazzoti-Morbidelli SMB design theory, our data shown complete separation to be also feasible at elevated flow rates such as 5mL/min. These results provide a promising case study for fractionating complex fluids with minimum impact on their naturalness and avoiding process-induced chemical alterations of the fractions isolated

Design and evaluation of flocculation based harvesting processes for microalgae biomass production

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The transition of a fossil fuel to a biobased economy in combination with the prospected increase in the world's population will lead to a growing demand for additional sources of biomass to complement traditional agricultural biomass. Microalgae are a promising new source of biomass. These aquatic microorganisms receive much interest today as a potential source of biofuels. The most pressing challenge, however, lies not in the production, but in the energy-efficient separation and downstream processing in a manner suitable for the production of bioproducts. Introducing a flocculation-based bulk harvesting step in the process can lower the energy needs with 10-fold. In our studies, we evaluated promising flocculation modes such as electro-coagulation-flocculation (ECF), flocculation using cationic starch and pH-induced flocculation. ECF was evaluated for a freshwater species *Chlorella vulgaris* and a marine species *Phaeodactylum tricornutum* in batch lab-scale experiments. This flocculation mode was shown to be more efficient using an aluminum than an iron anode resulting in algal recovery efficiencies > 90%. Power consumption of the ECF process was 2 kWh kg⁻¹ of microalgal biomass harvested for *Chlorella vulgaris* and 0.3 kWh kg⁻¹ for *Phaeodactylum tricornutum*. Because of the lower power consumption in seawater, ECF is a particularly attractive mode for harvesting marine microalgae. We evaluated cationic starch as a flocculant for harvesting microalgae. Our study revealed that cationic starch was an efficient flocculant for freshwater (*Parachlorella*, *Scenedesmus*) but not for marine microalgae (*Phaeodactylum*, *Nannochloropsis*). The cationic starch to algal biomass ratio required to flocculate 80% of algal biomass was 0.1, while for *Scenedesmus*, a lower dose was required (ratio 0.03). Cationic starch may thus be used as an efficient, nontoxic, cost-effective, and widely available flocculant to pre-concentrate freshwater microalgal biomass. We studied pH-induced flocculation to pre-concentrate microalgae, both for *Chlorella vulgaris* and *Phaeodactylum tricornutum*. For *Chlorella* was demonstrated that flocculation can be induced by increasing medium pH to 11. Although both calcium and magnesium precipitated when pH was increased, only magnesium (>0.15 mM) proved to be essential to induce flocculation. For *Phaeodactylum*, flocculation was induced at pH 10 but in this case, also calcium played an important role in the flocculation mechanism. The combination with precipitate formation models allowed us to predict pH induced coagulation conditions. Finally, floc characteristics (floc size, floc concentration, sedimentation velocity) and influence of the presence of algogenic organic matter (AOM) was evaluated for all modes and compared with conventional flocculation modes such as alum sulphate and chitosan flocculation. Practical implications such as cost, medium recycling and contamination risks were included in the discussion.

Functional analysis of the *Myceliophthora thermophila* C1 xylanase machinery

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Xylan is, after cellulose, the most abundant biopolymer in the plant kingdom. Its chemical structure is complex and characterized by a backbone composed of β -(1,4)-linked xylopyranosyl units containing several different substituents, such as arabinose, glucuronic acid and/or acetyl groups. These single components or partial degraded xylans/xylooligosaccharides, once released from the polymer, may be very valuable as building blocks for value added chemicals, as starting material for bio-based fuels or as functional ingredient for foods. The structure of xylan differs highly for different plant materials. Complete enzymatic degradation or modification of this structure depending on the particular application, therefore requires many different or more specific enzymes, respectively. Dyadic develops the filamentous fungus *Myceliophthora thermophila* C1 as a proprietary protein production platform for the efficient production of tailor-made enzyme mixtures for the degradation or modification of lignocellulosic biomass. This fungal strain has a high xylanolytic potential, since it is equipped with the full set of genes encoding for the complete spectrum of enzymes to degrade the complex structure of xylan. All thirteen xylanases found in the C1 genome have been produced individually in a dedicated C1 host strain, and subsequently purified and characterized. These enzymes have been classified in three different glycosyl hydrolase families (GH), namely GH10, GH11 and GH30. Results show that they all have differences in their mode of action and substrate specificities, even if they have been classified in the same GH family. Besides these thirteen different xylanases, also β -xylosidases and a large spectrum of accessory C1 enzymes, such as arabinoxylan hydrolases, α -glucuronidases and esterases, are produced and characterized. The differences in mode of action and substrate specificity for the xylanases will be discussed as well as their interplay with each other and with other xylan degrading/modifying enzymes.

Session 6

Multi-resource and multi-product biorefineries

Tuesday, April 9th 2013

Dorskamp room

A supply chain approach to biorefinery sustainability assessment

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There is a growing understanding that modern enterprises development needs being driven by a holistic assessment covering each aspect of the production and distribution network [1]. This approach, commonly refer to Supply Chain Management techniques (SCM) to steer investments on optimal networks design and planning decisions encompassing all the stages of production systems; i.e. raw materials supply, transportation, conversion and final product use and distribution [2] This kind of comprehensive analysis is of crucial importance at the early stage of new infrastructures design. In particular, this is true for biomass-based systems where preliminary integrated analyses capable of evaluating several alternative configurations may help defining a more comprehensive view of the system [3] and shed light on cost effective logistic solutions [4;5] In this context, Mixed Integer Linear Programming (MILP) represents one of the most suitable tools in determining the optimal solutions of complex SC design problems where multiple alternatives are to be taken into account [6]. This work proposes an optimisation tool to drive strategic policies and investments on biorefineries based upon the approach formulated for multi-period spatially-explicit resource-technology network modelling [7]. A MILP model is proposed as a quantitative tool to assess and optimise the upstream supply chain (i.e., analysing the network steps before the biomass conversion) of a new developing Organosolv process [8] for a wide variety of feedstocks. The optimisation model drivers are financial and environmental performance of the new biorefinery system encompassing a wide variety of crucial aspects, i.e. feedstocks mix seasonal supply, biomass geographical availability, transport logistics, biorefinery and storage capacity and location. Real-world case studies have been developed to show the efficacy of the modelling framework to drive strategic policy and investment on new biorefineries. Keywords: MILP, supply chain optimisation, biorefinery systems

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Fractionation of sustainable biomass from land and sea. Technology development at ECN

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The biorefinery concept aims at maximizing the revenue by fractionation of biomass into its main components and subsequent conversion of the fractions. Generally, the biorefinery concept consists of 1) pretreatment and fractionation of the biomass into its main constituents and 2) further processing of fractions via biochemical and/or thermochemical methods. ECN develops fractionation technologies for lignocellulosic and seaweed biomass. These are considered 2nd and 3rd generation biomass feedstocks, respectively. For lignocellulose, ECN develops organosolv-based biorefinery technology for the conversion of hardwood and herbaceous crops into 2nd generation biofuels, chemicals and performance products. ECN develops biorefinery technologies tailored for the use of cultivated seaweed biomass as a source of 3rd generation chemicals and fuels. Lignocellulosic biomass is comprised of three types of bio-polymers: hemicellulose and cellulose, which are carbohydrate polymers and lignin, an aromatic polymer. In the ECN organosolv fractionation process an organic solvent mixture with water is used to extract the lignin from the biomass. Simultaneously, the hemicellulose fraction is hydrolyzed and a cellulose-enriched pulp is produced. This high purity pulp can be used directly for paper manufacture. The pulp can also be easily (enzymatically) hydrolyzed into sugars for fermentation to biofuels or biobased chemicals. The main benefit of organosolv compared to other lignocellulose pretreatment technologies is that high-purity lignin is produced. This high purity lignin is very suitable for high-value applications. Seaweed biomass on the other hand contains no lignin, cellulose or hemicellulose. The main components are specialty carbohydrates (up to ca. 60wt%), protein and minerals. Examples of such specialty carbohydrates are mannitol, alginic acid, a valuable thickening and gelling agent, ulvans, which are bioactives, and rhamnose. The nature and contents of seaweed carbohydrates are species, season and location dependent. The proteins can contribute to alleviating the growing need for feed protein, whereas the minerals can be recycled into sustainable fertilizers. ECN is developing species specific biorefinery concepts which fractionate the seaweed biomass into proteins, carbohydrates and minerals. The carbohydrates are then fractionated into their components and can be hydrolyzed to produce sugar syrups suitable for fermentations or chemical conversion. In this manner seaweed carbohydrates can be converted to high value green chemical intermediates for bio-polymers and other applications and fuels. Our presentation will highlight the progress in development of the two biorefinery concepts. We will also discuss the complementary nature of the two raw materials. This will include results from our experimental program as well as conceptual design studies and economic evaluations.

Biorefinery of Microalgae: production of high value products, bulk chemicals and biofuels

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"Biorefinery" is a facility that integrates cell disruption, extraction, conversion and separation technologies of biomass. The biorefinery strategy is analogous to today's petroleum refinery, in which multiple fuel products and chemicals are produced from crude petroleum. Biorefinery includes the selective isolation of products (proteins, carbohydrates, lipids) from crude biomass. Biorefinery needs to be mild and efficient at the same time in order to maintain the functionality of the products (e.g. native protein conformation) and thus value. For the development of a sustainable biobased economy and to guarantee economical feasibility it is essential to use all biomass ingredients for high value and bulk product applications. Biorefinery will result in ingredients for a variety of applications (e.g. food, feed, fuel, pharma, chemicals) to cope with the worldwide scarcity of food and fuel in the coming decades. The feasibility of microalgae biorefinery production is presented from biomass concentration, cell wall characterization, cell disruption, extraction towards fractionation technologies. Although algae are not yet produced at large scale for high valuable or bulk applications there are opportunities to develop this process in a sustainable way. If biorefinery of microalgae is applied, lipids should be fractionated into lipids for biodiesel, as building blocks for the chemical industry and poly unsaturated fatty acids for the food and health market. Whereas the fractionated functional proteins and carbohydrates can be applied for food, feed, health and bulk chemicals. If in addition production of algae is done on residual nutrient feedstocks and CO₂, production of microalgae at large scale against low production costs the biorefinery approach for micro algae remains feasible. In this presentation case studies will be presented showing the progress on the different biorefinery stages for microalgae.

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Open innovation platforms on sugars, fatty acids and proteins: developing the high value products biorefineries

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By 2030, the role of the biobased economy is expected to grow significantly in Europe. The European biorefinery sector will evolve from the current industrial biorefinery operations known for products like (food, feed, biofuels, paper and board), to a broader, more mature sector. In 2030, the biorefineries will use a larger variety of feedstock and produce a wider variety of end-products compared to the current situation. Achieving this European Biorefinery vision will require a higher degree of integration, flexibility (multi feedstocks and multi products) and improved sustainability of the future biorefineries. To be able to address the challenges of future biorefineries, IAR cluster (France) and its members have developed an innovation ecosystem based on, among others, three open innovation platforms: BRI, PIVERT and IMPROVE. Each platform is dedicated to one type of feedstock: sugars for BRI, vegetal oils for PIVERT and proteins for IMPROVE. The whole approach is based on the biorefinery concept, playing on the different feedstock and platforms complementarities and taking into account the whole biomass valorisation, based on a zero-waste concept. These platforms will focus on developing competitive specialty and high value biobased products, such as food ingredients, chemicals and materials. BRI (Biorefinery Research and Innovation) is an open innovation hub focusing on biorefining and industrial biotechnology. BRI brings together the industrial biorefineries (sugar, starch & glucose) of Bazancourt-Pomacle (near Reims, F), academic partners, an applied R&D centre (ARD) and an open pre-industrial prototype (BioD emo). The platform aims at developing biotech processes from lab to pre-industrial scale. PIVERT is a centre for research, innovation, experimentation and training focused on oilseed biomass chemistry. With a provisional budget of about  246 million, the programme will involve more than 150 researchers over a 10-year period. It will also include the development of a flexible pilot centre where thermochemical, fermentation and chemical treatment will be available to design tomorrow's processes of valorisation of the entire oilseed biomass. PIVERT will develop the foundations of the oilseed biorefinery of the future. IMPROVE will focus on the valorisation of proteins. The aim is to create a shared platform dedicated to sustainable innovation in vegetable proteins. It will develop new features and technologies in markets such as food and feed positioning customers and partners in new target markets with strong growth potential: green chemistry (such as polymers, surfactants) and cosmetics. IMPROVE will offer a combination of high added value and unique facilities and expertise in a dedicated space, fostering innovation.

Biorefinery concepts from earth to space

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Through the production of multiple products with minimal waste streams, a biorefinery makes use of the differences in biomass components to maximize the value derived from the biomass feedstock. Biorefineries can be built on different platforms. The most cited ones are the thermochemical syngas platform and the biochemical sugar platform. More recently, a third platform emerged. Li and Yu (2011) describe two-stage bioconversion processes to achieve stepwise and directional substrate conversion. Organic matter is transformed into volatile fatty acids (VFAs) and other intermediates in a primary processing stage by mixed cultures. Subsequently, the VFAs are converted into bioenergy or biochemicals in a fine processing stage. In this abstract, two examples of biorefineries based on the carboxylate platform are presented. A first example investigates the synergies between waste, wastewater and energy to increase the energy efficiency of sewage treatment plants. In the SewagePlus concept, the organic fraction in sewage is concentrated and/or enriched because this allows the implementation of reductive technologies with production of energy carriers instead of energy consuming aerobic ones. Various technologies have been investigated at laboratory- and/or pilot-scale. The most innovative one consists of an acidification step yielding carboxylates, followed by the production of bio-electricity in microbial fuel cells. In this presentation, an overview will be given of future-proof concepts for sewage treatment, with emphasis on the scale of products that can be obtained and the associated research results and calculations performed. A second example concerns MELISSA or Micro-Ecological Life Support System Alternative. For long-haul manned space missions, the supply of oxygen, food and drinking water is a major challenge. Bioregenerative Life Support is the only approach that allows for maximal recovery of resources from the wastes produced by the astronauts. An international consortium of 11 partners including the European Space Agency, is working on the demonstration of the MELISSA concept, which consists of 5 interconnected biological compartments to essentially transform organic waste into food. In this closed loop, both low-value high volume and high-value low volume chemicals are produced. The first category includes carboxylates produced in Compartment I, the second one a cholestering lowering agent in the biomass of Compartment II and nutraceuticals and pigments in the microalgae present in Compartment IV. Acknowledgments The authors acknowledge the Environmental and Energy Technology Innovation Platform (MIP) for financial support to the SewagePlus project and Belgian Science Policy to MELISSA-related activities in Belgium.

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Session 7
New food and feed
ingredients from
biorefinery

Tuesday, April 9th 2013

Ir. Haak room

Biorefinery concepts aimed at healthy and sustainable food products

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Increased use of plant components in modern diets is a promising route to make food production more efficient, due to the fact that the production of plant based product requires far less resources than the production of animal-based products. In addition, plant proteins will become available in large quantities because of the expected increase in biofuel production. However, for optimal usage in food products, the proteins should be native, but not necessarily completely pure as current protein isolates.

The formulation of new plant-based alternatives strongly depends on the purpose of the replacing product. This can be illustrated with meat as an example. Apart from water, meat contains mainly protein and fat. Health benefits are expected when part of the meat is replaced through plant-based products that contain dietary carbohydrate fibers in addition to the protein. Plant materials, such as soy beans, are mixtures of proteins, fats and carbohydrates naturally. Keeping the natural structure in tact as much as possible through mild and selective fractionation can lead to several advantages with respect to nutritional value. Besides, the presence of carbohydrates in a protein matrix give rise to additional structuring options for making new products. Now, plant materials are decomposed to almost pure ingredients, in processes using copious amounts of water, energy and chemicals. When making the product, the pure ingredients are combined to obtain the final composition. This is clearly not efficient.

We therefore present an alternative approach to decompose plant materials aiming at the production of functional fractions. The use of those fractions is a method to increase the efficiency of food production, to provide additional options for creating new product structures and to give opportunities for creating plant-based alternatives with nutritional benefits. Main focus will be on dry and concentrated fractionation processes. The by-products of these fractionation processes can then be further processed into biofuels

Dry Fractionation for Production of Functional Pea Protein Concentrates

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Wet fractionation is the common route to isolate proteins from legumes and cereal grains. However, major drawbacks of this process are use of copious amounts of water and energy. Moreover, due to the harsh conditions, native functionality of proteins is lost. An alternative to wet fractionation is dry fractionation by fine milling and air classification. During fine milling the larger starch granules are physically disentangled from the smaller protein-rich particles to allow optimal separation during air classification. In our research we investigated milling and air classification as an efficient route for production of protein pea concentrates in combination with functional analysis of protein concentrates. Specifically, retention of native functionality of proteins could make the use of also less pure protein concentrates very attractive (Schutyser, 2011). Jet and impact milling were evaluated to produce flours with different particle size distributions as a function of air flow rates and classifier wheel settings using a pilot scale milling and classification facility. Milling to small average (7 μ m) particle size corresponded to a lot of damaged starch and poor yields, whereas milling to large average particle size (20 μ m) resulted in poor disentanglement of starch granules and protein bodies. Milling settings were selected to produce flour with an optimum particle size (13-17 μ m) and this flour was subjected to air classification using various settings. A range of protein concentrates were produced with a maximum protein content of 55 w/w%. Results from the air classification were compared to theory that related protein content and air classification settings providing a specific cut off size for separation. Finally, functionality of the protein concentrates was evaluated using water hydration capacity tests. It was found that dry fractionated concentrates (55 w/w%) had limited water hydration capacity compared to wet fractionated isolates (90%), which could be explained from the native functionality of the proteins present in the concentrate prepared by dry fractionation. Concluding, dry fractionation is an efficient method to obtain pea protein concentrates with interesting functional properties.

