

# Celignis USA



# Biomass Analysis Services

FROM A LAB DEDICATED TO  
ADVANCING THE BIOECONOMY

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# Celignis USA

After 11-years of succesful operations in Ireland, serving over 1,000 clients globally, Celignis is pleased to announce the opening of Celignis laboratories in the USA, operated by Celignis USA Corporation.

Celignis USA is a dedicated service provider for the bioeconomy. We provide our clients with the most precise compositional data and highly-informed process expertise in order to allow them to make the best use of their biomass feedstocks and optimise their biomass conversion processes.

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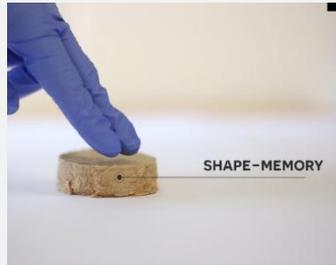


## ADVANCED BIOFUELS

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We can help to determine the value of your feedstock for the production of advanced biofuels. We determine sugars in cellulose and hemicellulose, as well as lignin, extractives, and ash. We provide data in one day with our unique rapid analysis models.



## BIOMATERIALS & BIOCHEMICALS

page

10-11

We are experts in the extraction of biopolymers from biomass and then in modifying these to obtain materials with diverse functional properties. Our team also uses advanced analytical equipment to find high-value biochemicals in feedstocks.



## BIOPROCESS DEVELOPMENT

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We provide valued analytical services to industrial and academic clients across the globe. We also have a top-class multidisciplinary team that can work with our clients on optimising their biomass valorisation technologies.



## Our Philosophy

“We believe that when people have accurate and comprehensive data the opportunities are limitless”



Celignis was born from research that targeted replacing fossil-fuels with sustainable biofuels. We found that feedstock composition was crucial, but that literature data could be highly misleading. There was a critical need for accurate analysis. We are driven to provide the best possible data and want to play our part in the development of the bioeconomy.

*Dr Dan Hayes, President of Celignis USA Corporation.*



### RENEWABLE NATURAL GAS

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16-19

We determine many properties relevant to the anaerobic digestion of biomass. These include the biomethane potential and the composition of the digestate. We also provide bioprocess consultation services to improve digestion efficiency.



### BIOCHAR ANALYSIS

page

20-23

These pages detail our wide-ranging analysis packages for the evaluation of biochar and pyrolysis feedstocks. We are available to discuss these results with you and suggest suitable applications for your biochar and potential process optimisations.



### SEAWEED COMPOSITION

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26-27

Seaweed has huge potential for the bioeconomy. We understand the unique chemistry of seaweed and the potential applications of polysaccharides and other components, and offer many seaweed-specific packages for sample evaluation.



## BIOMASS FEEDSTOCK ANALYSIS

# 1. Advanced Biofuels Feedstocks

**RELEVANT ANALYSIS  
PACKAGES:**
**P4 - Ethanol Extractives**
**P5 - Water Extractives**
**P7 – Lignocellulosic Sugars:**

Glucose, Xylose, Mannose,  
Arabinose, Galactose,  
Rhamnose

**P8 – Lignin Content:**

Klason Lignin, Acid Soluble  
Lignin, Acid Insoluble  
Residue, Acid Insoluble Ash

**P10 – Sugars, Lignin,  
Extractives, and Ash**
**P270 – Protein-Corrected  
Klason Lignin**
**P11 – NIR Prediction**
**P14 – Starch Content**
**P15 – Uronic Acids:**

Glucuronic, Galacturonic,  
Mannuronic, Guluronic, 4-O-  
Methyl-D-Glucuronic

**P16 – Acetyl Content**
**P17 – Biomass Amino Acids:**

Alanine, Arginine, Aspartic,  
Cystine, Glutamic, Glycine,  
Histidine, Isoleucine,  
Leucine, Lysine, Methionine,  
Phenylalanine, Proline,  
Serine, Threonine, Tyrosine,  
Valine

**P18 –Lipids as Fatty Acids**
**P19 – Deluxe Lignocellulose  
Package**
**P20 – Lignin S/G Ratio**


We can determine all the important parameters for the production of chemicals and advanced biofuels from cellulosic biomass.

Second-generation biofuels, such as cellulosic ethanol, offer huge potential in substituting for fossil-derived transport fuels. Similarly, biorefineries could produce a range of sustainable chemicals and bio-products from low-cost lignocellulosic biomass. The number of suitable feedstocks is massive and includes energy crops, agricultural residues, and municipal wastes. There can be huge variations in composition between different feedstocks and also within the same feedstock grown in different locations and under different conditions. Thus, it is crucial to use a laboratory experienced in the detailed and complex methods of analysis required to fully characterise these materials.





We provide all lignocellulosic analytical data in duplicate so you can see the precision of our work

## EXTRACTIVES

These are non-cell-wall components that can be removed using various solvents. Extractives can vary greatly in their compositions and amounts according to the feedstock and its stage of life. We recommend that extractives are removed prior to undertaking the lignocellulosic analysis of samples. We can use water, ethanol, or other solvents for extraction and can determine 14 different water-soluble carbohydrates present in the liquid extract. We also offer detailed analysis of the constituents in ethanol extractives.

## STRUCTURAL SUGARS

In lignocellulosic biomass the main structural polysaccharides are cellulose and hemicellulose. These are often the most important constituents when estimating potential cellulosic ethanol yields from the biological conversion of biomass.

We can determine the glucan content of biomass, a good estimate for the cellulose content, and we can also analyse for five other sugars present in hemicellulose (xylose, mannose, arabinose, galactose, and rhamnose) as well as uronic acids (galacturonic, glucuronic, mannuronic, guluronic, 4-O-Methyl-D-Glucuronic) and acetyl content.

## LIGNIN

This is a structurally important polymer in biomass and is often the solid residual output of biorefineries after the polysaccharides have been hydrolysed. It can be combusted or used as a feedstock for the production of chemicals and biofuels. In our acid hydrolysis process for liberating the structural sugars, we obtain Klason lignin as a solid residue and also acid soluble lignin which we determine using ultraviolet spectroscopy. With package P270 we can correct the lignin content for residual protein after hydrolysis.

## STARCH

Starch is a glucan polymer so we recommend, for relevant samples, that starch content is analysed to differentiate between lignocellulosic and starch-derived glucose.

## PRETREATED

Biomass pretreatment is a crucial step for the production of advanced biofuels and chemicals. There are a large number of different processes that can be used and a wide spectrum of potential products. We have a suite of analysis packages designed to fully evaluate the efficiency of pretreatment so that conditions can be appropriately engineered for the particular feedstock and desired end products.

In particular, the starting feedstock should be characterised in detail so that the different sources (e.g. lignocellulose, starch, extractives etc.) of sugars that may be liberated in pre-treatment are known. We strive to get as close to mass closure as possible for the whole pre-treatment process and this involves analysing in detail both the liquid and solid outputs.