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Bioactive food ingredients from agricultural by-products using green technologies

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Annually hundreds of tons of the agricultural raw materials are regarded as waste or by-products. Generally, these by-products are rich in phytochemicals that could be further utilized. Bioactive compounds from these raw materials e.g. phenolic compounds, anthocyanins, carotenoids and seed-oil can be used as ingredients on formulation of functional foods, nutraceutical or in the cosmetic industries. This has resulted in the increase interest in the use of agricultural by-product to extract high-value compounds that would otherwise be wasted. Extraction of these compounds, traditionally, requires the use of organic solvents and intensive heat treatment making the produced extracts of poor quality for further use as ingredient in functional food formulation. The use of green technologies such as pressurized fluid extraction [1] and supercritical fluids extraction [2] provides a promising alternative for the conventional extraction technologies, it is regarded as environmentally friendly over other conventional either solvent or enzyme extraction methods for recovering natural products. It is suitable extraction and fractionation for the food and pharmaceutical industries [3, 4]. The aim of this study is the shed the light on the possibilities of extracting bioactive compounds from agricultural by-products using green extraction technologies, stabilize and formulate the extracted compounds into food ingredients and to produce food products with functional properties.

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RubisCo: from Refinery to product Functionality

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Bio-economy, crucial part of the sustainability concept, refers to the maximal use of natural resources. An optimization between land use and benefits in total economy ranging from Food, performance materials to Energy. Food security, especially protein security, is a major issue. How to feed the world with highly nutritious foods, with minimal impact on natural resources such as land use and ecology. The most abundant protein in the world is RubisCo since it is the major protein in green plant-material. It has a good amino acid profile for nutritional application. Green plants like spinach, grass, sugar beet leaves, microalgae and alfalfa can produce more protein per m² than for example soy and other grains. However, application in food is minimal, mainly due to biorefinery and application issues. Based on our long history in protein technology, we addressed these issues by firstly developing a refinery process resulting in a colourless, low flavour protein isolate (>95% w/w) with highly soluble RubisCo proteins. The process was scaled up to pilot plant size and we showed the impact of spray drying (industrial scale) of these proteins. We will show results on the unique functionalities of this protein isolate in relation to industrial application, such as foaming capacity, gelling and emulsifying properties, but also nutritional aspects. Take an algal refinery for example. Renewal Algal Energy, LLC (RAE) has developed a novel and sustainable process to produce algae on large scale for biofuel, leaving proteins and other materials for alternative utilization. The resulting algal biomass is highly desirable for a range of animal feeds, but high-value products can be recovered from the biomass for nutraceutical application. An adjusted process was developed to extract proteins from the algal biomass that resulted in protein products that met the required properties for the targeted nutraceuticals market.

Autohydrolysis processing of oil palm by-product: xylooligosaccharides production and prebiotic potential

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Oil palm (*Elaeis guineensis* Jacq.), a tropical perennial crop, is an important commodity for palm oil production, mainly in South East Asia. Hemicellulosic-rich empty fruit bunches is generated as a by-product (20%) in every tonne of fresh fruit bunches being processed for palm oil. Currently, these materials are mainly used for mulch and others non-food applications. As these residues have up to 28% (dry weight basis) hemicelluloses and the xylan account for 75% of total hemicelluloses in this material, they could be potentially used for oligosaccharides production as a functional food ingredient. In relative to some cereal-based agricultural residues, the chemical characterisation of oil palm empty fruit bunches (OPEFB) fibre showed interesting compositions, being low in ash content and contained substantial acetyl groups, making autohydrolysis treatment a suitable option for the production of xylooligosaccharides (XOS) by allowing the treatment operation to be carried out at milder conditions. This process is generally considered as environmentally friendly as only water is used as the liquid medium during the treatment. The fibre of OPEFB was treated with non-isothermal autohydrolysis treatments at different operational temperatures (150-220°C) and with liquid to feedstock ratio of 8 g/g. The yield and compositions of the liquid and solid phases showed dependent on treatment severity (log Ro). Highest XOS concentration (14.84 g/100 g OPEFB fibre) which encountered for 63.0 % of feedstock xylan was obtained at a severity factor of 3.91 (210°C). The lignin and glucan content on the other hand was largely preserved while the hemicellulose fraction was being solubilised, thus the solid residues could be further utilised for other value-added product. This treatment however, produced some impurities in the reaction liquor which are mainly monosaccharide (12 %) and acetic acid, as well as other sugar degradation products such as furfural, 5-hydroxymethylfurfural and formic acid. Crude autohydrolysis liquor was purified using gel filtration chromatography technique and then freeze dried. Six mixed XOS fractions with average degree of polymerisation (DP) of 4, 7, 14, 28, 44 and 63 that were obtained is then used to evaluate the influence of molecular weight on the fermentation of xylooligosaccharides by human gut microbial populations. These purified fractions had purity of 75-83 %, of which 79-85 % was XOS. In the batch culture fermentation, oligofructose (OF), which is a known prebiotic, was used as a positive control and they were also compared with the commercial XOS. A considerable increase in short chain fatty acid concentration was observed in XOS fractions with average DP 4-14 compared to commercial XOS and OF at 24 and 36 h fermentation. Most of the XOS fractions were shown to be bifidogenic at 10 h of fermentation though there is a trend for the *Bifidobacterium* populations to increase with the decrease of molecular weight.

Alcorefinery of microalgal biomass - mild treatments integration for pigments and proteins fractionation from *Chlorella vulgaris*

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Chlorella vulgaris is a well-known green microalga. It is an interesting source of pigments (chlorophylls and carotenoids) and proteins that represents more than 10% and 50% of the biomass respectively (dry weight basis). The present work investigated a mild treatments integration for pigments and proteins fractionation directly from wet biomass. As the cell wall of *C. vulgaris* is quite resistant, it to and make the cell disruption step a key point for intracellular metabolites recovery. The biorefinery process was applied at a pilot scale. It started with a photobioreactor content (130 L at 0,5 g/l) or the harvested biomass (by centrifugation up to 5-10 g/l). The microalgal suspension was treated with a high pressure cell disrupter (operating pressure up to 2.7 kbar) in order to achieve complete cell breakage. Then the aqueous phase was put into contact with heptane in a Centrifugal Partition Extractor. This technic uses centrifugal acceleration to keep the solvent stationary while the feed is pumped through it. 70-80% of pigments could be extracted in less than 3 min without a preliminary dewatering step. An alkaline treatment was then applied to the aqueous residue for efficient protein solubilisation. The highest yield was 50.0±4%, when this operation was done at pH=12. The applied pressure for cell disruption appeared to increase the yield of proteins solubilisation. The proteins were quantified in terms of nitrogen contain using Kjeldahl and Lowry procedures. Molecular mass distribution was obtained using one and 2D electrophoresis. The 2-DE profile of extracted proteins (SDS-PAGE) showed a high diversity in protein composition; majority of proteins were separated in the apparent molecular mass range from 12.0 to 75.0 kDa . The bands with the highest intensity represented proteins with a molecular mass of about 25.0 kDa and 52.0 kDa. The major group had a pI range between 4.0-6.0 with a second group with higher pI (6.0-8.0). All the processes were conducted at room temperature. The cell disruption conditions were determinant for the post-processing efficiency. The proposed scheme seemed to be an interesting way to fractionate polar and non polar compounds and to optimize proteins recovery without any drying step. Perspectives would be discussed concerning the treatment of more concentrated biomass and necessary developments for scaling-up. This work was realized in the framework of a french research program ALGOREFINERY supported by the National Research Agency.

Session 8
Chemicals, Fuels and
Materials II

Tuesday, April 9th 2013

Dorskamp room

Growing Bioeconomy - Alberta Activities and Capacities

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Alberta is uniquely positioned with a large oil and gas industry and a wealth of agriculture and forestry resources. Alberta is focusing on growing its bioeconomy through diversification to enhance balanced and sustainable growth. This presentation will highlight Alberta Government activities to support bioeconomy initiatives in the areas of feedstock improvement, biomass conversion and product development. This presentation will describe work in Alberta around technology innovation and commercialization which has resulted in new bio-opportunities in fuels, chemicals, materials and food. Examples include the Canadian Triticale Biorefinery Initiative, the bio-polymer pilot plant and the Alberta Biomaterials Development Centre. Capabilities of the Food Processing Development Centre, AgriValue Processing Incubator and Agri-Food Discovery Place will be highlighted. The presentation will then describe Alberta's agriculture and forestry resource endowments as well as the policy landscape and programs designed to support the bioeconomy. Finally, some high level information on what makes Alberta an attractive place to do business will be presented.

How to develop a Profitable and Sustainable Biorefinery? - Success by maximizing value and being resource efficient

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Creating value is a core task for any business. The Arizona Chemical value creation model builds on offering the customer a green and innovative product with a high functionality, based on a profitable and sustainable biorefinery business. We believe that we know what we talk about when it comes to developing biorefinery business. The Arizona Chemical facilities have run modern refineries for products derived from trees for over 80 years. We have a proven track record of commercializing new products and bringing value to the customer. And we are truly market-driven with an “outside-in” perspective on business. Key factors contributing to the Arizona Chemical’s successful biorefinery business are maximizing value of green products for their applications and being resource efficient at the same time. Arizona Chemical applies its value creation model in each of its four focus markets. To the Adhesive market, Arizona Chemical supplies biobased resins for tack and adhesion in high performance end applications. We also offer tackifiers, which extend the applications of water based adhesives instead of solvent based ones. To Tire producers, we provide tread enhancement additives that work with the tire to improve traction and breaking distance. Our additives improve the polymer-filler interaction and thus lower the rolling resistance of the tire and save fuel. In the Roads & Construction sector, our innovations allow higher use of recycled material in pavements rather than being thrown into landfills, which earlier was often the case. Our resins also increase the durability and luminosity of road markings. Arizona Chemical also provides numerous sustainable Chemical Intermediates to enable many industries such as coatings, lubricants, metal working fluids, fuel additives, mining and oil field to go green. To achieve the resource efficient, low carbon economy, based on a profitable and sustainable biorefinery processes, Arizona Chemical advocates the implementation of four fundamental principles: 1. A natural raw material shall always be used to its full potential through cascading use 2. Sound business is based on market economics, not on incentives 3. Focus shall be set on creating highest possible value 4. A level playing field shall exist between users of the same resource The importance of the four principles can be exemplified with a real life example. Arizona Chemical’s main raw material is Crude Tall Oil (CTO), which is a co-product of kraft pulping. CTO is a liquid biomass, which is scarce, i.e. its supply is limited. A well-established application is the one Arizona Chemical represents, i.e. biochemicals. The presentation includes: • An outline of the Arizona Chemical value chain • Biorefinery business – how to develop a profitable and sustainable business model? • Market focus areas – why have they been chosen and how do we interact with our customers to understand their needs? • Green value creation – how is it done in real life?

Biomass pretreatment using superheated steam as a starting point for biorefining

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Lignocellulosic biomass is abundant worldwide, however, to use such biomass in biorefineries the lignocellulosic complex should be broken. It is known that this complex can be broken under acidic conditions at high temperatures. In case heating is carried out by steam injection, most of the time such steam is stagnant and saturated. The use of superheated steam (SHS) that flows through the biomass is an alternative with several advantages. By passing steam through heaps of e.g. straw an efficient and uniform heat transfer is established. Tests were carried out with wheat straw and reaction conditions were varied. The straw could be successfully pretreated within a few minutes at dry matter concentrations between 30% and 65%, steam pressures of 6 bara and temperatures of 160-180°C. High dry matter concentrations reduce heating costs and required acid concentrations are rapidly reached because of the low amount of water present. In addition, high biomass concentrations are favorable for the economy of subsequent conversion processes and downstream processing. A large part of the volatile compounds such as furfural was stripped from the biomass, thus reducing toxicity of the biomass. During superheated steam treatment hemicellulose was 90% hydrolyzed into its monomers, which could be recovered by extraction. The residual cellulose/lignin mixtures could be used in fermentation processes and in paper making. Alternatively, the complete SHS pretreated straw, including the hemicellulose fraction could be used in a fermentation process as well, postponing the product fractionation to a moment after the fermentation process. SHS makes the cellulose well accessible to catalysts and reactants. E.g. after cooling and neutralization the cellulose could be 90-95% converted into glucose by the use of enzymes. Pretreatment using SHS can be carried out in conventional steam dryers, with some modifications. In such systems the continuous feed in and out of relatively dry pieces of biomass through the superheated steam process, against a pressure drop of 6 bara, is feasible.

3-Hydroxybutyrate Production BY GENETICALLY ENGINEERED ESCHERICHIA COLI

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The production of bulk chemicals from renewable resources is a promising alternative to replace current petrochemical production routes. One of the alternatives to replace fossil based plastics is poly-3-hydroxybutyrate (PHB), a bioplastic naturally produced as a carbon and redox sink by a broad range of microorganisms. The ability to produce PHB has been transferred to different hosts, including *Escherichia coli*. When precise physicochemical characteristics are important in the target plastic, chemically polymerised PHB plastic is more tunable and reproducible than polymerisation in the microbe itself. Therefore, the microbial production of 3-hydroxybutyrate (3HB), the monomer of PHB, may be a better alternative. Apart from serving as a building block for the biopolymers/bioplastics industry, 3HB is also an interesting compound for the medical industry, for example for biodegradable sutures, bone plates, surgical mesh, pins, stents, etc, and for the fine chemical industry for example for vitamins and antibiotics. For all these applications the chiral purity of the 3HB is important.

Microbial fermentation is a convenient way to convert biomass into enantiomerically pure 3HB monomers, since a multitude of biochemical reactions can be catalysed simultaneously by one organism in one reactor with the help of stereoselective enzymes. Microbial processes can be either aerobic or anaerobic. Aerobic processes generally have lower productivities than anaerobic processes because of rate-limiting volumetric oxygen transfer. Yields are also lower in aerobic processes since part of the substrate is completely oxidised to CO₂ or converted into microbial biomass with high efficiency. To compete with petrochemical processes, both high productivity and yield are important. Therefore, we want our 3HB production process to be anaerobic.

Because product recovery is one of the most costly steps in producing products with a high water solubility and a high boiling point, like 3HB, we want to decrease these costs by producing 3HB in a mineral salts medium as compared to a complex medium, so it can be more easily purified. Achieving a pure product is important for later polymerisation steps.

In this study we metabolically engineered *E. coli* to produce both (*R*)-3HB and (*S*)-3HB in mineral medium under anaerobic conditions.

The use of biosourced phenolic compounds as a sustainable alternative to bisphenol A in the field of materials

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Epoxy resins constitute a major class of thermosetting resins and are extensively used in non-food and food packaging applications. These copolymers are elaborated from two different chemicals, a prepolymer bearing two or more epoxy groups per molecule and a curing agent (hardener). The most common epoxy resins are industrially produced by the reticulation of the glycidyl ether of bisphenol A (DGEBA) with polyamines, anhydrides or polyacids as hardeners. The negative impact of bisphenol A on human health and environment combined with the depletion and high cost of fossil resources impose to find an alternative source of phenols which is safer and sustainable. Natural polyphenols which are ubiquitous secondary metabolites in the plant kingdom can be used as phenolic resources for developing novel bio-based materials¹. Present in significant amounts in fruits and vegetables, they are daily consumed in our diet (in both fresh and processed foods). The polymerized forms are generally the most abundant source of polyphenols encountered in plants: lignins, condensed and hydrolysable tannins. Lignins are difficult to extract and to use for chemistry. In relation to epoxy resin application, condensed tannins are more readily depolymerized and have more phenolic hydroxyl groups. The first part of this project was to demonstrate the feasibility to synthesize epoxy resins from a constitutive monomeric building block of condensed tannins by chemical and chemo-enzymatic synthesis methods². Then, the quality of the epoxy resin obtained from the natural polyphenol was compared with that of the resin obtained from DGEBA. The properties of the resin containing the natural polyphenol are at least equal to those of the industrial epoxy resin. These results encouraged us to go further in this project by testing the synthesis of epoxy resins from industrial extracts of tannins. This stage is crucial for determining whether a preliminary step of depolymerization is required or not for the glycidylation of tannins.

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What is in store for bio-based polyamides?

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In recent years, Evonik has developed and is pushing a line of bio-based polymers under the trade name VESTAMID® Terra. Currently three products are available within this group of polyamides that are partially or entirely based on renewable feedstocks: VESTAMID® Terra DS, HS and DD. The castor bean plant (*Ricinus communis*), with its oil-based monomer derivatives, form the carbon backbone of these products. For example, Terra "DS" (a PA10,10 grade) is polymerized by 1,10-decamethylene diamine "D" and sebacic acid "S", which are both directly sourced from the castor bean. To guarantee not only the bio-based sourcing aspect, but also sustainable practices, a detailed life cycle assessment (LCA) has been conducted. Compared to similar polyamides (such as nylon 12 and 6,6) these products can greatly reduce greenhouse gas emissions by 42% up to 56%. It is also expected that these values will drastically increase as these bio-based products evolve from a novel to a mature business. In this respect, and in correlation with the market developments, customers are placing heightened importance on ecological certifications and bio-labelling. Yet, despite this trend, the main market pull of the VESTAMID® Terra line remains their superior properties. Technically speaking, this group of polyamides occupy the gap between the highperformance polyamides (such as 6,12 and 12) and the standard polyamides (such as PA 6 and PA 6,6). They are renowned for low water uptake, high temperature deflection, high dimensional stability, extreme chemical resistance, and other such properties which facilitate their use in demanding application areas. This makes them unique in the field of bio-polymers, as most others are focused on low-end applications, which include but are not limited to features like biodegradability. Thus in the traditional sense of the word "sustainable", Evonik's VESTAMID® Terra line are products that last while providing high performance with the added benefit of a reduced ecological impact.

Session 9

Small scale biorefineries

Wednesday, April 10th 2013

Ir. Haak room

Small beats large

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Currently, we are moving from a fossil-based economy to a bio-based economy. To enable this transition many different processes for biorefinery are being developed. Small scale biorefinery processes can be beneficial, not only socially and ecologically, but also economically. A smart and integrated process design can beat the advantages of economy of scale applied in large scale processes. The most important factor in design of small scale biorefineries is local re-use of materials, like water, minerals, organic matter, CO₂ and heat. This minimizes costs for recycling and transport. Examples of fully operational small scale systems will be given to illustrate our theory and specific design rules for such small scale biorefineries are defined. We will show that production of ethanol and starch is feasible on small scale. The focus in design for small scale processes should be on costs for investment, specifically costs on heat exchange. Moreover, it is shown that separation of relatively simple pre-processing at small de-centralized and more difficult processing at large centralized factories respectively is advantageous. Integration with a biogas and combined heat and power (CHP) unit will furthermore allow usage of residual material to produce the energy needed in the process.

A decentral approach for sustainable supply with raw materials and energy - Fraunhofer Innovation Cluster »Bioenergy«

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Worldwide, immense amount of wet biomass containing lignocellulose are generated decentralized. The spectrum ranges from grass and green waste via harvesting residues such as rapeseed straw, processing scraps from agricultural and silvicultural production to biowastes from private households. Today, wet biomass is utilized less intensely. The reason, on the one hand, is its high water content which results in a low calorific value (low energy density), and which makes its transport and storage more expensive. In addition, the inhomogeneity, the ash content and the bad processability of wet biomass make its utilization considerably more difficult. Quite often, biogas plants are not efficient for the conversion of the difficult to biodegrade biomass containing lignocellulose. Other utilization concepts are often not available. However, wet green biomass has huge potential since it can e.g. preferably be ideally coupled with food production in crop rotation. Fraunhofer UMSICHT has initiated the Innovation Cluster Bioenergy with the objective to increase the available and utilizable amount of biomass. Over the course of the 4-year project duration, new conversion technologies are to be developed that tap into the potentials of stalk-like as well as wet lignocellulose-containing biomass. To reconcile the food production and biomass utilization and to not create a utilization competition is the overriding objective of the cluster. In this context, new compatible options for the energy and raw materials supply are to be created and export technologies for a growing technology world market will be provided. In the talk, the concept a decentralized processing of wet and/or lignocellulosic bio-mass to intermediate products is presented, together with exemplarily outcomes of the technology development that might show the potential of the decentralized approach concerning feedstock availability, economic aspects, product processing and sustainability. The different technologies for biomass processing are developed in three main thematic areas: 1. Conditioning of wet biomass: technologies for mobile dehydration of wet bio-mass and for fractioning of press juice on the basis of press and membrane technologies 2. Decentralized biomass processing: development of a technology for mobile production of biogenic crude oil in proximity to the harvesting, based on thermochemical conversion, the ablative flash-pyrolysis 3. Semi-decentralized biomass processing: development of a method for station-ary use of wet biomass and residues based on hydrothermal carbonization (HTC)

A comparative supply chain sustainability evaluation of mobile pyrolysis plants and pyrolysis-based bio-refineries

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Operational and economic trade-offs arisen in different steps in the bio-refinery (BR) supply chains (SC) guide the decisions of both plant scale and location and biomass collection routes. In particular, integrating a mobile pyrolysis plant into a BR SC raises the interest on using decentralized regional biomass. This paper compares different BR SC design options with a focus on the installation of mobile pyrolysis plants and centralized or decentralized collection of biomass. The aim of this paper is to answer the following questions: (1) Is a mobile pyrolysis plant economically and environmentally sustainable at regional level?; (2) How does a regionally centralized pyrolysis-based BR compare to a mobile pyrolysis plant in terms of economic and environmental sustainability?; (3) What are the crucial factors influencing the trade-offs between aforementioned options?; and (4) What are the governance implications of these different BR SC design options? The empirical context of this paper is based on a scenario analysis for processing lignocellulosic biomass -particularly landscape wood, reed and roadside grass- available in the Overijssel region (east Netherlands). In particular three scenarios are compared: (1) Two mobile pyrolysis plants process the locally available biomass on-site and the obtained pyrolysis oil is sent to a regionally central pyrolysis-based BR for upgrading (heating and/or electricity); (2) Local biomass is collected and transported to a centralized pyrolysis-based BR in the region; (3) A mobile pyrolysis plant performs the on-site conversion and the obtained pyrolysis oil is provided to an oil refinery outside of the region (Rotterdam). We adopt a combined optimization-simulation approach to evaluate the scenarios taking into account the variables influencing the SC performance. We use the European Corine land cover data and the Dutch Top 10 and Top 25 vector data for their applicability in extracting weighted average centre points of landscape wood and reed. These centre points will be used as input for vehicle routing and optimizing the location of the BR. Data provided by Rijkswaterstaat (Dutch Ministry of Infrastructure and Environment) in ArcGIS files will serve us to quantify where and how much roadside grass biomass is potentially expected along roadsides. The social implications (e.g. employment created) of the scenarios will be further assessed to assist governments to develop sustainable policies. Moreover, variation analysis, in which combinations of technical and organisational options are studied, will be adopted to identify coherent development options for governments. The expected results of the paper would propose practical and managerial contributions for BR SC actors and investors regarding the economic feasibility, plant-scaling, and biomass collection. Moreover, it provides several governance implications for regional governments via lessons learned from the case example.

Small versus large scale microalgae production

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Microalgae are an attractive source for fuel production. Commercial large scale algae cultivation for biofuel production, however, does not yet occur but is expected for the near future. One of the current challenges is how to design and organise large-scale algae biofuel facilities which include a series of successive processing steps. Biomass cultivation, which may occur in various reactor designs, is the first step and depends strongly on the local weather conditions. The construction of algae cultivation plants in sunny regions seems from this point of view to be preferred. Planning a region for algae biofuel plants, however, does not solely depend on the local weather conditions for cultivation; the availability of water, nutrients and land can also be limiting. The size of a cultivation plant depends therefore on a combination of regional characteristics like climate, land & resource availability and infrastructure. Large plants require large quantities of resources that can only be supplied if the infrastructure satisfies, while small plants are preferred in area with a limiting water and nutrient supply. It is therefore essential to integrate algae cultivation and supply chain logistics to determine limiting factors, to find appropriate production plant sizes and to quantify the effect of regional characteristics and design on process feasibility. In this work, models on algae cultivation using various reactor designs were integrated with logistic models for transport towards and from the algae cultivation plants. The objective of the integrated approach is to produce algae with the lowest energy requirement for cultivation and transport of resources. The approach allows studying the suitability of regions for large scale production of algae biofuel, based on the climate conditions, the availability of resources and the existing infrastructure. The focus in this work is on salt water and CO₂ supply for algae cultivation and the local use of biodiesel. In the presentation we will show the effect of regional characteristics on the size of algae plants, the logistic supply of resources and infrastructure, and on the energy requirements using various scenario studies.

Acknowledgements This work was initiated at IIASA during the Young Scientists Summer Program (YSSP) 2011 with funding from the Netherlands Organisation for Scientific Research (NWO). The research was performed as part of the program of Wetsus, centre of excellence for sustainable water technology (www.wetsus.nl), with funding of the Dutch ministry of economic affairs.

Session 10

Valorisation of knowledge and technology

Wednesday, April 10th 2013

Dorskamp room

Bio Base Europe: Open innovation and education center for a sustainable biobased economy

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The bio-based economy is strongly developing today as a consequence of the strong price increase for fossil resources such as petroleum, the drive towards sustainable production processes and to reduce the emission of greenhouse gases such as CO₂. The combination of these factors causes a strong penetration of biobased products and processes in a multitude of industrial sectors, particularly in the chemical industry, the energy sector, and the agro-industry.

In Europe, the development of the biobased economy is seriously handicapped by a number of problems. First of all there is a serious gap in the innovation chain, caused by the lack of pilot and demonstration facilities. These facilities are required to scale up a process from a laboratory setting to an industrial production plant. The lack of pilot facilities for biobased processes seriously limits most industrial and academic players to realize their plans and to valorise their knowledge. As a second problem, there is a general shortage of well-trained process operators with experience in biobased processes. Apart from a generally decreasing interest for technical professions by youth, the problem is reinforced through the lack of specific training facilities for biobased activities.

Bio Base Europe is a joint initiative of the European Union, Belgium and the Netherlands to alleviate these problems. These have now united their forces in order to speed up the development of a sustainable biobased economy in Europe. Bio Base Europe has built research and training facilities for the biobased economy with an overall budget of 21 M€. The Bio Base Europe Pilot Plant is a flexible and diversified pilot plant, capable of scaling up and optimising a broad variety of biobased processes up to the 15 m³ scale. The pilot plant contains fermenters and reactors for green chemistry, as well as a variety of up- and down-stream processing equipment. The pilot plant is a one-stop-shop, capable of performing the entire value chain in a single plant, from the green resources up to the final product. The Bio Base Europe Pilot Plant is a completely independent facility that is operating according to the open innovation model. As such, the pilot plant can be used by all players of the bio-based economy to speed up their innovation process and bridge the valley of death.

The Bio Base Europe Training Center houses a number of training facilities for biobased activities, and is operating according to an open education model. Companies as well as schools and universities are offered tailor-made training programs for their personnel or students.

An overview is given of Bio Base Europe, an important building block for the development of the biobased economy in Europe. This research and training infrastructure is expected to improve economic growth, innovation and sustainable development. This will lead to a strong innovation dynamic, a flood of new projects, diversified contacts, networking and collaboration and in general a reinforcement of the open innovation and education model for biobased activities.

More information can be found on www.bbeu.org

Standardization: codifying and disseminating state of the art technology and best practices to the market

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The growing interest for the substitution of fossil resources with biobased alternatives has triggered a rapid growth of innovative technologies, and new biobased materials and products. The high level of innovation creates a very dynamic market for biobased products, in which companies are confronted with a large number of uncertainties. These may limit new products and technologies from growing into full scale commercial products. The market needs to consolidate the state of the art. Thereby developing a solid basis for commercialisation of new technologies and products and creating a point of reference as a basis for further research. Development of standards in close relation with research is essential to create levels of stability in this market, while still supporting innovation and development of new technologies. The European Commission shares this vision and has submitted several requests (mandates) to CEN, the European Standardization Organization, for the development of standards in the field of biobased products. Based on these mandates, CEN initiated a new Technical Committee: CEN/TC 411 "Bio-based products". This CEN/TC, with a Dutch Chair (DSM) and Secretariat (NEN) has committed to the development of a comprehensive set of standards for biobased products. It covers subjects like terminology, determination of bio-based content, sustainability criteria and lifecycle analysis (LCA), as well as product-related (vertical) standards for bio-based solvents, including bio-degradability. The close connection with research is arranged via the project KBBPPS (Knowledge Based Bio-based Products' Pre-Standardization) which started under the FP7 EU research programme. Led by NEN and involving several Dutch and other European research institutes, the project executes pre- and co-normative research for bio-based products, to be used directly in the CEN standardization process on the same topic. The presentation will give an overview of the subjects in the field of biobased products for which European standards are being developed. Detailed examples will be discussed of how standards are used as guidance for research and how the research results are being incorporated in new and improved standards for the market.

Green extraction of natural products as tools for biorefinery

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This presentation will introduce a new and innovative area in the frontiers of chemistry, biology and processing: green extraction with special emphasis on natural products. Green extraction is a part of the sustainable development concept; its history, concept, principles and fundamentals will be described. We will pay special attention to the strategies and the tools available to make biorefinery greener. The representation will present the innovative research in this area these past five years in term of innovative techniques (microwave, ultrasound, pulse electric field...) and alternative solvents (ionic liquids, sub and supercritical fluid, agrosolvents, water...) applied to this new area green extraction of natural products with special examples applied to biorefinery concept. With the increasing energy prices and the drive to reduce CO₂ emissions, chemical and food industries are challenged to find new technologies in order to reach a significant improvement in energy efficiency. Extraction technology is one of the promising innovation themes that could strongly contribute to sustainable growth of the European economy. Extraction is one of the key technologies of the process industry, often requiring up to 50% of investments in a new plant and more than 70% of total process energy used in food, fine chemicals and pharmaceutical industries. A general definition of green chemistry is the invention, design and application of chemical products and processes to reduce or to eliminate the use and generation of hazardous substances. In relation of green extraction of natural products, this definition can be modified as follows: "Green Extraction is based on the discovery and design of extraction processes which will reduce energy consumption, allows use of alternative solvents and renewable natural products, and ensure a safe and high quality extract/product". The listing of the "six principles of Green Extraction of Natural Products" should be viewed for industry and scientists as a direction to establish an innovative and green label, charter and standard, and as a reflection to innovate not only in process but in all aspects of solid-liquid extraction. The principles have been identified and described not as rules but more as innovative examples to follow discovered by scientist and successfully applied by industry.

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Integrated valorisation of biomass

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This contribution summarizes the results of a recently finished study in The Netherlands on the future development of the Biobased Economy. Biomass is expected to become an increasingly important factor in meeting our need for functional materials, fuel, food and animal feed over the coming decades. To minimize the environmental impact, new efficient value chains need to be developed which provide at least 2 times more outputs and require at least 2 times less inputs. A crucial element in this approach is the integrated valorisation of biomass used across all relevant domains. For this, a better understanding of the connections between the different uses of biomass is required, not only from a technological point of view, but also in terms of cooperation and commitments between the involved stakeholders (governments, industry and knowledge institutions).

The results of this study are summarized in a roadmap towards an integrated and optimised system for the valorisation of biomass. For each stage, the starting points for stakeholders are described, as well as the barriers they must overcome to realise the transition to the next stage. From these barriers the issues on which the stakeholders need to work are derived, including the stimulation of cross-domain partnerships, expanding and applying knowledge, and developing new technologies.

Posters

Palm oil residues for the biobased Economy while maintaining soil nutrients

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Introduction The EU needs to import an estimated 100 million tons of biomass to fulfil its ambitions for bioenergy and biochemicals in the Biobased Economy by 2020. This will be an attractive new market for underutilised palm oil residues if concerns about nutrient removal and GHG emissions can be addressed. Approach We evaluated alternative oil palm mill set-ups in which POME and EFB are used for anaerobic digestion while fibre and shells are used for production of electricity, or exported, or converted to pyrolysis oil and exported. Scenario 1: Business as usual Scenario 2: POME + EFB for anaerobic digestion + combustion of remainder for local energy Scenario 3: POME + EFB for anaerobic digestion for local energy + pyrolysis of the remainder for high density oil and char for nutrients Scenario 4: POME + EFB for anaerobic digestion for local energy + partial export of shell and fibre for external commercialization Result: The analysis (Table 1) shows that GHG emission reduction and selling biomass can bring an extra revenue of up to US\$ 198,-/ha while making the main product palm oil more sustainable with better recycling of nutrients. Furthermore GHG emissions were reduced by up to 8 ton CO₂-eq /ha/year

Table 1. Scenarios for exploitation of biomass residues (Per ha per year) Scenario 1 BAU Scenario 2 grid connection Scenario 3 no grid connection Scenario 4 no grid connection
 Electricity generation kWh 323 5027 2384 2378 Steam (MJ) 21447 - 20839 20957 Power for local grid (kWh) 0 2062 2062 2056 Surplus power grid (kWh) 0 2643 0 0 Pyrolysis oil (ton) 1 - Exporting fibre and shell (MJ) 38 GHGs reduction (ton CO₂-eq) - 8.4 6.6 6.6 Cost Investment US \$/ha - 896 568 499 Annual US \$ - 56 34 29 Pyrolysis cost - - 100 US \$/ ha/year - Revenue Nutrient saving (US\$) - 15.3 15 12.8 Carbon credit (5 US \$/ ton CO₂) - 42 33 33 Selling electricity (US\$) - 120 - - Pyrolysis oil (US\$) - - 150 Exporting fibre and shell 57 Total revenue (US\$) - 177 198 103

Conclusions More options with new technologies should be analysed to enable maximum recycling of local nutrients, with the export of condensed biomass commodities. In this way, the requirement of the biobased economy will be fully covered by carbon, hydrogen and oxygen leaving other elements as nutrients at the biomass production site.