## We are the Only Company to Provide Lignocellulosic Data within One Day and at Low Cost!

We can follow standard chemical analysis methods for determining the lignocellulosic composition of biomass. However, these chemical analysis methods can be slow, taking up to two weeks for a sample, as numerous steps and items of equipment are involved. To date, the length of this analytical process has meant that the number of samples that can be analysed has been restricted by time and by finance limitations.

But we at Celignis have the solution! As an alternative to our chemical analysis packages you can get your solid biomass samples analysed using our unique rapid-analysis Near Infrared (NIR) method. This involves us scanning your sample and then applying our proprietary algorithms to predict the content of 13 different lignocellulosic parameters. This means that we can provide you with data within one day for as low as \$70 per sample. No other company is able to provide this service for advanced biofuel feedstocks.

### CHEMICAL ANALYSIS



### CELIGNIS RAPID ANALYSIS



You are no longer limited in the number of samples you can evaluate!

# Benefits of our Rapid NIR Method

## 1 ACCURATE – DATA YOU CAN RELY ON

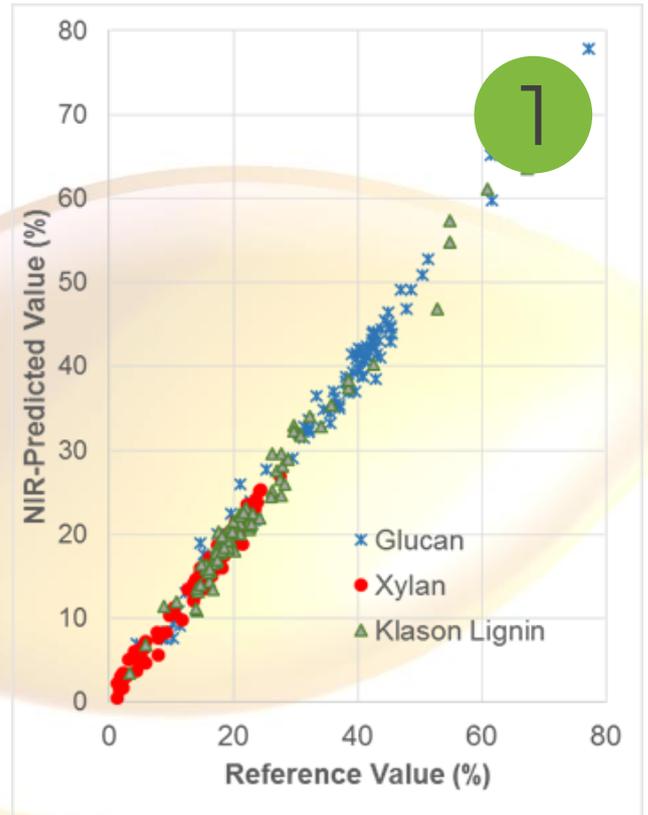
When the data from our NIR prediction models were compared with data from standard chemical analyses we found a very close correlation ( $R^2$  of  $\sim 0.97$  for the main constituents of lignocellulose). Furthermore, as well as providing data for the predicted composition, our NIR package also provides an estimate for the error (deviation) in the prediction and, if we find this to be high, we will undertake the chemical analysis at no extra charge.

## 2 SUITABLE FOR ALL BIOMASS TYPES

Our models have been demonstrated on thousands of samples covering a wide variety of feedstocks including energy crops, crop residues, organic wastes, pre-treated samples, and process residues.

## 3 FITS WITH YOUR REQUIREMENTS

The rapid turnaround and low cost mean that you can quickly see the value of your samples or process conditions and make responsive decisions or modifications accordingly. You can also analyse many more samples than before, improving your chances of finding the optimal sample!



## BIOMASS FEEDSTOCK ANALYSIS

## 2. Analysis for RINs Credits



Celignis USA has expertise in chemical analysis for a wide variety of feedstocks. We can help our clients to determine cellulosic RINs for EPA unapproved D3 feedstocks or mixed feedstocks for liquid biofuels and biogas.

Since the early 2000s the USA has seen a rapid expansion in biofuel production. While most is sourced from corn, most future biofuel facilities will process cellulosic feedstocks (e.g. wood, grass, agricultural residues etc.). There are major financial supports (e.g. D3 RINs) for such facilities, oftentimes dependent on the cellulosic proportion of the feedstock. The Celignis family has analysed tens of thousands of samples for their lignocellulosic composition, using methods approved by the US EPA. Our reports provide in-depth compositional data as well as important summaries (e.g. adjusted cellulose content) required under the RFS.

We have provided services to many US start-ups and multinationals looking to develop technologies to valorise lignocellulose, with an array of analysis and bioprocess development services focused on understanding pre-treatment, hydrolysis, and fermentation processes. We also recognise the potential for higher-value (non-biofuel) products from biomass and have helped clients to understand the most profitable valorisation approach for a given feedstock or technology.

# How our Analysis Can Help with RINs Credits Applications and Renewable Volume Obligations

## ADJUSTED CELLULOSE

Our methods of lignocellulose analysis are approved techniques according to the EPA guidelines on determining adjusted cellulosic content.

We differentiate between structural sugars (such as those in cellulose and hemicellulose) and those present in sucrose, starch, and the extractives and report each of these structural sugars individually, as well as the lignin content (as both Klason and acid soluble lignin).

Our online Database and Excel and pdf reports also sum these structural sugars (i.e. cellulose and hemicellulose) along with the lignin content and express the result (the adjusted cellulosic content) on a dry-ash-free basis so that you can see if the sample meets the 75% threshold or if partial allocation of D3 RINs, based on the the proportion of total mass that is cellulosic, is required.

## BIOGAS FEEDSTOCKS

We can determine the percentage cellulosic RINs and maximum achievable RINs for biogas production from D3 and D7 unapproved feedstocks (i.e. RFS EMTS (EPA Moderated Transaction System) reporting codes 335 and 336) by doing biomass composition analysis and biomethane potential (BMP) tests.



BIOMASS VALORISATION

# 3. Advanced Biomaterials



Advanced biomaterials are designer materials developed by modification and functionalisation of polymers derived from biomass. They are playing important roles in many sectors, from packaging to tissue engineering.

The types of biomaterials range from bioplastics to hydrogels and aerogels. Design and characterisation of these materials require trans-disciplinary knowledge of biomass chemistry, chemical engineering, materials engineering, and molecular chemistry. Celignis’s multi-disciplinary team has successfully designed biomaterials from marine and terrestrial biomass for clients and in a number of research projects.