Biorefinery of coffee pulp for the production of valuable pectins and polyphenols - Economic potential and results of technology development to date

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Coffee is one of the most important agricultural commodities in the world. The total amount of green bean coffee produced in 2011 was 7.8 million ton; production is estimated to grow 10% in the next decade. Mechanization of coffee production has been carried out for the last twenty years to cope with the growing demand; this has created new challenges in the coffee value chain. On one hand mechanization increases productivity and lowers cost of production; on the other hand mechanized production generates more agricultural waste and reduces labour force in the plantation. Because of mechanization, coffee discarded streams are being concentrated in the washing stations, where the coffee cherries are transformed into green beans. Today the traditional wet method of coffee processing is being replaced by the semi-wet method. In the semi-wet method the pulp is separated from the beans by mechanical means, the mucilage is removed by friction and mixed with the pulp. The discarded streams are mostly composed of the skin, pulp, mucilage and silver skin of coffee cherries. This discarded stream contains high amounts of polyphenols, polysaccharides and sugars, as well as limited quantities of caffeine. These compounds makes the discarded stream unfit for use as compost or livestock feed, generating pollution in the coffee regions. Coffee pulp represents 40% of the total fresh weight of the coffee cherry. Traditionally coffee pulp was used in small amounts as fertilizer and composting but these applications are not technically efficient. The pulp biomass is rich in polyphenols, caffeine and complex polysaccharides such as pectin. These substances can be extracted from the pulp, by separating and refining the products following the biorefinery approach. By separating the coffee pulp biomass, the pollution loads will be reduced and the refined material transformed into valuable biobased compounds for use in the food and pharmaceutical industries. The aim of this research is the development of an efficient process for extraction and purification of pectin from coffee pulp. Pectin is a hydrocolloid widely used in the food industry as a thickening and stabilizing agent. Coffee pectin was studied in the past, but discarded as a suitable source of pectin due to the lack of gelling properties with calcium ions. New insight in pectin structure allows the modification of pectins with the use of enzymes, providing new functionalities for different applications. Coffee cherries from Arabica were collected in Colombia. The pulp was separated from the coffee beans and freeze dried. Pectin was extracted from the dried material by means of acid and alkali solutions, precipitated and dried at low temperature overnight. Pectin was analysed for sugar composition and uronic acid content. The extracted material shows high contents of galacturonic acid and arabinose. Further research will focus on optimization of the extraction, purification process and pectin functionalization.

The effect of large-scale algae cultivation for biofuels on coastal eutrophication in Europe

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The effects of large-scale algae cultivation for biofuels on coastal eutrophication in Europe. We analyze the consequences of large-scale production of biodiesel from micro-algae for eutrophication in European coastal waters of the Atlantic Ocean, the Baltic Sea, the Black Sea and the Mediterranean Sea. To this end, we analyze scenarios for the year 2050, assuming that in the 27 countries of the European Union fossil diesel will be replaced by biodiesel from algae. To replace all the current transport fuels by biofuels in the 27 EU countries 0.4 billion m³ biodiesel is needed per year (Wijffels and Bardosa, 2010), of which 0.3 million m³ for heavy traffic such as heavy duty trucks, freight vessels and airplanes (Eurostat, 2011). We estimate the required fertilizer inputs needed to grow algae for these volumes of biodiesel, and quantify how this may increase the nitrogen (N) and phosphorus (P) content of coastal waters, potentially leading to eutrophication. We use the Global NEWS (Nutrient Export from WaterSheds) model to estimate the river transport of N and P to the European coastal waters (Mayorga et al., 2010). We analyze scenarios for the year 2050 that differ in assumptions on biodiesel use, waste water treatment and recycling of fertilizer N and P. Coastal eutrophication is a serious problem in Europe. Current policies are aimed at reducing N and P loads. In our scenarios assuming no biodiesel use, river export of N and P is indeed decreasing over time. Our results indicate that the amount of N and P in the coastal waters may increase considerably in the future as a result of large-scale production of algae for the production of biodiesel. We also show that the N and P losses from algae production depend largely on the assumptions about waste water treatment and recycling of waste water in algae production. When assuming a 95% recycling or removal of the fertilizers from waste water, river export of N and P is 20-50% higher than in scenarios without micro-algae production. Our calculations are relatively sensitive to the assumed recycling and removal rate. In scenarios with 70%-90% recycling or removal the N and P inputs to coastal waters are considerably higher. For a recycling and removal of 70% we calculate that river export of N and P would be a factor of 3 and 5 higher, respectively, than without the production of biodiesel from micro-algae.

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Characterization of solid bases as catalysts for the optimized isomerization of glucose to fructose

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In the transformation of biomass to renewable fuels solid bases have shown to perform as catalysts for several reactions [1]. To understand the reaction mechanism and to be able to optimize catalysts a complete characterization of the catalytically active centers is necessary. Many research applications limit the characterization of solid basic catalyst to the specific surface area and the composition of the catalyst. The understanding of basicity is still difficult but essential. Methods for the investigation of basicity of solids described in literature are IR spectroscopy, UV/Vis spectroscopy, temperature programmed desorption (TPD), titration, various test reactions and many more. To be able to develop an experimental standard for characterizing the basicity the abilities and drawbacks of these methods have to be explored. UV/Vis and IR spectroscopy as well as TPD and titration have been compared. The investigation shows that TPD with CO₂ is the simplest and most convenient method. Unfortunately the reliability of the results is only given when the catalyst is not changed by the method. This happens for thermally instable materials like hydrotalcites. The big advantage of UV/Vis spectroscopy exploring basicity is working in the liquid phase. That way it can be adapted to settings similar to the reaction conditions. This is also the case for the Hammett titration method. IR spectroscopy gives the deepest insight into the nature of the available basic centers. Where the basicity determined by UV/Vis spectroscopy or TPD can only be classified in weak, medium and strong basic centers and the Hammett titration method gives pH areas of the active centers, IR spectroscopy can discriminate between the different types of interaction between the probe molecule and the basic surface centers. The drawback of all methods is the sensitivity to small changes in the procedure. That makes it difficult to compare results to literature. In this study the described methods have been applied to solid base catalysts used for the isomerization of glucose to fructose. In general, the mechanism of the production of HMF from glucose is proposed to go via this step [2]. Therefore a cost effective production of HMF relies on efficient processes for the isomerization of glucose into fructose. Previous studies described that besides other properties the basicity has an influence on the yield of the reaction. E.g. for MgAl hydrotalcites doped with tin and BaO @ Al₂O₃ doped with Mg the yield increased with increasing number of basic sites. A careful assessment of the described methods to characterize number and nature of basic sites of solid bases for the isomerization of glucose appears essential to enable an application oriented material design.

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Producing knowledge for the paper industry within the Biobased Economy

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The Competence Centre Paper & Board (KCPK) -the research arm of the Dutch paper & board industry- states innovation on behalf of and in cooperation with the industry as its mission. Its long-term strategy entails the contribution -through knowledge provision- on the continuity of the industry in the Netherlands, within an ever changing regulatory landscape and shifting global, regional and national trends. With the industry facing increasing competition for lignocellulosic resources, KCPK attaches high priority on new, more economical and -if possible- locally available raw materials. Its vision includes, therefore, the incorporation of side streams from both within and outside the paper industry to the feedstock supply. Projects are or have been examining -among others-:

- The use of the Organosolv process for the production of cellulosic fibres from agricultural/agro-industrial side streams (wheat straw, corn stover, rapeseed straw, camelina straw, grass),
- The development of a process for the fractionation of wheat straw into feedstock for the paper and chemical industries and for the energy sector. Furthermore, KCPK participates in the BIOcab project, aiming at Biobased Economy (BBE) growth in the northern Netherlands. Within it, new raw materials for the paper industry -from miscanthus and beetroot pulp to digestate and residues from greenhouse agriculture- are being considered. KCPK's approach to the integrated BBE is not limited, however, to alternative resources from side streams of other sectors; the paper industry's own side streams need to be valorized, either within or outside its boundaries. Ongoing projects examine:
- The separation of cellulose from paper industry sludge for the production of polylactic acid,
- The utilization of fatty acids within the paper industry process water as feedstock for the production of bioplastics (PHAs),
- The use of paper industry sludge as raw material for other paper qualities,
- The use of paper trimmings as a feedstock for the production of composite materials. Furthermore, new ideas -such as the production of BTX chemicals, as well as wood-plastic composites- are under consideration or development and supplement KCPK's promotion of a paradigm shift: from minimizing the costs incurred by side streams towards achieving the full utilization of their value potential within the rising BBE. KCPK is also setting its sights on a higher-level goal: making the paper industry able to steer the production process in such ways that side streams of desired characteristics come out of it, ensuring the maximum possible valorization of feedstock. KCPK has forged -and will continue to do so- ties and partnerships with various actors and economic sectors. Projects that bring the paper industry together with the chemical industry, the agricultural sector, energy producers, process technology and biotechnology developers, universities, research institutes and government agencies are carried out.

Biotechnology at Evonik

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Evonik, the creative industrial group from Germany, is one of the world leaders in specialty chemicals. Its activities focus on the key megatrends health, nutrition, resource efficiency and globalization. Profitable growth and a sustained increase in the value of the company form the heart of Evonik's corporate strategy. Evonik benefits specifically from its innovative prowess and integrated technology platforms. Evonik is active in over 100 countries around the world. In fiscal 2011 more than 33,000 employees generated sales of around €14.5 billion and an operating profit (EBITDA) of about €2.8 billion. Evonik is active in bioeconomy. Evonik's portfolio comprises amino acids and derivatives, active pharmaceutical ingredients (APIs), (bio)catalysts/products for the production of biofuels and platform chemicals, biobased polyamides, biobased polyester polyols for coatings/adhesives, cosmetics actives, emollients, enzymatically produced chiral α - and β -amino acids/chiral alcohols/chiral amines, peptides and keto acids for pharma/food/cosmetics, pharmaceutical aids for peroral/parenteral controlled release drug delivery systems, and surfactants. Evonik innovates and serves the bioeconomy markets. According to OECD bioeconomy comprises three segments - health, agriculture, and industry. To say it in short: Evonik is active in all three segments of bioeconomy. Products are Biolys®, DYNOCOLL® Terra, RESOMER®, ThreAMINO®, TrypAMINO®, and VESTAMID® Terra. Evonik delivers high performance products to its customers. An important performance attribute is sustainability. Life cycle assessment characteristics for Evonik's amino acids and biobased polyamides are outstanding. Another important benefit to the customer is global supply security. Evonik's global amino acid supply network and long tradition in large-scale production of feed amino acids by industrial biotechnology are highly valued by our customers.

Sustainable Insect Biorefinery

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Current protein production for human nutrition can't keep up with the global protein demand. In Europe consumption of whole insects is controversial. Hence alternative uses (as a non-visible food ingredient) are a nice alternative. This poster presents current state of the art & short term research focus. Briefly: Insects use side streams of food industry as a feed resource. A Biorefinery has been set up to provide safe insects as such or components thereof. Guidelines will be formulated when these components can be used as Food / Feed applications. Current research focusses besides the proteins also on valorisation of fat, chitin and other carbohydrates. Focus Points in research are for instance: Biorefinery, Preservation / Safety, Consumer Acceptance, Product development, LCA / sustainability, Quality comparison to traditional animal ingredients & plant alternatives. Hurdle examples are: Legislative / Novel Food Regulation & the creation of a new sector of insect breeders for large scale production.

Biorefining organic waste into biochemical & biofuels

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Sustainable production of fuels and chemicals are demanded to avoid the depletion of and pollution from fossil fuels. To recover resources from waste can be an ideal alternative for supplying chemicals and fuels. Mixed-culture Biological Chain Elongation (MBCE) is an innovative environmental technology that can produce Medium Chain Fatty Acid (MCFA, C5~C9) from organic waste and low grade biomass. MBCE employs microorganisms to achieve this production, which consists of two main steps: First, compositions in organic waste such as carbohydrate, fat, and protein are hydrolysed and acidified into Volatile Fatty Acids (VFA, C2~C4), H₂, and CO₂; second, these acidification products together with (dilute) ethanol are converted into MCFA. Both steps can be performed in mixed culture fermentation, which implies the robustness, resilience and potential cost-reduction of the production system compared with pure- or co-culture fermentation. Organic waste as feedstock also provides MBCE several benefits such as low cost, large availability and free of food competition. MBCE is so far still under development in lab-scale, and its MCFA production rate has been enhanced into the same range of other fermentation technology such as anaerobic digestion and ethanol fermentation. However, there are four main obstacles that need to be overcome: first, the present high production rate is based on artificial waste, and the researches performed on real organic waste result in lower production rate and yield. Shifting the research focus from artificial waste to real waste is therefore of importance. Second, the addition of (dilute) ethanol implies potential high cost and impact, which need to be replaced. Alternatives can be, for instance, utilising waste streams containing ethanol. Third, the promotion of MBCE into a conceptual biorefinery is required. The researches related to MBCE mostly focus on the fermentation step. There are several other steps to be investigated in order to realise the implementation of MBCE; examples are the separation of product from fermentation broth and the post-treatment of the fermentation waste. Last but not least, the overall environmental performance of the MBCE conceptual biorefinery has to be evaluated. The evaluation will be used to compare MBCE with other MCFA production technologies or other organic waste recovery techniques. By tackling these four obstacles we expect to promote MBCE into a conceptual biorefinery with the least environmental impact that can be implemented in the future.

Integrating study on high effective biofuel energy recovery with multistage biorefinery process

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The energy safety issue is always a pressing consideration in Taiwan with almost 100% imported fuel and high dependence on fossil fuel. A 3-year integrating study on biofuel energy recovery with multistage process was launched in 2009 by Taiwan's National Science Council. Five research groups from different fields including biochemical engineering, life sciences, biotechnology and environmental engineering were integrated to focus on various bioenergy production and residue treatment. The research scope covered basic research on bioethanol and biobutanol production from cellulosic materials and the wholesome treatment process with biohydrogen and biomethane production. In order to improve the limiting step of hydrolyzing lipid and cellulosic materials, metagenomic study was conducted to clone innovative hydrolyzing enzymes. Eight cellulolytic sequences and five lipase sequences were obtained. These enzymes were confirmed as outer-membrane characteristics and the hydrolyzing kinetics tests should be further investigated. Genetic engineering was also conducted to improve cellulolytic efficiency and bioavailability of cellulosic materials. Ordered gene assembly in *Bacillus subtilis*(OGAB) method was adopted to construct a high efficiency cellulose degrading organism with butanol production ability. A group of functional genes, including cellulose originated from *Clostridium thermocellum* and butanol fermentation originated from *C. acetobutylicum* ATCC 824 were transplanted into *Bacillus subtilis*. Different gene assemblages were also tested to evaluate the performance of cellulolytic ability and butanol production. Real-time PCR was adopted to monitor the RNA expression level of each cellulolytic gene. The result indicated that the gene engineered *Bacillus* can express related cellulolytic genes in RNA level. Three enzyme assays of endo-glucanase, exo-glucanase, and xylanase were also revealed that the closer a gene near the promoter the higher activity can be obtained. On the other hand, the fermented residues presented somewhat refractory cellulosic and oily materials that should be degraded furthermore by the selective syntrophic microbial community. In this integrated process, aerobic solid state fermentation was presented for residue transformation. Refractory cellulosic material such as lignin can be converted by fungi to produce high concentration lignin modifying enzymes and mineralization of the organic residue simultaneously. Besides, kitchen waste and napiergrass were fed into a biohydrogen and biomethane recovery process. Lactate was observed easily converted from kitchen waste and our study revealed that by controlling the volumetric loading rate under 15 kg-COD/m³-d can effectively transformed in the biohydrogen tank. Oil, grease and cellulose was remained in bioH₂ tank but can be further utilized in bioCH₄ tank. The COD recovery can achieve 80% and transformed as methane gas.

Study of self-assembling properties of yeast-derived acidic glycolipid biosurfactants

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Sophorolipids are yeast-derived, water-soluble, glycolipids with very low toxicity, antimicrobial properties and greater biodegradability compared to the petroleum-based counterparts and therefore they gained more and more attraction as alternative surfactants within the last decade. Their synthesis occurs from renewable feedstock, achievable using the yeast *Candida Bombicola* with a production yield of $>300\text{g l}^{-1}$ [1]. The biosynthesis of this chemical compound is an attractive combination between green chemistry and lower carbon footprint providing less waste.

Numerous industrial applications exist such as cosmetics (ex, skin care products) and cleaning formulations because of their low foaming potential and high surface activity properties[2].

Like other biosurfactants, sophorolipids exhibit an interesting physicochemical behavior[3]. They can form giant ribbons as well as micellar structures or tubular aggregates through self-assembling under specific conditions[4].

The design of this molecule bearing a COOH group at the end of an alkyl chain attached to a sophorose moiety provides a pH responsive compound. The specificity of sophorolipids consists in being an asymmetrical bolaamphiphile which has unique characteristics with regard to pH, temperature and ionization degree.