## 1 EXTRACTION AND PURIFICATION OF BIOPOLYMERS

With the expertise of the Celignis team in biomass chemistry, we design and develop processes for extraction and purification of biopolymers. It involves a multi-stage approach: (1) Analysis of feedstock for desired polymers; (2) Design of extraction strategy to obtain the polymers in the most native form; (3) Establishing proof of concept and process optimisation by lab-scale experiments; (4) Techno-economic analysis and life cycle assessment of the process; (5) Scale-up design and testing.



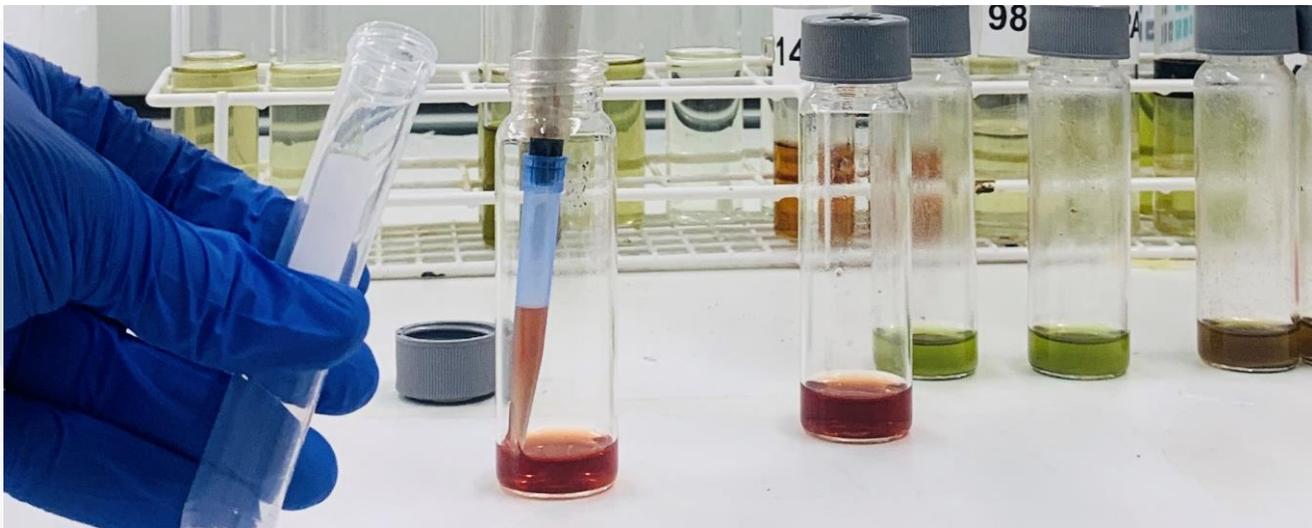
## 2 DESIGN AND TESTING OF BIOMATERIALS

We offer services for design of biomaterials such as: films, foams, hydrogels, and aerogels with specific functionalities.

Our expert team provides analytical testing for the biomaterials designed at Celignis or at clients’ locations. The testing services include thermal behaviour testing; physical, chemical and biochemical functionalities.



# 4. High-Value Biomolecules



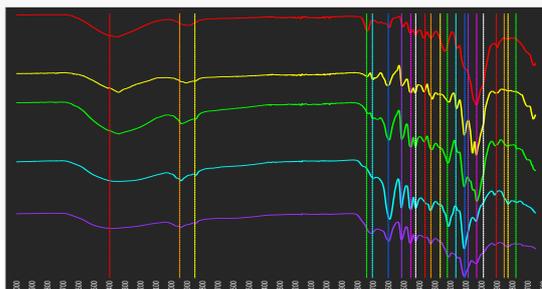
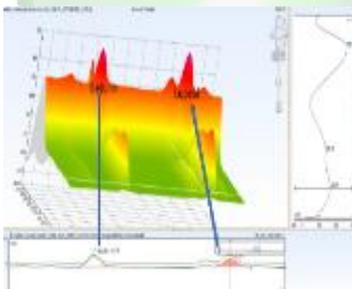
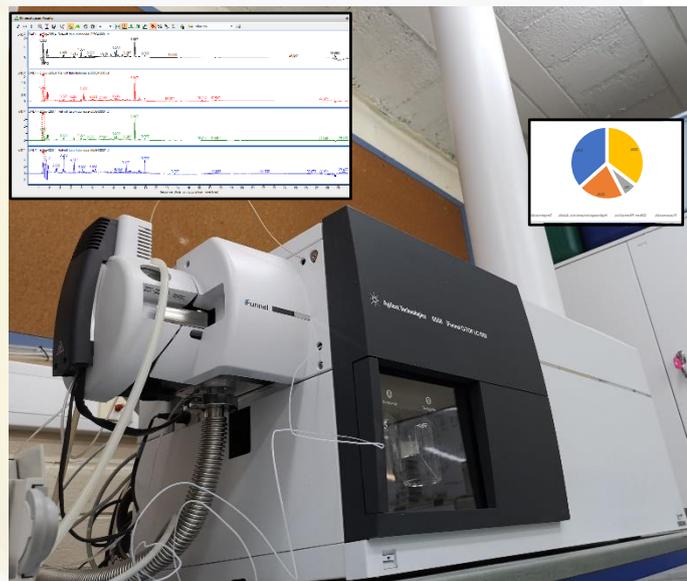
Biomolecules are derived from biomass and microbes. They have a wide range of applications such as cosmetics, paints and coatings, food and nutraceuticals and biomedicine.

## DISCOVERY, EXTRACTION AND PURIFICATION

Our multi-disciplinary team of biomass chemists, analytical chemists, and microbiologists work together to discover the novel molecules from biomass and microbial sources. Discovery of molecules from biomass involves multiple extractions in a range of polar and non-polar solvents. The extracted fractions are subjected to LC-QTOF-MS. The selected molecules, based on the novelty or abundance, are isolated and purified, following the principles of green chemistry, and are tested for purity.

## STRUCTURAL AND FUNCTIONAL CHARACTERISATION

The purified biomolecules are analysed for functional groups and linkages using spectroscopic techniques. Functional characterisation of the purified molecules is performed by performing a wide variety of custom-designed microbial, biochemical, and chemical tests.



## SERVICES TO INDUSTRY

# 5. Bioprocess Development



We develop and refine processes to efficiently valorise biomass. We can work on individual process stages, or develop a bespoke vertically-integrated technology for your feedstock or desired end-product. This can be done at lab-scale up to the 1m<sup>3</sup> level. Our understanding of biomass chemistry, our extensive array of bioprocessing equipment, and the biological, engineering, and commercial experience of our Bioprocess team, all play crucial roles in ensuring that our projects are well-designed and focused on our clients' end-goals.

1

## Biomass Extraction Processes

Biomass can be rich in bioactive compounds of high value for food, feed, cosmetic, and pharmaceutical applications. We develop bespoke extraction methods suitable for your needs with high selectivity, efficiency and low environmental impact.

3

## Biomass Hydrolysis

For the hydrolysis of biomass to monomeric sugars either chemical or biological approaches can be used. We can use both methods at scales ranging from flask-level to 1m<sup>3</sup>. We have particular expertise in enzymatic hydrolysis.

5

## Fermentations

We're experienced in many fermentations and can help you determine and optimise yields of an array of different fermentation products.

2

## Pretreatment Processes

The choice of pretreatment method varies with the type of biomass and the end-product requirements. At Celignis we can determine the most suitable pretreatment for your feedstock and determine the optimum conditions in lab-scale trials followed by higher TRL scale-ups.

4

## Application of Enzymes

Enzymes are biological catalysts that have a wide variety of applications in the bioeconomy. We are experts in the design and use of enzymatic approaches for the enhanced valorisation of lignocellulosic biomass.

6

## Downstream Processing

How the various outputs (solid and liquid) of a bioprocess are dealt with is often overlooked until later in bioprocess development, leading to excessive costs and complications.



Accurate data are not enough. We have in-depth understanding of the implications of composition and can design processes to fully valorise biomass

## 7 Lab-Scale Optimisations

We consider that optimising a bioprocess at the lab-scale is the most-cost effective approach to explore a range of different scenarios in search of optimal process conditions. Based on the outputs of these experiments we can then test the chosen set of conditions at higher TRL levels.

## 9 Techno-economic Analyses

Our techno-economic experts can evaluate your bioprocess, considering various scale, tech, and feedstock options. We apply accurate costing models to determine CAPEX/OPEX of simulated and pilot scale processes which are then used to determine key economic indicators (e.g. IRR, NPV).

## 8 Scale-Up to Higher TRLs

At our dedicated Celignis Bioprocess laboratories we have all the necessary upstream and downstream apparatus to undertake bioprocess projects up to a technology readiness level (TRL) of 6, with reactor and processing capacities of up to 1m<sup>3</sup>.

## 10 Full Project Development

We work closely with you to understand your objectives and timelines. We then propose a project, usually covering a series of deliverables and stage-gates. Often our projects involve optimising conditions at the lab-scale before replicating the conditions at higher TRL levels.

# Types of Bioprocess Projects

## 1 PRODUCT-CENTRED BIOPROCESS

In this scenario the final-product is locked-down and, instead, the bioprocess development can focus on the best approach to sustainably and profitably get it. This means there is some flexibility regarding the feedstock and processing technologies to be employed

## 2 FEEDSTOCK-FOCUSED BIOPROCESS

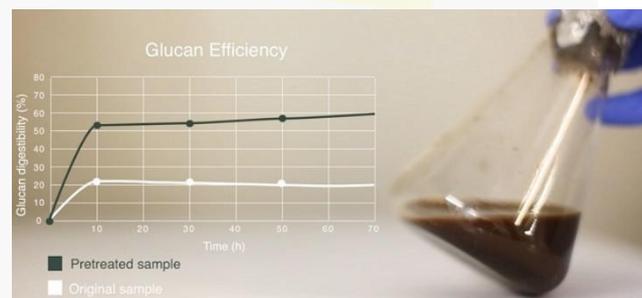
Here the feedstock is locked-down but the technological approaches and final end-product(s) are open. The Celignis team will consider the existing infrastructure of the client when designing the bioprocess as well as any existing chemical demands of their main process.

## 3 REFINEMENT OF AN EXISTING BIOPROCESS

This improvement can target the whole bioprocess or specific nodes. The targets for the improvement can be: product yield, OPEX reductions, and improved product quality, among others.

## 4 NEW FULL-VALUE-CHAIN BIOPROCESS

We can also work on Bioprocess Development for an entirely new bioprocess, covering all stages of the process-scheme. This means we start with the original feedstock and develop all stages involved in processing it and obtaining the targeted product(s). Developing such a comprehensive bioprocess requires considering, and developing approaches for, a number of key aspects, including: Feedstock Selection and Preparation; Primary-Conversion Technology; Downstream Processing Steps; Side-Stream Valorisation; Product-Recovery and Purification; Waste Management; Process Integrations.



## SERVICES TO INDUSTRY

# Bioprocess Project Case Studies



We are proud of the knowledge, passion, and work ethic of our team. They have played key roles in the formulation, optimisation, and commercial evaluation of biomass valorisation processes in industry and academia and, together, we have the multidisciplinary expertise to evaluate all stages of your bioprocess and suggest real improvements

## Glycerol from Industry Side-Streams

This project focused on the hydrolysis of cellulosic side-streams from an industrial process, followed by the fermentation of the liberated sugars into a variety of products, including glycerol, ethanol, and organic acids. Following the lab-scale work, we worked on a TEA analysis of the bioprocess, considering several different scenarios. The outputs of this TEA informed a follow-on bioprocess development project, incorporating the changes deemed to give greatest impact to the process in terms of commercial and environmental sustainability. The final stages of this project will involve scaling up the developed approach to the 100-litre level.

## Sugars from Paper Side-Streams

This project involved the optimisation of process conditions to allow for the production and recovery, in high yields, of monomeric sugars from recycled paper/cardboard streams. The project involved examining a number of important process variables, including: pretreatment conditions; the type and loading of enzymes; and the duration and conditions of the hydrolysis stage. The final outputs of the project were selected optimum conditions for each of the feedstock types and recommendations for further optimisations of the process and future scaled-up activities.

## Other Bioprocess Projects

- Production of Propionic Acid.
- Biomaterials from Caribbean Seaweed.
- Bioactives from Tropical Hardwoods.
- Sustainable Downstream Purification Process Developed.

## Bioethanol from Palm Residues

This was a lab-scale vertically-integrated project covering pretreatment, and separate hydrolysis and fermentation (SHF). The project involved a series of lab-scale experiments focused on optimising the pretreatment conditions so that the yields and commercial viability of the process as a whole could be improved. The next stage involved optimising the type and dosage of enzymes, as well as other factors (e.g. solid-loading), to maximise ethanol yields from the targeted biomass components.

## Oligomers from Biomass

We have undertaken a number of projects, for different clients, focused on obtaining oligomeric sugars from biomass, or biomass-derived polymers. In some cases there have been specific requirements in terms of the preferred oligomer chain length and the ratio of monomeric to oligomeric sugars in the final liquid output. Optimising the bioprocess required a carefully-formulated DoE considering relevant factors (e.g. temperature, enzymes, pretreatment) in the context of the chosen feedstock and the final product requirements. In all such projects that we have undertaken to date we developed an improved bioprocess that allowed for greater proportions of the total carbohydrates in the liquid phase being in the client's targeted product range.