Small angle scattering is a powerful tool to characterize the diversity of morphologies sophorolipids can offer. The investigation of micellar structures by Small Angle Neutron Scattering (SANS) and transmission electron microscopy under cryogenic conditions and the effect of pH was described by Baccile *et al.* in 2012[3]. They reported an unusual pH-dependent self-assembly behavior with regard to the amount of negative charges on the carboxyl group.

The goal of this contribution is to bring further insight into the understanding and control of the wide range of supramolecular assemblies of sophorolipids as a function of pH and temperature in order to use them as biobased products for the synthesis of new materials.

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Xylose dehydration to furfural in the presence of glucose

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To achieve an all-embracing alternative for the use of fossil resources in modern economies the production of platform chemicals from lignocellulosic biomass should be considered. Lignocellulosic biomass mainly consists of cellulose and hemicellulose (and smaller amounts of lignin). Cellulose is a homogeneous polymer of glucose units, whereas hemicellulose is a heterogeneous polymer of various (both C6- and C5-)saccharides, but predominantly xylose. When platform chemicals are to be produced from either the hexose or the pentose content of the biomass, the reaction mixture will inevitably contain both hexoses and pentoses. The current study focuses on the dehydration of xylose to form the platform chemical furfural in the presence of glucose. Experiments have been performed in a one-liter autoclave reactor under dilute acidic (HCl, 50 mM) and saline (NaCl, 500 mM) conditions. The pressure in the reactor was equal to the saturation pressure of the mixture. The sugar and furfural concentrations were analyzed using an HPLC apparatus equipped with both an RI and a UV detector. First, series of experiments with pure furfural (50 mM) and series of experiments with pure xylose (50 mM) at reaction temperatures between 160 and 200°C were performed. Next, both experimental series were repeated, but with addition of 50 mM glucose. Furfural conversion and furfural yields were then compared for the cases with and without glucose. Furthermore, for all these experiments the kinetic (Arrhenius) parameters were determined by minimizing the sum of squared normalized errors. The experimental results clearly indicated a strong influence of the glucose on the degradation of furfural. Furfural is degrading significantly faster when glucose is present compared to the experiments with pure furfural. Conversely, the furfural yield of the dehydration of xylose with glucose present did not differ largely from the experiments without glucose. However, in these experiments the formed furfural was decreasing faster at longer reaction times when glucose was present. This observation confirmed the higher furfural degradation rates with glucose present. It is concluded that glucose enhances the degradation of furfural, whereas it has no significant effect on the dehydration of xylose towards furfural. From experiments with furfural and varying concentrations of glucose it turned out that an additional (second-order) reaction had to be added to the reaction mechanism of furfural degradation in order to satisfactorily predict the experimental data. This additional reaction incorporated the initial glucose concentration as a constant in the Arrhenius expression for the reaction rate constant.

Protein behaviour in ionic liquids

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Ionic liquids are gaining more attention as novel tunable solvents due to their unique physical and chemical properties. Ionic liquids are widely used as reaction media in biocatalysis and extractions of metals. However in order to use these ionic liquids for extraction of biomolecules, in particular proteins, it is necessary to understand the stability of proteins in ionic liquids. Interaction between proteins and ionic liquids is not well understood. The aim of this work is to understand the stability of model proteins BSA, IgG and Rubisco in Ammonium and Phosphonium based hydrophilic ionic liquid. The protein stability was investigated over wide range of ionic liquid concentrations. Non-reduced gel electrophoresis gives an indication about the native form of the protein. Additionally the stability of the proteins was also investigated using Differential scanning calorimetry (DSC) and size exclusion chromatography (HPSEC). Protein extractions using ionic liquids are normally studied using aqueous two phase extraction system. The studies were further extended to stability of proteins in aqueous two phase extraction system. The preliminary studies reveal that low concentration of ionic liquids favours protein stability.

RuBisCo protein from sugar beet leaves

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RuBisCo protein from sugar beet leaves RuBisCo protein is a protein from plant leaves that has a role in the photosynthesis and that is present in all green plants. RuBisCo also is an interesting protein for application in food products. First of all it possesses good functional properties like gelling and foaming [1]. Second, it is very nutritious because of a good digestibility and a high rating in essential amino acids (AAS 87%) [2]. Third, it has a very low allergenicity [3]. In the Netherlands sugar beets are grown on 70.000 hectares with an average yield per hectare of 70 ton. Besides the sugar beets, 30 – 40 ton per hectare of leaves and stems are produced which, at present, are discarded on the field. These leaves however, contain 1% RuBisCo protein [4] and together with 4 SME's, TNO has developed and tested a process which gives a functional, colorless and off-flavorless protein. The process is currently being scaled up. The process steps that are under study are:

- The harvesting of the leaves without stems and dirt,
- The pressing of leaves with a resulting juice, low in fiber and high in protein
- The removal of chlorophyll from the juice whilst leaving the protein unaffected
- The concentration of the protein to a concentration that can be spray dried
- The spray drying of the concentrate with a dry, soluble and functional protein as a product.

In the harvesting campaign of 2012 a production line for the production of 30 kg of RuBisCo protein per day is set up. The end product will be a spray-dried protein powder which will be tested on functionality and application in food products. The proposed presentation will show the results obtained from this production, it will address the possibilities of this novel protein source and the place this process has in the transition towards the use of more vegetable proteins.

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Microalgal extracts as natural alternatives for synthetic antioxidants

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Over the last decades, microalgae received much attention as energy crop, but the cost of microalgal biofuels is at present not competitive with fossil fuels. To maximize the value of the biomass, it is important that as many high-value compounds as possible are extracted. Antioxidants are potentially high-value products that can be used to improve UV-stability of plastics or in food and feed applications to replace synthetic antioxidants. Most of the research on microalgal antioxidants focused on lipophilic carotenoids or antioxidant enzymes. Much less is known about the presence of more polar antioxidants such as phenolics, an important class of antioxidants in terrestrial plants. We examined antioxidant properties of 32 samples of microalgal biomass (commercial samples and samples from own cultivation on pilot installation). Extracts were prepared using two extraction procedures. The first procedure aimed at extracting all antioxidants using a one-step extraction with an ethanol/water mixture. The second procedure aimed at fractionating the different antioxidant types by subsequent extraction with hexane, ethyl acetate and hot water. Antioxidant activity was measured with 4 assays, i.e. TEAC, FRAP, AAPH induced oxidation of linoleic acid and square wave voltammetry (SWV). SWV allows determination of antioxidants in extracts when traditional photometric assays cannot be applied (e.g. due to chlorophyll interference or turbidity of the samples). The strongest antioxidant activity was found in the ethanol/water extracts. Lowest antioxidant activity was measured in the hexane fractions, while more activity was found in the more polar ethyl acetate and hot water fractions. The fact that the hexane fraction is low in antioxidant activity is interesting as this solvent is commonly used to extract lipids. Thus, after removing lipids from microalgal biomass, antioxidants can still be extracted with a medium polar solvent or with hot water. It was further found that antioxidant activities determined by SWV correlate well with results from other established antioxidant assays and that SWV provides additional information on the antioxidant profile of microalgal biomass. Hence, SWV serves as an excellent tool for optimizing fractionation processes to recover antioxidant components from microalgae. Besides screening microalgal species for their antioxidant potential, we studied the effect of nutrient availability on antioxidant formation in 3 species. It was found that antioxidant activity of microalgae is lower under nutrient limitation but can be increased by elevated nutrient levels. Finally, antioxidant activity in microalgal preparations was compared with extracts from other well-known natural antioxidant sources (i.e. grape seeds, green tea, rosemary and gall nuts). From this, it could be concluded that microalgal extracts can be a good alternative for traditional plant extracts (e.g. some microalgal extracts showed TEAC values similar to rosemary).

Highlighting the different enzymatic degradation pattern of plant tissues by separation of particles of ground maize stems

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Enzymatic saccharification of lignocellulosic material is performed on ground particles that originate from tissues differing in their biochemical composition. The grinding of the materials and the extent of degradation of individual particles will depend on the tissue. The objective of the present work was to isolate particles through a dry separation process in order to study their enzymatic degradation pattern. Maize was considered both as a model and crop plant. Whole plants without ears were ground in a hammer mill with a mesh size of 1 mm and fractionated by dry separation process. Three fractions were obtained by turbo-separation. Particles of fine and medium size were submitted to electrostatic sorting to obtain positive and negative electrode deviated fractions. Both the biochemical and particle size evolution during enzymatic degradation were followed using a prototype of reactor built for visualisation of particles during hydrolysis. The prototype allows setting temperature, stirring, and automatic image acquisitions at given times. Particle size was quantified by assessing granulometric curves using image texture analysis. Sugar release was measured off line by colorimetric method after sampling the media at given times. Fractions had different lignin amount and structure. Large fractions were richer in lignin and fine fractions exhibited a higher ratio of syringyl over guaiacyl units. For all the fractions major sugars were glucose, xylose and in a lesser extend arabinose and uronic acids. Large particles had a higher amount of xylose compared to fine particles. Fine particles were richer in arabinose and uronic acids. Fractions were incubated in the reactor for 7 h with a combination of cellulase, xylanase and glucosidase. Fractions differed in their behaviour towards enzymatic degradation. The large particles were the less degraded both considering the particle size and biochemical evolution. Fine particles were the most degraded with a concomitant release of sugars and decrease of particles size. Positive deviated medium size particles had a particular behaviour with most of the sugar released during the first 30 min without any noticeable change in particle size. The negative deviated medium size particles behaved like the large ones. The present work shows that plant heterogeneity should be taken into account in the biorefinery chain. Next steps will be to relate particles degradation pattern to stem histology and to refine the separation process to select fractions of interest.

Disruption of Green Microalgae by Bead Milling as the First Step in Biorefinery

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Background Scarcity in fuel based resources and the climate change are important drivers for the promotion of fuel and chemicals from renewable feedstocks. Within this respect, microalgal biomass and its constituents are regarded as the future feedstock for sustainable production of renewable products because microalgae have much higher growth rates, photosynthetic efficiencies and process optimization possibilities than conventional terrestrial plants. The major economic bottleneck cited in the literature is algal productivity, followed by labor and harvesting/extracting costs. In order to overcome these economic problems it is vital to promote better yields in downstream processes which increases the product yields. Aims The aim of this study is to investigate the mass transfer between the supernatant and solid phases during bead milling of green microalgae biomass. Methods *Neochloris oleabundans* was chosen as the model green microalgae due to the increasing interest of this microalgae. The cultivation was done under photosynthetic conditions by using Bold's Basal Medium. Bead milling (BM) was chosen as the model cell disruption method for processing biomass. BM was carried out according to Schwenzfeier et al. (2011). After the cell disruption the supernatant and solid phases were separated by centrifugation. The untreated, treated-solid and treated-supernatant phases were analyzed to determine the total biochemical compositions. Folch, BCA and Dubois methods were used for total lipid, protein and carbohydrate determinations respectively. Solid phase extraction was carried out for crude lipid fractionation. All of the analyses and bead milling studies was done in triplicates. Results The results are summarized in Figure 1. The data showed that the distribution of biochemicals is non-selective. All the distribution values obtained from biochemical analyses are similar to biomass distribution values, being 35(\pm 5)% for supernatant (Fig. 1). In other words the biochemical composition of untreated, treated supernatant and treated solid samples are similar. All three phases consisted of 34.5 (\pm 2)% lipid, 53 (\pm 2)% protein and 11 (\pm 1)% carbohydrate. Summary / Conclusions The mass transfer between supernatant and solid phases during bead milling of *Neochloris oleabundans* biomass was investigated. The results showed that the distribution of biochemicals among supernatant and solid phases is fully related to the biomass distribution. This observation validates the hypothesis that bead milling causes a non-selective release of biochemicals. Mild cell disruption methods which provides selective biochemical release will be investigated in further studies to be able to compare the results with this study. In this way we will be able to look at cell disruption process with a biorefinery point of view.

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Biorefinery: key for a successful biobased bioeconomy

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The emerging biobased economy is causing shifts from fossil oil based resources towards renewable resources. Because of market mechanisms, current and new industries utilising renewable commodities will attempt to secure their supply of resources. Due to the initiated transition towards a biobased economy, renewable resources like lignocellulose, agro-food residues and aquatic biomass are intensively investigated for new applications such as advanced biofuels, green chemicals and materials.

Crucial step in developing biomass-based industries is to develop concepts for integrated biorefineries for conversion of biomass into valuable components like cellulose, sugars, lignin and protein and finally to products. Despite the great relevance of biobased products for many industries, experts still see numerous technical, strategic and commercial challenges that need to be overcome before any large-scale commercialization of the industry can succeed. Biorefineries will have to employ the most feasible technologies to ensure that biobased products break even. This will require the concerted action and cooperation of many non-traditional partners like biomass producers, chemical-, food and feed- and energy companies. It is required to cover all aspects of the complex biomass value chain, from feedstock production to end products and end-user distribution as well as technologies that are applied for pretreatment, extraction and conversion of biomass. Besides technological innovations, there is a need to translate scientific and economic information on renewable resource availability, conversion techniques and application into practical accessible information for interested layman and entrepreneurs.

Wageningen UR (WUR) is a strong player in the field of biorefinery research as a multidisciplinary approach is required. WUR can use its in-house available expertise from biomass cultivation to value chains and biobased end-products combined with conceptual process design and policy know-how. Value chain information is combined with knowledge on biomass logistics and converted to more accessible information, often in cooperation with the Dutch Government [1-3]. In this presentation examples from research projects which are performed in cooperation with other knowledge institutes and companies will be highlighted. For example the fractionation of various types of biomass (on lab-scale or bench-scale) into cellulose, sugar, protein or lignin fractions will be presented. Also the valorisation of these fractions into valuable biobased chemicals and building blocks will be highlighted.

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Catalytic cracking of bio-based oils – a bio-jet-fuel-option for biorefineries

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In 2009, the global aviation stakeholders, including the International Air Transport Association IATA, have agreed upon a set of goals to limit aviation's contribution to the global warming, including a 50%-reduction of CO₂ emissions by 2050, relative to 2005 levels. Improved aviation technology will be one important part of the sustainability strategy – optimizing infrastructure and navigation procedures being another. But there will be no chance to reach these ambitious goals without partly substituting fossil fuels [1]. Aviation jet fuels like Jet A-1 show a very narrow boiling range. Unlike gasoline and diesel fuels, they have to contain certain amounts of aromatics in order to ensure seal swelling. Catalytic cracking – a flexible biorefinery module Catalytic cracking of bio-based oils proves to be very robust concerning feedstock compositions and impurities. It is an ideal pathway to transform bio-based oils as well as their wastes and fatty acid-containing by-products from plant oil processing into high-quality jet-fuel components. Route one: standard bio-based oils ⇒ jet-fuel isoparaffins Components for bio-jet fuel production can be achieved by catalytic cracking of fatty oils and fatty acids over activated carbon catalyst. At reaction temperatures of about 450 °C, mainly C15- and C17-n-Alkanes that can be isomerized for jet fuel-usage are produced. They can be used for bio-kerosene after isomerization. Route two: High-oleic oils ⇒ jet-fuel n-paraffins Introducing high-oleic feedstock like HO-sunflower-oil and slightly raising the reaction temperature leads to high amounts of n-alkanes in the jet-fuel boiling range. Route three: bio-based oils ⇒ jet-fuel aromatics Using alternative catalysts leads to an enhanced production of alkylated benzenes which are indispensable for aviation jet fuel. With plant oils like Jatropha Curcas-oil or plant oil wastes, remarkable amounts of alkylated benzenes can be produced. Benzene and toluene were not detected; the typical chain-length of the branched alkyl is C4 to C11. They can be used for supplementing the aromatic content of bio-jet-fuel, which the products in most of the alternative process routes lack.

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Outlook for the co-production of electricity and ethanol in Brazil using different biomass feedstocks

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The goal of this research is to give an future outlook of the ethanol and electricity production costs in Brazil, based on the historic trends and expected development in biomass feedstock cultivation and conversion technologies. Ethanol is an important biofuel for transportation, and gained increasing interest as renewable feedstock for bio-based chemicals. Historically, the Brazilian ethanol industry was based on the fermentation of sugar juice derived from sugarcane stalks, the so-called first generation technology. Van den Wall Bake (2009) observed an significant cost reduction of sugarcane cultivation costs and ethanol production costs in Brazil, since the 1970's. While in recent years the sugarcane production costs increased, see figure below. Future improvement options, among others, incorporate the utilization of other sugar crops which can be harvested outside the harvesting window of sugarcane and the implementation of second generation technology. Due to mechanical sugarcane harvesting sugarcane trash will become available for ethanol production or electricity generation. The utilization of 2nd generation technology will also open doors for other (lignocellulosic) biomass feedstocks for ethanol or electricity production, especially in cultivation area's less suited for sugarcane cultivation. Van den Wall Bake (2009) determined the technological learning in first generation ethanol production from sugarcane in Brazil. In the current study we would like to determine the contribution of changing yield, land costs, labour costs, fertilizer prices and amount of cultivation inputs to the cultivation costs with a multi-factor learning curve. By doing so, insight is given in the historic development of the cultivation costs reduction of sugarcane. This is the basis for the future outlook for cultivation costs of sugarcane, energycane, sweet sorghum, eucalyptus and elephant grass. The development of first generation technology also historically underwent a significant costs reduction has been observed. Based on available data and expert opinion and future outlook is given for the industrial production costs of first and second generation technology. Preliminary results show that the costs reduction in sugarcane cultivation is due to, among others: yield development and lower harvesting costs. Sugarcane yield increased more rapidly than increasing land costs, thereby resulting in lower land costs per tonne harvested cane. Reduced harvesting costs are due to increased output per worker, with relatively stable daily wage. Furthermore, the use of sweet sorghum is economically feasible, as a larger operation window of the ethanol production facility would pay-off the increased biomass feedstock costs. Final results are intended to depict the historic and future development of total ethanol production costs, up to 2030 in Brazil, taken into account the development in biomass cultivation and biomass conversion to ethanol.