Through Innovation, Passion, and Determination,  
We at Celignis Strive to Make a Difference in The  
Development of The Bioeconomy



## Relevant Experience & Infrastructure at Celignis Bioprocess

1

### Dedicated Bioprocess Building

Celignis Bioprocess was opened in 2022 and provides a dedicated 500m<sup>2</sup> facility for the at-scale (TRL7) processing of biomass. The site is also used for lower-TRL optimisation experiments in order to find the most suitable process conditions that can then be validated in larger reactors. The nearby-located Celignis Analytical building provides all of the necessary analytical support regarding the evaluation of the feedstocks, products, and side-streams.

3

### Track-Record in Research

In addition to our successful Bioprocess Development Services projects that we have undertaken for our corporate clients, Celignis has a long and impressive history as a valued participant in collaborative research projects. We are a spin-out of a research project designed and written by Celignis founder Dan Hayes and have, to date, participated in 20 research projects, funded by the European Union's Horizon research programme, focused on the development of innovative bioprocesses. 9 of these projects are currently ongoing.

2

### Experienced Bioprocess Team

For a commercially-viable bioprocess it is necessary to consider many aspects, both technological and financial, of the value-chain. Celignis Bioprocess is populated by a diverse team with multidisciplinary and complementary expertise. There are biologists, chemists, electricians, engineers, fermentation specialists, techno-economic analysts, and chemometricians. In 2021 Celignis (Ireland) was awarded "Innovation of the Year" (Irish Lab Awards) for one of its bioprocesses.

4

### Bioprocess Infrastructure

- Array of bench-top (1-100 litre) advanced bioreactors.
- Anaerobic fermentation systems (1-20 litres).
- Gas fermentation systems.
- Extracellular flux analysers for metabolic burden analysis.
- Library of industrially-relevant non-recombinant strains.
- 96-wellplate UV-Vis and fluorimetry analysers.
- QTOF-LC/MS for microbial metabolites analysis.
- Ultrafiltration systems for downstream processing.
- Extensive chromatography lab for fermentation products
- 1m<sup>3</sup> bioreactors for scale-up studies.

## BIOMASS FEEDSTOCK ANALYSIS

## 6. Anaerobic Digestion Services

We provide a wide range of analysis & consultation services to the anaerobic digestion sector. Our multidisciplinary expertise spans feedstock chemistry, biology, process optimisation, and techno-economics. It's the interaction of all these factors that will allow for feedstocks to be efficiently valorised and for the most impactful RNG projects to be developed.

A key motivation in establishing Celignis facilities in the US was the rapid national expansion of the anaerobic digestion (AD) sector for Renewable Natural Gas (RNG) production. Over the years Celignis (Ireland) had built a strong reputation, not only as a high-quality lab for the provision of analytical data to biogas stakeholders, but also as a centre of biological and technical expertise in the AD sector.

In previous years Celignis (Ireland) had several US clients from the biogas sector that expressed frustrations with the absence of any US-based lab with this kind of wide-ranging expertise. They wished to pursue more significant projects with Celignis but the volatile nature of many AD feedstocks meant that overseas shipments were not possible. We are delighted that the launch of Celignis USA now allows us to provide our valued services to RNG stakeholders across America.

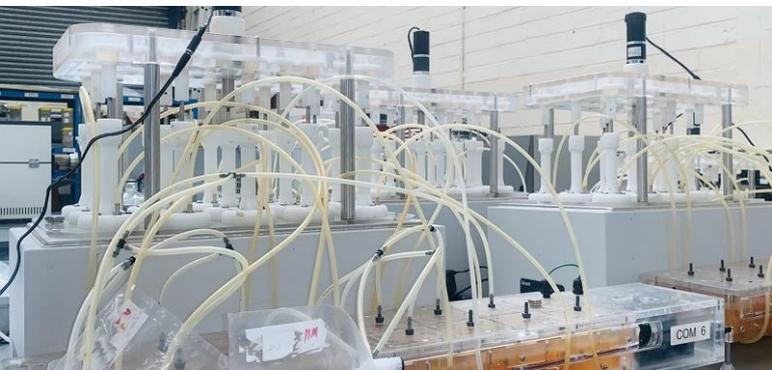


### BIOMETHANE POTENTIAL

The biomethane potential (BMP) can be considered to be the experimental theoretical maximum amount of methane produced from a feedstock. In our laboratory, we have twenty BMP systems, comprising 300 reactors, that allow us to digest your samples and determine the biogas yield over periods of between 14 and 40 days.

### CONTINUOUS DIGESTIONS

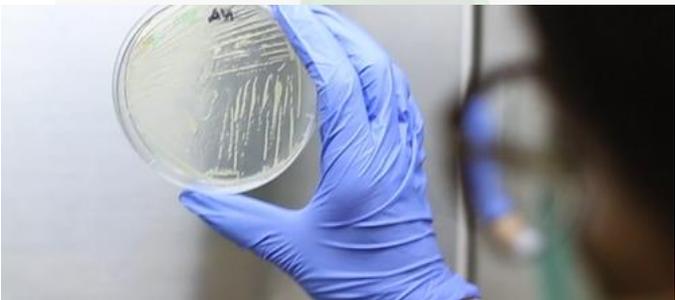
To help you evaluate how well your anaerobic digestion feedstocks will behave in real-world conditions we can undertake continuous digestion experiments. These operate at scales up to 12 litres and typically run for 3 months. We target maximum achievable organic loading rate (OLR) and biomethane potential.



We provide analysis and consultation services to help evaluate feedstock and process suitability for maximising biomethane yields and RINs credits

### TOXICITY ASSAYS

The waste streams used in RNG that arise from process industries may contain toxic or bacterial inhibitory compounds (e.g. antibiotics, polyelectrolytes, detergents). Our anaerobic toxicity assays can determine the presence of such toxicities and suggest feeding limits for feedstocks.



### SPECIFIC MICROBIAL ACTIVITY

AD is a microbial process involving a sequence of stages (hydrolysis, acidogenesis, methanogenesis) to convert a complex feedstock to methane. We analyse samples collected from digesters and undertake tests to investigate how well they proceed with each of these stages of digestion. Tests undertaken include: Specific Hydrolytic Potential (SHP), Specific Acidogenic Potential (SAP), Specific Methanogenic Potential (SMP). Each test involves 5 days of substrate digestion and monitoring of the biogas produced.

### DIGESTATE ANALYSIS

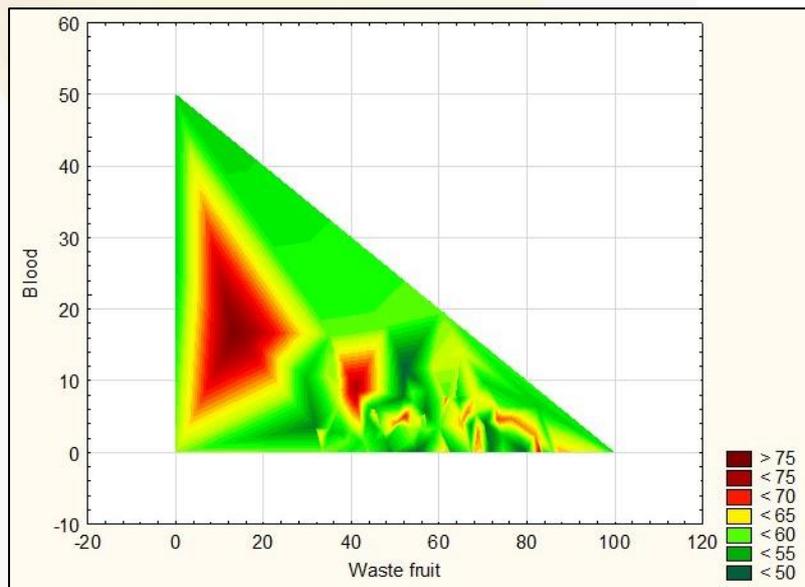
Digestate can potentially have value as a soil fertiliser. We offer a range of detailed analysis packages for digestate, allowing you to fully assess this resource and to determine the best use for it. Our team can also assist in evaluating digestate valorisation options.

### PROCESS OPTIMISATION

There are many factors in running an RNG facility. Optimising these allows for improved plant performance and revenues. We can design and experimentally-validate such optimisations at the lab-scale prior to you implementing them at your facility. This approach allows for greater benefits and lower costs than optimising at the commercial scale. For example, we can suggest optimal values for major and minor elements in the digester as well as upper and lower threshold values. This allows us to formulate a bespoke cocktail of additives according to the requirements of the digester.

### BIOLOGICAL CONSULTANCY

We are experts in the biology of anaerobic digestion. We pour through operational data from biogas plants and identify correlations between process parameters and digester performance. This leads to understanding on the specific biological conditions of the digester and recommendations as to how performance can be improved and made more stable.



SERVICES TO INDUSTRY

# RNG Project Case Studies



At Celignis, we understand the critical role that accurate and timely data plays in the AD sector. With a decade of expertise in biomass analysis and valorisation, and over 1000 clients, our comprehensive suite of analytical and consultation services will enhance your AD feasibility studies and optimise your AD plant operations.

## Toxicity Assays

A biogas plant started underperforming when a new feedstock was used as co-feed. Since the plant received a gate-fee for this feedstock they did not want to stop using it but instead to use it in a controlled manner. Celignis analysed the feedstock and then custom-designed and ran Anaerobic Toxicity Assays for the waste stream based on the resulting analytical data. This work allowed us to determine threshold feedstock-loadings, in order to avoid toxic/inhibitory effects.

## Biological Advice

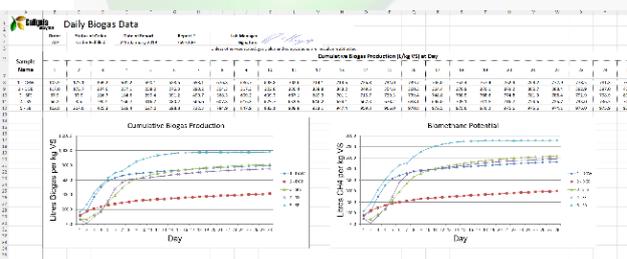
We helped a large AD company to optimise plant operations for more consistent outputs and reduced downtime. Our support involved us analysing process data and providing operational limits and green, yellow, and red zones for each process indicator. We also made a tool to allow self-design of major and minor elements (nutrients) for their plants based on feedstock chemistry. The tool was suitable for mono & co-digestion and allowed for shifting feedstocks and adding a new feedstock to the mix, without lowering plant performance.

## Continuous Digestions

We undertook continuous digestions, for a company producing biogas from the organic fraction of municipal solid waste, to determine the maximum achievable organic loading rate (OLR) and optimum feedstock mixtures. We also determined the minimum OLR to maintain plant health in scenarios of limited feedstock availability. These data, combined with the specific microbial activity tests on the digestate, provided the plant with adaptation strategies for the new feedstock. The full suite of tests and data analysis allowed the operator to understand feedstock limitations, feedstock underload/overload effects, optimum feedstock loadings, and process indicator ranges at different OLRs and feedstock mixtures. This allowed for bespoke adaptive strategies for maintaining plant health under feedstock supply and composition variations.

## Our Global Client Base

The Celignis brand has provided valued analytical and bioprocess services to over 1000 clients globally, including many in the RNG sector. We understand how the focus of AD projects can differ between countries and have advised a global network of clients on their RNG projects.



# Celignis RNG Packages

P#	Package Description	Price per Sample (\$)	Comment
P38	Major and Minor Elements	150	On feedstock or digestate, bulk savings possible
P81	Biomethane Potential - 14 Days - Basic	530	\$1,728 per batch of 4 samples (\$432 per sample)
P82	Biomethane Potential - 14 Days – Deluxe	835	\$2,908 per batch of 4 samples (\$727 per sample)
P84	Biomethane Potential - 21 Days - Basic	660	\$2,112 per batch of 4 samples (\$528 per sample)
P85	Biomethane Potential - 21 Days – Deluxe	955	\$3,292 per batch of 4 samples (\$823 per sample)
P87	Biomethane Potential - 28 Days - Basic	755	\$2,416 per batch of 4 samples (\$604 per sample)
P88	Biomethane Potential - 28 Days – Deluxe	1,050	\$3,596 per batch of 4 samples (\$899 per sample)
P90	Biomethane Potential - 40 Days - Basic	1,080	\$3,456 per batch of 4 samples (\$864 per sample)
P91	Biomethane Potential - 40 Days – Deluxe	1,315	\$4,636 per batch of 4 samples (\$1159 per sample)
P93	Feedstock Chemical & Biological Analysis	155	Included in the Deluxe BMP packages
P94	Digestate Chemical & Biological Analysis	155	Included in the Deluxe BMP packages
P95	Residual Biogas Pot. – 14 Days - Basic	390	Run on digestate, longer digestions possible
P220	Chemical Oxygen Demand (COD)	36	Falls to \$24 per sample, depending on order size
P221	Biological Oxygen Demand (BOD)	36	Falls to \$24 per sample, depending on order size
P222	Volatile Fatty Acids (VFA) Speciation	90	Falls to \$42 per sample, depending on order size
P223	Carbon Dioxide Evolution Rate	70	
P224	Specific Oxygen Uptake Rate	70	
P225	Digestate Impurity Content	110	Falls to \$70 per sample, depending on order size
P226	Ammoniacal Nitrogen	35	Falls to \$25 per sample, depending on order size
P227	Nitrates	30	Falls to \$20 per sample, depending on order size
P228	Viable Weed Seeds	120	Undertaken on digestate
P240	Specific Hydrolytic Potential (SHP)	495	<a href="http://www.celignis.com/specific-microbial-activity">www.celignis.com/specific-microbial-activity</a>
P241	Specific Acidogenic Potential (SAP)	495	<a href="http://www.celignis.com/specific-microbial-activity">www.celignis.com/specific-microbial-activity</a>
P242	Specific Methanogenic Potential (SMP)	495	<a href="http://www.celignis.com/specific-microbial-activity">www.celignis.com/specific-microbial-activity</a>
P243	SHP; SAP; and SMP	990	<a href="http://www.celignis.com/specific-microbial-activity">www.celignis.com/specific-microbial-activity</a>
P245	Sludge Granule Size Analysis	90	
P246	Sludge Activity Test	90	
P247	Anaerobic Toxicity Assay	895	<a href="http://www.celignis.com/anaerobic-toxicity-assay">www.celignis.com/anaerobic-toxicity-assay</a>
P249	Biological Consultation	750 a day	<a href="http://www.celignis.com/ad-biological-consultations">www.celignis.com/ad-biological-consultations</a>
P250	Continuous Anaerobic Digestion	1,650*	<a href="http://www.celignis.com/ad-continuous-digestions">www.celignis.com/ad-continuous-digestions</a>
P386	Digestate Germin Inhibition/Stimulation	495	Falls to \$355 per sample, depending on order size
P388	Digestate Plant Growth Trials	545**	Falls to \$325 per sample, depending on order size