A thermophile under pressure: Transcriptional analysis of the response of *Caldicellulosiruptor saccharolyticus* to different H₂ partial pressures

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Caldicellulosiruptor saccharolyticus is an extremely thermophilic, grampositive anaerobe which ferments a broad range of substrates to mainly acetate, CO₂, and hydrogen gas (H₂). Its high hydrogen-producing capacity make this bacterium an attractive candidate for microbial biohydrogen production. However, increased H₂ levels tend to inhibit hydrogen formation and lead to the formation of other reduced end products like lactate and ethanol [1]. To investigate the organism's strategy for dealing with elevated H₂ levels and to identify alternative pathways for reductant disposal, the effect of the hydrogen partial pressure (PH₂) on fermentation performance was studied [2]. For this purpose cultures were grown under high and low PH₂ in a glucose limited chemostat setup. Transcriptome analysis revealed the up-regulation of genes involved in the disposal of reducing equivalents under high PH₂, like lactate dehydrogenase and alcohol dehydrogenase as well as the NADH-dependent and ferredoxin-dependent hydrogenases. These findings were in line with the observed shift in fermentation profiles from acetate production under low PH₂ to a mixed production of acetate, lactate and ethanol under high PH₂. In addition, differential transcription was observed for genes involved in carbon metabolism, fatty acid biosynthesis and several transport systems. The presented transcription data provides experimental evidence for the involvement of the redox sensing Rex protein in gene regulation under high PH₂ cultivation conditions. Overall, these findings indicate that the PH₂ dependent changes in the fermentation pattern of *C. saccharolyticus* are, in addition to the known regulation at the enzyme/metabolite level, also regulated at the transcription level.

Biohydrogen production from glycerol by *Thermotoga maritima*: proposed pathway and bioenergetic considerations

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Hydrogen gas (H₂) is considered an attractive alternative to fossil fuels, as it burns cleanly, without emitting carbon dioxide (CO₂) or any other environmental pollutants. Biohydrogen is produced either by biophotolysis, microbial electrolysis, photo-fermentation, using light-dependent organisms, or by dark fermentation. Biohydrogen production by dark fermentation is an anaerobic process, involving heterotrophic fermentative bacteria or archaea that convert biomass or biomass-derived hydrocarbons mainly to H₂ and acetate. One of the potential substrates for this process concerns crude glycerol, which is an inevitable by-product of the production of biodiesel. The accelerated growth of the biodiesel industry has generated a surplus of glycerol, that resulted in a 10-fold decrease in crude glycerol prices. Its availability, low price and its potential to mitigate possible environmental hazards make glycerol an attractive carbon source for industrial microbiology including the dark fermentation processes. The production of biohydrogen from glycerol, by the hyperthermophilic bacterium *Thermotoga maritima* DSM 3109, was investigated in batch and chemostat systems. *T. maritima* converted glycerol to mainly acetate, CO₂ and H₂. Maximal hydrogen yields of 2.84 and 2.41 H₂ per glycerol were observed for batch and chemostat cultivations, respectively. These data suggest that glycerol is fermented to acetate, CO₂ and H₂ in a ratio of 1:1:3. End products commonly found in mesophilic glycerol fermentation by enterobacteria or clostridia, like ethanol, butanol, 1,3-propanediol, 1,2-propanediol, succinate, or formate, were never detected. For batch cultivations: i) hydrogen production rates decreased with increasing initial glycerol concentration, ii) growth and hydrogen production was optimal in the pH range of 7-7.5, and iii) a yeast extract concentration of 2 g/l led to optimal hydrogen production. Stable growth could be maintained in a chemostat, however, when dilution rates exceeded 0.025 h⁻¹ 25 glycerol conversion was incomplete. A detailed overview of the catabolic pathway involved in glycerol fermentation to hydrogen is proposed. Based on comparative genomics the ability to grow on glycerol can be considered as a general trait of *Thermotoga* species. The exceptional bioenergetics of hydrogen formation from glycerol is discussed.

Qualification of a biomass as a renewable energy source as a substrate for bioethanol production

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Promotion of a renewable energy by imposing on member states duty to definite support systems is one of a key element of European Union energy policy. Such approach seems be natural in accordance to the limited quantity of primary resources (such as coal) and climate changes which we deal with in the last decades. Simultaneously it should be underlined that diversifying the structure of individual fuels in the energy balance is increasing energy security of the state and improve efficiency of using available resources. A support for biofuels and biocomponents is an additional component of widely understood promotion of renewable energy sources. The main aim of the paper is an evaluation of actual and future EU and polish legal acts conected with promotion and producing energy from renewable energy sources, biofuels and biocomponents in particular taking into consideration biomass for energy purposes. Those issues seems to be especially important in context of legislative changes and fundamental meaning of biomass in process of producing renewable energy in Poland.

Valorization of waste streams, "From food by-products to worm biomass"

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A new technology is investigated to produce a high quality animal feed source by converting safe industrial food wastes into worm biomass. The freshwater worm *Lumbricus variegatus* (common name: blackworm) has been selected for this purpose. This species can be used to reduce and concentrate municipal wastewater sludge and recover its nutrients. Its simple life cycle and high stress tolerance make this worm suitable for a controlled application. The worm's macronutrient composition also makes it a promising aquaculture feed source. The levels of protein, lipid and sugar are in accordance with the general fish feed requirements. The goal is to develop an eco-effective bioreactor for the production of worms based on a low-grade waste stream. First, standardized growth experiments are performed to investigate the performance on different food wastes varying in macronutrient composition and origin (vegetable, bacterial and animal). For this purpose a suitable test setup was developed and in the meantime several successful tests with plant sources have been performed. Recent tests with wheat gluten and starch delivered more insight in the digestibility of protein and carbohydrates and resulting worm growth. Secondly, a prototype worm production reactor is designed, tested and optimized. The key element in this is the use of a carrier material which acts as a separation layer between the worm-feed compartment and the water column. In this concept, streams of worm feed, worms and worm feces can be separated. The challenge is to control the conditions for sustainable growth and efficient biomass production. To determine the efficiency of the reactor, mass flows for carbon and nitrogen are determined in combination with worm growth performances. In two years' time, experiments with different food wastes of interest will result in various criteria for worm feed selection. Development of the reactor will mainly depend on the growth performance on a specific worm feed suitable for reactor application. The commercial success will largely depend on the acceptance of blackworms as a safe feed for cultured animals and the legalization of food waste products for worm feed application.

Deterministic ratchet technology for fractionation of particulate suspensions

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We report on the design and scale-up of a new separation technology, which allows recovery of particles from suspensions [1, 2]. The separation technology employs so-called ratchets, which consist of periodic arrays of obstacles in a flow channel. Suspended particles below a critical hydrodynamic diameter move in the main flow direction through the channel, whereas particles exceeding the critical particle size are horizontally displaced by the obstacle array. The objective of the current study is to evaluate deterministic ratchet technology for separation of concentrated suspensions at large-scale.

Two experimental deterministic ratchet devices were developed to study particle separation and fractionation. Critical to the separation of particles is the distance between two adjacent obstacles. In the current study two devices were constructed with critical distances of 200 μm and 800 μm , respectively. Model suspensions were prepared by suspending polyethylene and polystyrene particles in water. Subsequently, separation was evaluated by analyzing composition of outlet fractions as a function of ratchet design and operational conditions such as inlet concentration and flow rate (or hydrodynamic conditions).

Surprisingly it was found that separation efficiency process was strongly dependent on hydrodynamic conditions. At $Re=1$ no separation occurred whereas for $Re>1$ suspensions could be concentrated up to a factor 3 into 1 of the 5 outlets for both 200 μm and 800 μm devices. The latter corresponds to a recovery percentage of 60% of the particles in the specific outlet. Maximum inlet concentrations for the devices found were 1.6 and 4.5 v/v%, respectively. From CFD simulations and high speed camera imaging it further appeared that at these increased Reynolds numbers, a pair of vortices developed behind obstacles. Both the presence of vortices and the increasing number of flow lanes could explain the observed increased separation efficiency.

The large separation efficiency, which improved at increased flow rates, and the tolerance for high inlet suspension concentrations may open up ways for design of a novel efficient separation process.

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Biorefinery mapping and Biorefinery indexes for regional policies: an application in Emilia-Romagna Region

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This work concerns decision making for biorefineries applied to a regional scenario. The first part of the work focuses on value chains which are actually running in Emilia-Romagna Region (Northern Italy) with the purpose of measuring and assessing the chains integration and biorefinery output capacity. Considered value chains are based on palm oil, rapeseed and sunflower oils, biogas and biomethane from maize and manure. Two indexes are proposed: value chain activation (VCA) index and biorefinery output (BIO) index. VCA index is intended to measure how diverse are the economic activities along the value chain. Diversity is based on statistical NACE (National Classification of Economic Activities) European code which makes possible to measure the number of different codes involved in a specific bio-based value chain. The work illustrates how system boundaries, aggregation and exemptions have been treated. The biorefinery output index measures the diversification of products (e.g. power, heat, biochemicals, food, feed, etc.) obtained along a biomass-based value chain; a weight is assigned to the different types of products to get a weighted index. Results show how the biogas and bio-methane value chain obtain the highest result for VCA, but that the highest result of the BIO index are currently obtained by a palm oil biorefinery plant. The second part of the work deals with potential biorefineries in the region and aims to foster a sustainable and versatile biomass based value chains. The key concept is the development of mapping systems at regional scale, for a more complete picture of the spatial distribution of relevant elements for future bio-based chains. These include biomass and related activities (upstream side), primary and secondary treatment systems, marketable products and related distribution system (downstream side). The work focuses on three of these potential value chains: biodiesel from used dietary oils, Polyhydroxyalcanoates (bioplastics) from residual biomass, and polylactic acid (PLA) from the cheese industry sector. The work shows a map related to the potential value chain of biodiesel from used oil.

This work has been initiated together with the Energy Department of the Emilia-Romagna Region (Italy) and sees the collaboration of the consortium for innovation and technology transfer in Emilia-Romagna ASTER.

Roadmap Biobased Economy

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In a biobased economy a wide range of products (materials, chemicals, fuels, food and feed, etc.) is produced from a wide range of resources (wheat, rapeseed, wood, grass, algae, organic residues, wastewater etc.). These production chains are currently developing along parallel pathways that are getting more and more interconnected. Side products from one process are used as a resource for other processes. An overview of all the possibilities and interconnections cannot easily be fitted in a figure on paper. A web tool named Roadmap Biobased Economy (<http://www.biobasedeconomy.nl/routekaart/>) was developed to provide a deeper insight into the biobased economy. The web tool shows raw materials, products, intermediates, processes and interconnections that play a role within the biobased economy. Each item is provided with a short explanation and a picture. Relevant routes are highlighted when browsing over the raw materials, products, intermediates and processes. The routes can be viewed in more detail and in coherence with other routes. The explanation provides insight in the considerations that determine the choice for a given raw material, intermediate or process. Infographics provide additional information on most routes. Altogether this web tool will help entrepreneurs, scientists, policy makers and interested people to find their way in the web of the biobased economy.

Exploring new magnetic and photonic functions in coccolith of *Emiliana huxleyi*

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The algae coccolithophores is a spherical plant cell and its proliferation in the ocean over the past several billion years had a large impact on the earth's carbon cycle. The interesting point in coccolithophores is the coccolith, which is a curved disc of 2~10 μm in diameter, being made of calcium carbonate and the discs covering the cell surface. The function of coccolith discs has not yet been clarified sufficiently, but it is conjectured that the discs are regulating the exposure to the solar irradiations. One of the recent literatures reported that holo-coccolith discs act as a UV light filter for the cell inside. The aim of this study is to explore the possible effects of strong static magnetic fields on the optical properties of coccolith discs in the coccolithophores. Two kinds of coccolithophores, *Emiliana huxleyi* and *Calcidiscus*, were investigated for the structural color analyses. The observation of a coccolithophore with accumulated coccolith discs under a dark field illumination revealed that the stack of calcium carbonate crystals generated a slight iridescent color distribution by the optical interference between coccolith discs. The magnetic field sweep-up to 5 Tesla (T) enhanced the green, blue, and red colors of the discs. While in the sweep-up to 10 T, yellow colors dominated the coccolith disc arrays covering the cell surface. Particularly at 10 T, the outline shape of agglomerated coccolith discs became slim when it was observed in parallel to the applied magnetic fields. Therefore, it was speculated that the calcium carbonate discs became partially aligned under strong magnetic fields. Consequently, the placements of coccolith discs were modulated and the light interference between the coccoliths with periodical crystal structures changed the color intensity. The results suggest that the utilized magnetic fields can control the solar light harvest by modulating the photonic structures in coccolith stacks, and that it must have an important role in photosynthesis. In addition, we separated the coccolith discs from cells and prepared a suspension of the isolated discs. The condensed suspension of discs provided an optical interference between discs, and the side light illumination was strongly scattered by the discs when the discs were showing Brownian motion in the suspension. It was clearly observed that the applied 5-T magnetic field restrained the light scattering by discs. The bright white color in the suspension changed to blue under the magnetic fields of more than 4 T. The individual color of the discs separated into red, yellow or blue. The obtained results indicated that the aggregation of coccolith discs behaved as photonic crystals because the calcium carbonate crystals were oriented under magnetic fields. It is expected that the coccolith discs can be utilized in the application for bio-sensing phonic devices in the future.

Mitochondrial activity in *Euglena* under light and magnetic field exposures

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Recent drastic climate change and social problems concerning constant fuel supply require a new method for enhancing the production speed of biodiesel, which can be obtained from algae. Also, a huge number of agrichemical and biochemical approaches were applied for the improvement of the process, but few studies were carried out using a physical approach, especially in magnetism. For the purpose of clarifying the responses of green algae to magnetic fields, measurements of the spectral profile of *Euglena gracilis* in near-infrared light regions and microscopic behaviors under magnetic fields of up to 10 Tesla (T) were carried out in this study. *Euglena* culture was filled in an optical chamber, and three optical fibers, a light emitter, a detector and light stimulator, were put on the chamber. The metabolic response in *Euglena* was evaluated by the near-infrared (NIR) lights at 780nm~830nm, which was an indicator of cytochrome oxidase concentration in the mitochondrion of *Euglena*. The magnetic field exposure at 10 T caused a clear increase in the absorbance of NIR lights, and the switching off of the magnetic fields induced a transient decrease which accompanied a negative peak. For comparison, the effect of green light stimulation was measured in the *Euglena*. After the light stimulation, a decrease and a negative peak in the absorbance of NIR lights appeared, and the rate of decrease depended on the intensity of the green light. Also, the result showed a relatively small decrease under a red light stimulation. The results indicated that both the green light and the 10-T strong magnetic fields exhibited a transient decrease in the NIR light absorbance at 780nm ~ 830 nm. *Euglena* shows green-light-taxis, and therefore, we conjectured that the light stimulation induced the acceleration of activity in the mitochondrion. The transient decrease after the light stimulation was probably a rebound of the response to the stimulation. Furthermore, we compared the responses of two kinds of *Euglena* strain, normal type and chlorophyll deficiency type. Interestingly, the chlorophyll deficiency type showed no response to the light stimulations; however, as the normal type, it exhibited the same kind of transient decrease after the magnetic exposure. In order to clarify the mechanism of the *Euglena*'s responses to the physical stimulations, we recorded the motion of *Euglena* under 10-T fields in real-time by using a CCD camera, and an orientation of *Euglena* directing its length perpendicular to the applied magnetic fields was observed. It was speculated that the mitochondria in the *Euglena* were also oriented perpendicular to the applied magnetic fields, and, possibly, the orientation resulted in the observed change in cytochrome oxidase concentration. The most important finding in this study is the similarity of the magnetic field effect with the green light effect in the mitochondrion response.

Proteins from micro- and macro-algae

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In the search for proteins to meet the growing global food and feed demand, novel green sources such as microalgae and macroalgae are of great interest. The microalgae *Chlorella vulgaris* and the macro-algae *Ulva lactuca* can contain up to 60% and 45% of protein based on dry weight respectively. Utilisation of green biomasses will only be economically feasible if a biorefinery approach is applied in which multiple components of interest are extracted, such as proteins for food and feed or lipids for biofuels. In this research project, the development of a mild, continuous and scalable technology for protein extraction while keeping its functionality and minimizing the energy requirements is investigated. The potential of enzymatic cell wall hydrolysis and Pulsed Electric Field will be investigated, since these two techniques are considered mild. The effectiveness of these mild techniques will be evaluated with respect to the proven technique of beadmilling. Subsequent to cell wall disruption, the products of interest need to be extracted by an extraction method able to solubilize both the hydrophobic (e.g. lipids) and hydrophilic (e.g. proteins) compounds. Ionic liquids, surfactants and polymers are able to do so and are as well considered mild extraction methods.