\* Per reactor/month. Price dependent on reactor size (5-100 litres) and on the analyses measured in the period.

\*\* Larger (tray) trials using a dedicated greenhouse can also be arranged.

## Biogas and Biomethane Potential (BMP)

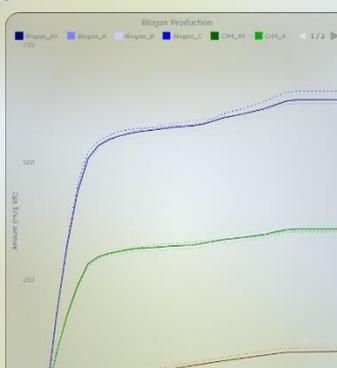
After 28 Days of Digestion at 37°C

### Biogas and Biomethane Yields

Biogas Produced (litres)	BMP (litres/kg VS)	Biogas Produced (litres/kg VS)	BMP (litres/kg VS)	Biogas Produced (litres/kg VS)	BMP (litres/kg VS)
Av. 633.66	358.65	364.48	206.32	285.68	161.71
Rep 1 625.46	353.25	359.92	203.21	282.82	159.27
Rep 2 623.12	352.20	358.45	202.84	280.95	158.03
Rep 3 652.20	378.43	375.10	211.99	284.99	157.62
SD 16.15	16.21	8.20	5.88	7.29	6.61

### Additional Biogas Statistics

Stat	Day
Max. biogas production reached at day	28
70% methane biogas reached at day	4
80% methane biogas reached at day	4
90% methane biogas reached at day	11
95% methane biogas reached at day	11
98% methane biogas reached at day	9



Page 1 of 10

**CERTIFICATE OF ANALYSIS**

Customer # 201      Order # 705      Report # 705-COPI

Order Status      Order Fulfilled      Date of Report 21th January 2019

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Biogas and Biomethane Potential (BMP) - Summary Data

Sample Name	Total Biogas Produced (litres/kg VS)	Biomethane Potential (litres/kg VS)	Final Weighted Biogas Composition (%)
1 - DMW**	16.60	19.62	28
2 - DMW**	11.69	18.51	28
3 - SPB**	6.43	13.65	28
4 - SPB**	8.88	11.90	28
5 - SPB**	13.07	10.90	28

\* Data can also be viewed online at [www.celignis.com/outputs/biogas\\_potential](http://www.celignis.com/outputs/biogas_potential)  
 \*\* Gas yields are biomass-normalised, unless otherwise stated.  
 \*\*\* Gas composition not normalised (abstracted for specific periods) due to low net gas production. See 'Biogas Composition' during Market table for more information.

Lab Manager Signature:

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**ALL WORK IS UNDERTAKEN SUBJECT TO OUR TERMS AND CONDITIONS**

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FEEDSTOCK TO BIOCHAR

# 7. Advancing Biochar

**RELEVANT ANALYSIS PACKAGES:**

- P350 – Biochar Production**
- P360 – Specific Surface Area**
- P364 – Pore Size Distribution**
- P388 – Biochar Plant Growth Trials**
- P386 – Germination Inhibition**
- P384 – Biochar Polycyclic Aromatic Hydrocarbons (PAH)**
- P382 – Water Holding Capacity**
- P383 – Cation Exchange Capacity**
- P33 – Ultimate Elemental Analysis (C, H, N, S, O)**
- P372 – Inorganic Carbon**
- P38 – Major and Minor Elements:**  
Al, Ca, Fe, Mg, P, K, Si, Na, Ti, Sb, As, Cd, Cr, Co, Cu, Pb, Hg, Zn, Va, Ni, Mn
- P373 – Thermogravimetric Analysis**
- P34 – Calorific Value and Elements:**  
Gross Calorific Value, Net Calorific Value, Ash, CHNSO
- P42 – Ash Melting Behaviour**
- P371 – Ash Content (815 °C)**
- P381 – Electrical Conductivity**
- P387 – Scanning Electron Microscopy (SEM) Imaging**

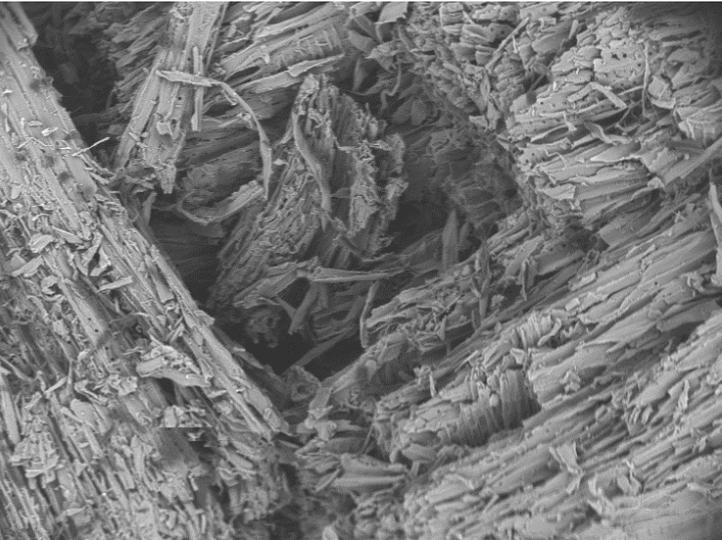


We can determine the most relevant properties of biochar and provide a comprehensive assessment of the results

There is a huge buzz, all across the US, about the role that biochar can play in the development of a more sustainable economy. Potential applications include soil-amendment, carbon sequestration, waste reduction, biomaterials, and energy production. However, many factors need to be considered in order for the biochar approach to be successful. These include the characteristics of the feedstock, the conditions of the pyrolysis process, and the properties of the resulting biochar.