Medium chain carboxylic acid production from non-food feedstock for biomaterials

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Medium chain length carboxylic acids are important building blocks for polymers and resins. Conventional production processes of these chemicals have the disadvantage that petrochemical or food compounds are used as substrate. The use of agricultural residues or municipal solid waste as feedstock for the production of these (bio-based) chemicals is a promising alternative. However, the utilization of these waste streams into bio-based chemicals can only be feasible when an energy efficient process is used. In this project, a novel, economic and eco-friendly bioprocess concept for the production of medium-chain-length carboxylic acids will be developed. The first step in this process involves the anaerobic digestion of low-grade biomass into mainly volatile fatty acids (VFAs). Subsequently, the VFAs are separated and fermented to medium chain fatty acids (MCFAs) such as caproic acid, enanthic acid, caprylic acid and pelargonic acid in a second anaerobic mixed culture fermentation step. Finally, the fermented products are purified in DSP steps. After selecting an appropriate substrate, the two anaerobic fermentation processes are developed and coupled. The final goal is to demonstrate proof of principle for the entire process on lab-scale and to confirm its economic potential. It is important to select the right process conditions to maximize VFA production from the organic waste and to aim for high production rates and high conversion efficiencies. Other design criteria are minimal input of energy and additives as well as minimal formation of waste or co-products.

Tagetes for biocontrol and bio energy

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Search for cost-effective Tagetes cultivation against harmful nematodes, in stead of chemical input by soil fumigation. Tagetes cultivation: Great effect against *Pratylenchus* spp. and *Paratrichodorus pachydermus* for example, but also very cost intensive for farmers. Question: is it an option to produce biogas from the aboveground part of Tagetes, in order to compensate for the high cultivation costs? Research: HLB and Proeftuin Noordbroek performed several field trials with Tagetes on practical scale, to test biomas production and harvesting techniques. Benefits of Tagetes: nematode control (especially *P.penetrans*); soil health (organic matter, soil biology); but also a good energy source? Final results in 2011: Tagetes delivered more than 8 ton dry organic matter/ha, which provides about 3.000 m³ gas. Conclusion: Tagetes appears to be a good and save alternative for chemical soil fumigation, and could be well alternated with energy corn. In corn a number of disadvantages are known for soil, nematode population and landscape quality, which are repaired by Tagetes.

Sustainable processes for solid and concentrated particulate foods

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Sustainable processes for solid and concentrated particulate foods M.A.I. Schutyser, Y.S. Lubbersen, P.J.M. Pelgrom, J. Perdana Food Process Engineering, Wageningen University, Wageningen, The Netherlands Different novel sustainable processes for solid and concentrated particulate foods are presented. In all these processes, the dynamic behaviour of micron-sized particles is critical. Separation processes in food industry are often carried out under extreme conditions with dilute product streams; for example, the wet fractionation of wheat into gluten and starch. Wet fractionation not only leads to excessive energy use, but also negatively affects native ingredient functionality. Separation processes are presented that can fractionate concentrated or dry product streams; specifically, fine milling and air classification for pea protein concentrate production (1) and deterministic ratchets for ingredient suspension fractionation (2). Deterministic ratchets employ periodic arrays of obstacles. The obstacles deflect the particles from the fluid stream and sort particles on the basis of their size. Drying technology is used to manufacture powdered food ingredients or solid foods. Freeze drying is conventionally used for mild drying of heat sensitive ingredients, e.g. enzymes and probiotics. Spray drying is three times as energy efficient, but ingredients may suffer from severe heat inactivation. This quality loss can be minimised if optimum product formulation and spray drying conditions are known. A novel high throughput single droplet experimentation method on spray drying is presented to establish these optimum conditions (3). 1) Schutyser and van der Goot, Trends in Food Science & Technology (2011), 22:154-164 2) Lubbersen et al., Chem Eng Sci (2012), 73: 314-320. 3) Perdana et al., Food Bioprocess Technol (2013) 6(4): 964-977.

Systems Biology Natural Technology Facility; Symbiont – ModuTech

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Symbiont is a facility to design microbiological processes to create scientific breakthroughs in new process principles for food, chemical and environmental industries relying on biomass such as plants, algae, micro-organisms and residual organics, as a resource. As an initial phase of Symbiont, the Modular Technology Facility ModuTech of Wageningen University has been set up. A national and international extension is under development. In the ModuTech facility microbial reactors, online analytical tools, and dynamic modelling and ICT services are provided to talented researchers to design new efficient microbial processes in interaction with innovative start-ups, SMEs and industry, and in cooperation with other universities. ModuTech accommodates experimental technological setups for students, PhDs and postdocs of the Sub-department of Environmental Technology and other research groups with which we co-operate (Attero, LeAF, Waste2Chemicals). This lab consists of 24 experimental cabinets in which custom built reactor systems are prepared by the researcher in close collaboration with the supportive staff. Twelve of these cabinets are temperature controlled and most of them are equipped with additional gas sensing equipment to create safe working spaces for practically work on new approaches and ideas. Within ModuTech 4 large experimental cabins are available for semi-technological research on municipal sewage in set ups up to 200 L. In ModuTech we have continuous access to real wastewater from the sewage treatment plant Bennekom, and tanker stored agro-industrial water, or salt or brackish water to use in research. In the second phase, SYMBIONT will provide a modular high-through-put (HTP) BIOCONVERTER platform for researching the dynamics, regulations and networking in open and defined microbial biomass production and processing systems. The BIOCONVERTER is a facility in which complex bioprocesses are monitored via on line at line and off line analysis to obtain complete information about complex bioprocess such that it is mechanistically understood in order to optimize the bioconversion.

Bodec Process Technology: From knowledge to industrial realization

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For nearly 20 years Bodec is an established name in (mainly) the food industry. The company thinks along in innovative, practical solutions for process development and optimisation. In the beginning of 2013 Bodec will move to the FoodTechPark Brainport in Helmond, where they open their own Pilot & Production Plant, in where optimized processes will be developed. Various technologies will be compared, tested and can even be scaled up to pilot production, what implies that the customer is able to postpone its investment in its own production. Bodec is founded by Paul Deckers & Frank the Boeff and has 25 employees. The development and optimization of production processes and the translation of new innovative technologies into practice (business), this is in short the scope of Bodec. The focus is on isolation, separation, concentration and drying in the food industry. Bodec aspires for the realization of projects that extend from Concept Development, testing, engineering until the complete implementation and optimization of the process, such as valorization projects or even complete new production processes. Bodec works closely together with > 20 universities and research institutes worldwide to be able to closely follow the latest developments. Bringing together knowledge and business could not be fully accomplished without appropriate tools. In the first quarter of 2013 Bodec will be running a flexible Pilot Plant and Tolling Facility specially designed for innovation in food and bio-processing. This new facility is part of the so called Food Tech Park Brainport in Helmond - The Netherlands. The Pilot Plant has a capacity of processing 5 kg/h of dry product. It includes a 6 columns Simulated Moving Bed (SMB) system, an Agitated Thin Film Drier (ATFD), Evaporator and a Spray Dryer (SD). Additional units operations like ultra/microfiltration, rotary disc column, freezecon and freeze drying will be added in the future. The total capacity of the Tolling Facility will be 50 kg/h (figure 1). In parallel with equipment design & operation, detailed dynamic modeling is also being conducted. The idea behind this development is to offer a TOOLBOX with which companies at all levels can challenge their projects and ideas. Such toolbox will allow proof of concept, process development, validation, optimization and scale up studies. It is being thought as a mean for easy access to knowledge and for realization of business opportunities. Our strategy therefore makes possible a solid link between research and industry. Knowledge is taken to industry with a pragmatic approach while industry can access knowledge in a seamless way. Improvement, innovation, cooperation and excellence will remain our main drivers. Figure 1. Bodec's Pilot Plant (5kg/h) and Tolling Facility (50kg/h). Short term scope (blue) and Long term scope (orange).

Hydrothermal Pretreatment of Wheat Bran

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While the world demand for energy carriers and commodity chemicals increase continuously, the main resource - fossil fuel - covering this demand becomes scarce. Despite the new discoveries of fossil deposits, the total amount of it on earth is finite and the complete depletion is expected within a foreseeable future. It is inevitable that the global economy switches to a sustainable system utilizing renewable resources. The production of energy and industrial products via biochemical conversion of glucose derived from corn has been investigated and partially industrialized over the last decades. However, the concern on sustainability of food crops for nutrient purposes favors the concept of second generation biomass utilization. In this respect, sustainable biorefineries, which enable the utilization of multiple biomass fractions, will take the key position. For this purpose, various lignocellulosic feedstocks (e.g. wood, straw, corn stover, etc.) are in the focus of interest of research and development. In this context, wheat bran may also possess the appropriate characteristics for utilization within a biorefinery. As a by-product of the production of white wheat flour, it accrues in enormous quantities and hitherto it is mainly used as a low cost ingredient in animal feed. Comparing the composition of wheat bran with that of other commonly considered raw materials (wood, straw, etc.), some differences become evident. While bran contains a lower amount of cellulose and lignin, it contains a reasonable high amount of proteins, starch and minerals. On the one hand these aspects could add an extra value to bran when used as a feedstock, but on the other hand it could also complicate the necessary technological biorefinery system. Whilst the enzymatic hydrolysis of the starch fraction can rather easily be done by commercially established enzyme processes, the disintegration of more recalcitrant fractions like cellulose, hemicellulose and to some extent also proteins requires harsher methods. For this purpose, various so-called pretreatments (hydrothermal, steam explosion, acidic, organosolv, etc.) are commonly proposed. All these methods bear specific advantages and disadvantages. As relatively little has been done on this specific field with wheat bran as a feedstock and as the effects of the pretreatments vary widely with the used raw material, experimental trials are essential. The hydrothermal pretreatment has been investigated in conjunction with an enzymatic hydrolysis. Using a 2 L batch reactor, temperatures ranging from 140 to 200°C and reaction times between 10 and 30 min have been applied. Enzymatically, a glucose yield of 90% and a hemicellulose hydrolysis of approx. 50% could be achieved. About 70% of the proteins could be dissolved; however, very little amounts were obtained in the form of free amino acids. The formation of undesired degradation products inhibiting the further biochemical conversion such as furfural starts at 180°C.

Production of Pure Microbial Oil (PMO) with oleaginous yeasts

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A potentially promising alternative for the production of Triacylglycerides (TAG) could be the use of oleaginous yeasts that produce Pure Microbial Oil (PMO). These micro-organisms have substantial advantages compared to other production systems; - High volumetric production rates (doubling time of two hours, > 60 g biomass/kg broth). - More efficient use of rural area (vertical versus horizontal cultivation). - No competition (yet) with the food market. These second generation bio fuel producing micro-organisms have a high potential for the use in the bio based economy (bio diesel, cosmetics, paint, fine chemicals, etc.). After initial shake flask screening on glucose, seven promising strains were selected and screened further on their robustness with respect to cultivation parameters (pH and T). Altering the medium composition to higher C/N ratios results in nitrogen starvation and forces the cells to produce lipids and glycogen. Various carbon sources were tested on their ability to be metabolised by these strains. Selection of the desired strains was based on several parameters; - Ability to grow at a broad T-range and at low pH-values - High percentage of fatty acids in the cells - Low percentage of other storage carbohydrates (e.g. glycogen, trehalose) - Ability to grow on a broad range of carbon sources. Results will be presented of this research.

Mono-digestion of dairy manure: energy from waste or waste of energy?

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Our current economy is relies gravely upon the usage of fossilised resources for its energy supply and for the production of chemicals and materials (such as plastics). The reserves of fossil fuels, however, are running thin and a transition towards a biobased economy which makes use of biomass as a renewable resources of biological origin is necessary. The province of Fryslân wants to provide for 8% of its total energy consumption with biogas produced by anaerobic digestion (AD). Fryslân is a highly agricultural region with a strong dairy industry and thus has access to vast amounts of biomass in the form of grass and dairy cattle manure. With the lifting of the European milk quota in 2015 it is expected the dairy industry will grow, resulting in an increase in available biomass for AD. However digestion of manure is economically unfeasible and suffers greatly from ever increasing prices of codigestion products. Pretreatment technologies could prove the key to cracking the lignocellulosic fibers in dairy manure and thus increase biogas production.

Production of lactic acid using lignocellulosic biomass as feedstock: Effect of inhibitory side products

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Plastic is one of the main sources of oil consumption. One of the mostly produced plastics worldwide is Polyethylene Terephthalate (PET), with a market of 60 million tonnes per year (2010). PET is produced on a large scale via petrochemical processes. Polymers of lactic acid molecules (PLA) can be used as a biobased replacement for PET. PLA has many similarities with PET in structure and behaviour, while it can be either recycled or composted to minimize waste production. Also, PLA does not require the input of oil, which is beneficial for the environment. Lactic acid molecules, which are required for the production of PLA, can be produced via fermentation processes. Lignocellulose is an interesting starting material for this process. Lignocellulose is the most abundant biopolymer on earth, since is present in nearly all plant material. Lignocellulose consists for roughly 60-75% of sugar molecules. An interesting source of lignocellulose is agricultural waste material such as corn stover, straw or sugarcane bagasse. Although lignocellulose consists mainly of sugar molecules, the sugar is tightly packed in the lignocellulosic structure, and therefore not accessible for micro-organisms used in the fermentation. A pretreatment is required to acquire the sugars in a fermentable state. In this pretreatment, two stages convert the sugar polymers to monomers. The first stage of the process is a chemical pretreatment, which is followed by an enzyme treatment. Due to the severe conditions in the chemical stage of the pretreatment, not only monomeric sugars are formed. Lignin can be degraded to phenolic polymers and monomers, while hemicellulose is deacetylated. Depending on the exact process conditions, monomeric sugars can be dehydrolysed, which forms furanic compounds. These compounds can significantly decrease the growth and productivity of micro-organisms when they are present during fermentation. In this project, lignocellulosic substrate is analysed for the presence of side-products formed during pretreatment. These compounds will be tested for inhibitory effect on micro-organisms. Based on their abundance in the lignocellulosic derived substrate and toxic effects on lactic acid producing micro-organisms, key compounds will be identified. With this information, methods can be developed to overcome the inhibitory effect of these compounds. To determine the composition of the lignocellulose material after pretreatment, different methods have been set up. An UPLC-MS method has been developed to measure the phenolic compounds. Furthermore, acids are measured using a different HPLC method, while sugars are measured using HPAEC-PAD. Currently, a new method is set up for the determination of level of toxicity of inhibiting compounds on lactic acid producing bacteria using small volumes.

SOLANYL and FLOURPLAST thermoplastic starch based plastics and OPTINYL masterbatches: CREATING NEW OPPORTUNITIES FOR THE BIOPLASTIC INDUSTRY

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Rodenburg Biopolymers developed over the last year various bioplastic materials to their portfolio of products consisting of FlourPlast precompounds, Solanyl end-compounds and Optinyl masterbatches. This paper describes an outline of the structural features (using SEM, WAXS and other advanced techniques) and various properties of products containing compatibilised thermoplastic starch and flour based compounds (i.e. Solanyl) or precompounds (i.e. FlourPlast). Grain or cereal flour or even purified starches are themselves not thermoplastic materials. The FlourPlast and Solanyl portfolio of products are based on thermoplastic flour (TPF) or starch (TPS) and are made from a unique combination of natural based grain by-products or reclaimed starch based sources from the food processing industry and a novel compatibilising polymer system making it thermoplastic materials, which can be processed on standard plastic processing machines. The FlourPlast portfolio of products is as such shown to be highly compatible with natural or petrochemical based biodegradable aliphatic (co-) polyesters and various polyolefins such as polypropylene giving the opportunity to make dedicated bioplastics. By combining FlourPlast with other plastics, it is shown that improved processing conditions are obtained and enhanced or novel properties of the end formulations (compounds). By making different combinations of the various grades of the FlourPlast (i.e. building block system of precompounds) with other polymers it will be shown that it is possible to obtain a range of products with different properties and good functionality. This made it possible to process the components into products by the compounding industries suitable for various applications such as injection molding, extrusion and thermoforming, and film blowing and casting. The Solanyl portfolio consists of ready-to-use biodegradable and biobased compounds directly suitable for converters to obtain biodegradable or biobased products. The portfolio consists of various compounds suitable for injection moulding (the C1*** series), (sheet) extrusion and thermoforming (the C2*** series), and film extrusion (blowing or casting) (the C8*** series). The various compounds offer a complete portfolio each compound having dedicated properties similar to LDPE, HDPE, PP or PS. Specially designed for FlourPlast, Solanyl or other biopolyester plastics Rodenburg Biopolymers offer Optinyl masterbatches offer the opportunity for converters to fine tune other properties such as colour, flow, impact and many more. Some remarkable examples are described.

Sustainable production of food and fuel in Mali

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In recent years, there has been an increasing concern about the growing demand for agricultural land for energy production in the developing world, especially in Africa, by foreign investors and the potential risk this may have for the food production. However, there is also a growing interest in many developing countries to attract foreign direct investment as a basis for reinvigorating agricultural development. Though many studies have been carried out showing a negative impact of foreign direct investment on smallholders, Mali Biocarburant (MBSA) presents an “inclusive business models”, where food and energy security are promoted through agroforestry production systems. Mali Biocarburant SA (MBSA) is a private company with smallholders as shareholders that produce biofuel in a way that supplements farmers' incomes, contributes to poverty alleviation and respects the environment. Since 2007, the company has been working with more than 8000 small *Jatropha curcas* (*Jatropha*) farmers in three regions of Mali and two regions in Burkina Faso. MBSA focusses on local production, local processing and local consumption. The company encourages small-scale farmers in Mali to intercrop their fields with *jatropha*. Farmers harvest the *jatropha* nuts and sell them to MBSA, which then extracts their oil. MBSA refines the oil into biodiesel, which is then sold locally and used to power generators and cars that have diesel engines. The extraction process produces residual “press cakes” that are used to produce electricity while the residual organic fertilizer helps to improve soil fertility and glycerin that is used to produce soap. This bio-fuel model integrates *Jatropha* production into the smallholder farming system. A large number of extension staff train farmers to improve food production by introducing drought tolerant maize varieties and/or improved sweet sorghum varieties and they helping them to have access to credit. This value chain approach has led to increased availability of food, while only the toxic oil seeds of *jatropha* are used for bio fuel production. Further, the agroforestry system results in increased food production through reduced soil erosion, water infiltration and better soil structure. As a next step the company is leading a consortium that will embark on a programme to set up small scale bio refinery systems of local food, feed and fuel production. This leads to increased availability of sweet sorghum grains, animal feed from the sweet sorghum stalks, while the leaves used to produce proteins for improved food quality. Finally, 2nd generation biofuel processes will be tested for bio-ethanol production from cellulose. Conclusion: MBSA shows that if the appropriate business model is chosen that both food and energy security can be promoted.

The C1-technology platform for versatile, robust enzymes enabling a variety of second generation biofuels and chemicals processes

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Dyadic develops the filamentous fungus *Myceliophthora thermophila* C1, as a proprietary protein expression platform for the efficient production of enzymes. Recently, re-sequencing of the C1 genome has been completed and approximately 10.000 genes were identified by automated annotation. At present, this knowledge is being exploited to improve the performance of C1 production strains and the enzymes produced by dedicated genetic modification. The annotated genome of C1 revealed an impressive number of carbohydrate active and oxidative enzymes, which in an appropriate composition can decompose lignocellulosic biomass completely. This notion prompted the development of C1 as a producer of low-cost enzyme mixtures for the conversion of lignocellulosic biomass into fermentable sugars for biofuel and chemicals production processes. Such processes are very diverse as a result of the many different types of lignocellulosic biomass, pretreatment procedures and microbial conversion systems. An enzyme system that is robust and highly active on a variety of lignocellulosic biomasses is therefore imperative for successful broad application. By recruiting the genes from C1 which were shown to encode the most effective carbohydrate active enzymes in dedicated production strains, high quality and cost-effective enzyme mixtures were developed. These enzyme mixtures efficiently converted a variety feedstocks, e.g. corn stover, wheat straw, sugar cane bagasse and paper waste, into fermentable sugars. Importantly, the enzyme mixtures developed showed high activity at broad temperature and pH-ranges, enabling their use in non-conventional biofuels and chemicals processes. In particular the thermophilic nature of C1-enzyme mixtures in combination with their activity at higher pH was shown to be a unique advantage over the traditional lignocellulosic enzymes produced by other fungi, like *Trichoderma*. An important aspect of the C1-enzyme mixtures developed is that they are produced by a single strain, facilitating the enzyme production process and its implementation on site. An overview will be given covering the latest results of the development of C1 as a producer of enzymes that can be applied in a range of lignocellulosic biorefinery concepts.

Combining a bioanode and biocathode in a Plant Microbial Fuel Cell for sustainable electricity production

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The plant microbial fuel cell (PMFC) produces bioelectricity from solar radiation. Plants grow in the anode compartment and excrete organic matter via photosynthesis. Electrochemically active bacteria break down this organic matter and produce protons, electrons and CO₂. The electrons are collected by the anode and transported via an external circuit to the cathode, where preferably oxygen is reduced to water. In previous research, often unsustainable ferricyanide is used as the electron acceptor. The reduction of oxygen is very slow and limits the performance of the PMFC. However, the reduction reaction can be catalysed by microorganisms. The objective of this research is to show that a bioanode can be combined with a biocathode to produce sustainable electricity with a PMFC. For the experiment three flat plate cells were constructed. Each cell had one anode compartment with a size of 19 x 3 x 19 cm (l x w x h) and a planting area of 57 cm². Two cathode compartments were connected to each cell on both flat sides of the anode. The anode and cathode were separated by a bipolar membrane (BPM) to minimize unwanted crossover of ions and to maintain the pH difference between both compartments. The anodes were made of graphite felt. Two cells had also graphite felt as cathode material and one cell had platinum coated titanium cathodes as a reference at pH 7. The graphite felt cathodes were inoculated with mixed culture microorganisms. The pH in the cathodes of one cell was controlled at pH 2, in the other at pH 5. The salt water plant *Spartina Anglica* was planted in the anode compartments and kept waterlogged (anaerobic) at all times. The catholyte was continuously aerated and recirculated. The anode, cathode, membrane and cell potential were logged every minute. The cells were controlled at 1000 Ω. Regularly polarization experiment were performed with resistance boxes and a potentiostat from OCV down to an external resistance of 1 Ω. Polarization curves before and after the inoculation of microorganisms showed that the microorganism catalysed the reduction of oxygen. For both cells with a biocathode the maximum power output increased by an order of magnitude to 0.329 W/m² for the pH 2 biocathode and to 0.597 W/m² for the pH 5 biocathode. The maximum power output of the platinum reference cell was clearly lower (0.047 W/m²) indicating that the biocathode catalysed the oxygen reduction. This indication was confirmed by the resistance of the cathode which decreased eight times for both biocathodes after inoculation, while the reference cathode had the same resistance at both times. The performance of the anodes decreased during the experiment, mainly due to oxygen diffusion through the membrane which resulted in an high anode potential. The plants in the anode connected to the biocathode at pH 2 died, likely due to a low pH of 3 in the anode, indicating that the BPM was not able to maintain the pH difference between both compartments.

Combined oil production and protein extraction from rubber seed

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Rubber trees are mainly cultivated for latex production. The seeds are rarely used except selectively in small amounts for seeding. Rubber seed kernel contains 48% oil and 18% protein. Oil is the largest fraction and has potential applications, including drying oil and biodiesel. Oil production from rubber seed will also produce a residue with high protein and amino acids content that has potential value for use as animal feed or as a starting material for biochemicals. Valorizing this fraction will increase the benefit in the entire chain, including oil and latex production. Protein was extracted from rubber seed kernel, with or without prior oil removal. Before treatment, the kernels were dried at 60°C for 72 hours or 105°C for 24 hours. Protein extractions were performed using 0.1M NaOH. At 25°C, 0.10 g-protein/g-biomass was extracted from full-fat rubber seed kernel at 24-27% purity. Increasing the temperature to 60°C did not significantly improve the yield and purity. In a second type of experiments, oil was removed from the kernel before protein extraction. For this, hydraulic pressing (60°C and 100°C, 25 MPa) and solvent extraction in a soxhlet unit (n-hexane, b.p., 6 hours) were applied, resulting in press cake (26-41% oil, 19-22% protein) and meal (3-14% oil, 23-27% protein). Protein extractions were performed at the same conditions as protein extraction from the kernel. The yield from the press cake and meal were 0.11-0.15 g-protein/g-biomass (30-40% purity) and 0.12-0.13 g-protein/g-biomass (36-37% purity), respectively. Press cake that was pre-dried and pressed at 60°C gave the highest yield and purity compared to the other materials. The yield and purity were only slightly improved when protein extraction temperature was increased from 25°C to 60°C. Our study clearly showed that oil removal improves subsequent protein extraction from rubber seed kernel compared to kernels with oil. However, temperature and heating duration of the pre-drying and the method of oil removal also affected the results of protein extraction from these materials.

Minifors 'Biobased': saccharification and subsequent fermentation at laboratory scale

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Lignocellulosic (waste) streams are increasingly used as feedstock for the industrial production of "green" chemicals or fuels through fermentation. "Bio-based" processes, such as SSF (simultaneous saccharification and fermentation) or CBP (consolidated bioprocessing), generally involve high solid loads. Although this may not be a major issue on a large scale, on a laboratory scale the limitations of the currently available equipment readily become apparent. Combinations of e.g. different lignocellulolytic enzymes, duration of the process and specific process conditions applied, together determine the amount of sugars released (e.g. glucose, xylose and arabinose). Because saccharification efficiency has great influence on both final yields of desired fermentation products and overall process economics, researchers generally invest a lot of effort in optimizing this part of the process. To find the optimal conditions for saccharification of a specific lignocellulosic waste stream, regular fermentation systems designed for low- or average viscous liquids, are commonly used. These systems however, are less suitable to mix viscous media or media with high solid loads (e.g. 10-40% dry matter). Poor mixing may result in prolonged saccharification processes or even in incomplete saccharification. For optimal conditions during both saccharification and fermentation currently separate vessels are needed. However, a single unit that can be used for both processes is often preferred. The recently developed Minifors 'Biobased' (Infors HT Benelux, Doetinchem, NL) now offers a unique platform for handling high solid loads at laboratory scale. The system is designed for both, saccharification and fermentation of high solid feedstock in a single unit, either simultaneously or sequentially. The system features a stronger engine, specifically designed impellers for improved mixing of high dry weight contents and an improved heating and cooling system. In this poster, the performance of the Minifors 'Biobased' is evaluated by performing enzymatic hydrolysis of pretreated wheat straw and subsequent fermentation to ethanol by yeast. The Minifors 'BioBased' provides a robust platform for both, hydrolysis purposes of feedstocks with solid content up to 30% (w/w) and (semi-) anaerobic microbial cultivation in a single vessel. Furthermore, the Minifors 'Biobased' offers a broad temperature range, allowing for flexibility in terms of thermophilic hydrolysis and/or fermentation. Additionally, high solid loads can be successfully mixed in the Minifors 'Biobased', in contrast to standard fermenters.

Innovative production of lactic acid from lignocellulose and starch

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Oil palm empty fruit bunch (EFB) was hydrolyzed to get hemicellulose sugars by the combined use of dilute H₂SO₄ and H₃PO₄, which has been shown to have a synergistic effect in improving xylose yield compared to using H₂SO₄ alone. Hemicellulose sugars reached >100 g/L in the hydrolysate without any additional concentration steps. Furfural, 5-hydroxymethyl furfural and acetic acid in hydrolysate were efficiently degraded by simply adding the microorganisms that were isolated from the nature in Singapore. Cellulose was hydrolyzed to glucose by using whole cell cultures without supplementing any exogenous enzymes, which was further enhanced by simultaneous saccharification and fermentation. Thermophilic bacteria were isolated from the natural environment and used to convert all lignocellulose sugars to L-lactic acid at 50°C in a simple medium with high yield (>95%), titer (>200 g/L) and productivity (>5 g/L/h). Genetic modifications of the isolates are under way to construct mutants to produce D-lactic acid. The isolated thermophilic bacteria also have a great potential in industrial production of L-lactic acid from starch as they are able to directly convert starch (up to 200 g/L) to L-lactic acid in 1-pot, 1-step simultaneous saccharification and fermentation process at 50°C without the requirement of sterilizing the medium.

Bioeconomy Innovation Cluster

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European Society faces a huge looming crisis of resources. Europe is especially dependent on importing oil and minerals to feed its industries. Yet Europe has the highest consumption of raw materials per capita in the world. As these traditional resources become scarcer, their availability will become more politically controlled, leaving the EU vulnerable to highly politicised negotiations and pricing. Within a successful Bioeconomy the biomass resources have to be produced sustainable and have to be synergetically converted into both human food, animal feed, chemicals/polymers, materials, fuels, power and/or heat by means of high-efficient bio-cascading and biorefining processes.

For the time being, biomass resources are often cultivated unsustainable, whereas they are used for food OR non-food applications, and in the latter case most often only for relatively low-value animal feed and energy applications. Implementation of high-efficient sustainable biorefining chains, are only taking place at a slow and low level, mainly caused by:

- The current state-of-the-art and time-to-market of some of the key chain composing technologies –biomass fractionation & product separation technologies, advanced conversion technologies, and downstream processing technologies of biomass intermediates to marketable products.
- The lack of co-operation between: i) stakeholders covering the full biomass value chain from biomass production till end-use, ii) stakeholders normally operating in separate market segments (not knowing each other), and iii) stakeholders with different expertise and roles (industry/SMEs, research institutes/universities, (regional) governments) necessary for successful technology development, deployment and final implementation.
- Suboptimal or not using available expertise, facilities, and funding opportunities at a regional level.
- A lack of co-operation at an international (European) level.

The bottom-up co-operation of triple-helix partnerships – government, industry/ SMEs, institutes/universities – at regional level offers the synergetic deployment force of budgets, policies, entrepreneurship, and innovative knowledge/technologies to facilitate the development and implementation of high-efficient sustainable biorefining chains. Bioeconomy innovation cluster (BIC) aims to promote the transition towards bioeconomy, focusing on regional research and strengths at first instance as well as technological development of concepts, technologies, processes and full value chains for efficient and sustainable biomass production and use for both food and non-food applications. BIC covers variety of biomass production and valorisation, aquatic biomass (microalgae, seaweeds), and primary (agro), lignocellulosic and fresh biomass as well as manure and sludge to (co)produce human food, animal feed, chemicals, materials, fuels, power and/or heat. High-efficient and sustainable use of the biomass resources by applying both biocascading and biorefining principles, including the improvement of already established biomass valorisation processes (more efficient in terms of water, energy and chemicals) and shared facilities forms the major focus of the innovation cluster.

The activities within the cluster are organised in programs relating with the type of raw materials allowing development of raw material specific chains, partnerships and technologies. Furthermore, it is facilitated that there is interaction between these programs to make cooperation possible also between different value chains, cross contamination leading to value added applications, efficiency in developing generic technologies and minimised risk in investments in facilities and finally implementation.

The chemical pretreatment proces and enzymatic hydrolysis of hardwood derived from arable land

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The limiting factor for effective conversion of lignocellulosic feedstock into ethanol is a complex structure of biopolymers, including the presence of lignin which is highly recalcitrant to biodegradation. The process of enzymatic hydrolysis is not sufficient to attain an acceptable level of fermentable sugars. Therefore, for achieving high fermentability of the wood hydrolysates, a pretreatment process is a required step for the bioconversion of lignocellulosic biomass before the enzymatic hydrolysis process. The objective of this study was to evaluate the effectiveness of two methods of chemical pretreatment: alkaline and acid hydrolysis. In this thesis the various lignocellulosic feedstock derived from arable land - poplar, willow and acacia - were compared. These plants belong to the group of hardwoods with 40-50% of the cellulose, 12-23% of the hemicelluloses, and 16-20% of lignin of the dry matter (DM) of wood chips. The other soluble compound and ash have been studied at the level of 13-23% DM and 15-25% DM, respectively. Additionally, in the resulting wood hydrolysates the water-soluble carbohydrate (WSC) content was investigated using the anthrone method. The level of WSC in the hydrolysates after pretreatment processes occurrence was about 20-30% DM. In the case of alkaline hydrolysis with NaOH it has been shown that the hemicelluloses fraction mainly dissolves to about 5% DM. Compared to alkaline hydrolysis pretreatment, acid hydrolysis with H₃PO₄ acid has been proven to be more efficient for treating lignocellulosic materials. In the solid residues after dilute acid hydrolysis of hardwoods there was an observed decrease in the hemicelluloses and celluloses fraction to 0,21% DM (in the case of the poplar hydrolysates) and to 27,73% DM (in the case of the willow hydrolysates), respectively. It should be noted that in some hardwood hydrolysates there was an observed reduction of lignin to about 4% DM. However, the various compounds alongside fermentable sugars, which are formed by degradation of lignin, may interfere with the fermentation process.

Cost-efficient protein extraction from tea leaf residues

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Leaf protein (LP) is considered to be a potential resource for feed or food since 1960s, but its applications are limited due to high proportion of insoluble protein and low cost-efficient processing. To overcome these problems, the alkaline extraction method and correlated parameters are re-evaluated by using green tea leaf residue (GTR) as starting material. In our study, protein extraction of GTR could be maximized up to 95%. In the extraction, temperature and NaOH are crucial and interacting parameters that affect extraction efficiency and final protein molecular weight. By controlling the process with these two parameters it is possible to get high recovery of proteins in the extracts that are still at high molecular weight, which benefits the subsequent recovery. The economical optimal one-step method produces 53% pure protein from tea leaves at only 130€/ton product. In addition, when Ca²⁺ was introduced in the processing, protein product with very light color can be obtained. This new technology can be also applied to other leaves, which increases the chances for cost-efficient applications of leaf protein.

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