The Celigis USA team has extensive experience in the analysis of biochar and in the evaluation and testing of its potential applications. For clients that are considering pyrolysis as a means to valorise their feedstocks or residues, we are able to screen their samples for suitability and produce lab-scale quantities of biochar for further analysis and application testing. For producers of biochar, we have over 30 different packages to evaluate their samples and test the suitability for various applications (e.g. plant-growth trials).





We are equipped to run all the necessary analyses for the EBC and IBI methods

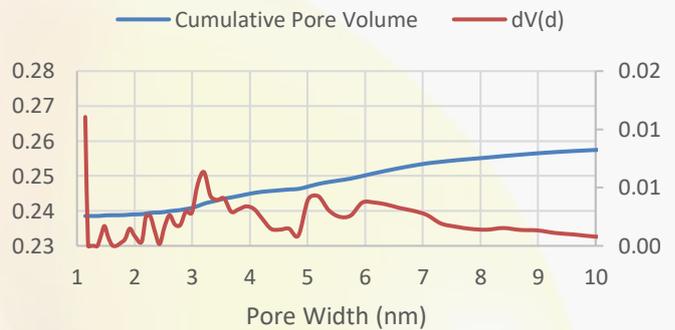
### SURFACE AREA & PORE ANALYSIS

During pyrolysis, the release of volatile components present in biomass provides biochar with a characteristic honeycomb structure with a relatively high surface area. This porous nature makes biochar suitable for applications such as: an adsorbent to decontaminate air and water; a soil supplement for improving plant development; or as an additive for upgrading biogas production. The production of biochar is carried out by optimizing parameters such as residence time, temperature, heating rate, inert gas flow rate, and particle size according to each type of feedstock. This variety of operative parameters results in porous, but also non-porous biochar. Therefore, it is essential to fully characterize the porous profile of each type of biochar to have a clear picture of its potential applications. Celignis USA offers a wide range of packages for such analyses.

### THRESHOLD

Biochar can have constituents that may prohibit its use in certain applications. We determine the concentrations of 23 different polycyclic aromatic hydrocarbons (PAHs) and several heavy metals. Our reports provide PASS/FAIL tables where the results are compared against threshold values set by the EBC for various end-uses.

### Pore Size Distribution



### Feedstock Analysis - Summary Data

Test	Method Reference	Units	As-Received	Dry Mass Basis	Dry Ash-Free Basis
Moisture	EN 14774-1:2009	%	8.49	-	-
Total Solids	Calculated	%	91.51	-	-
Ash	EN 14775:2009	%	6.85	7.49	-
Volatile Solids	Calculated	%	84.66	92.51	-
Carbon	EN 15104:2011	%	42.25	46.17	49.91
Hydrogen	EN 15104:2011	%	5.23	5.72	6.18
Nitrogen	EN 15104:2011	%	1.83	2.00	2.16
Sulphur	EN 15289:2011	%	0.14	0.15	0.16
Oxygen	By Difference	%	35.21	38.47	41.59
Aluminium	EN ISO 16967:2015	ppm	101	110	-
Calcium	EN ISO 16967:2015	ppm	4,669	5,102	-
Iron	EN ISO 16967:2015	ppm	120	131	-
Magnesium	EN ISO 16967:2015	ppm	3,313	3,620	-
Sodium	EN ISO 16967:2015	ppm	51	56	-
Phosphorus	EN ISO 16967:2015	ppm	2,994	3,272	-
Potassium	EN ISO 16967:2015	ppm	11,853	12,953	-

### SOIL APPLICATIONS

Biochar can be relevant in carbon accounting schemes, as it can be considered to sequester carbon whilst also potentially enhancing soil fertility and plant productivity. However, biochar can sometimes inhibit the development of plants. For example, when it is produced from hazardous feedstocks such as municipal solid waste, the presence of heavy metals and other detrimental minor elements can be problematic. Therefore, the starting material and the obtained biochar should always be analysed prior to considering using biochar for soil amendment. We have many suitable analysis packages in this regard and can also undertake plant growth trials in our laboratories, using biochar-amended soil compared against controls.

## FEEDSTOCK TO BIOCHAR

# Celignis USA Biochar Packages

Pkg #	Package Description	TAT		Per Sample Price (\$)					# Samples in Order			
		(wks)	P1	P2	P3	P4	P5	L2	L3	L4	L5	
<b>Biochar Production</b>												
P350	Biochar Production	2	655	545	435	325		5	10	30		
<b>Biochar Chemistry</b>												
P10	Ligno. Constit., Extractives, and Ash	2	550	495	445			5	10			
P226	Ammoniacal Nitrogen	2	35	30	25			30	50			
P227	Nitrates	2	30	25	20			10	30			
<b>Biochar Physical Properties</b>												
P51	Particle Size	1	42	30				10				
P53	Bulk Density	1	25	20	15			10	30			
P360	Specific Surface Area	2	305	275	220			10	30			
P364	Pore Size Distribution	2	360	325	250			10	30			
P366	Pore-Size Distribution Deluxe	2	435	385	330			10	30			
P368	Pore-Size Distribution Ultimate	2	655	600	545			10	30			
<b>Biochar Thermal Properties</b>												
P2	Moisture Content	1	30	25	20			10	30			
P3	Ash Content	1	30	25	20			20	50			
P31	Volatile Matter	2	60	50	40			5	10			
P33	Ultimate (Elemental) Analysis	2	54	42	36			20	40			
P34	Calorific Value and Elements	2	150	115				10				
P35	Chlorine and Sulphur (Requires P34)	1	31	25	19			10	30			
P42	Ash Melting Behaviour (Reducing)	2	120	100	75			10	30			
P370	Inherent Moisture	1	28	17	11			10	30			
P371	Ash Content (815C)	1	42	30	18			20	40			
P372	Inorganic Carbon	2	54	42	36			25	50			
P373	Thermogravimetric Analysis (TGA) (N <sub>2</sub> )	2	215	165	140	130	110	10	30	40	50	
P374	Thermogravimetric Analysis (TGA) (Air)	2	215	165	140	130	110	10	30	40	50	
<b>Biochar Soil Amendment Packages</b>												
P38	Major and Minor Elements	2	150	125	100			20	40			
P381	Electrical Conductivity	2	55	45	35			10	20			
P382	Water Holding Capacity	2	105									
P383	Cation Exchange Capacity	2	165									
P384	Polycyclic Aromatic Hydrocar. (PAH)	2	325	275	215	165		10	20	30		
P385	Liming	2	85	65	55			10	20			
P386	Germination Inhibition	3	495	435	385	355		5	10	20		
P387	Scanning Electron Microscopy (SEM)	2	545	435	385	325	275	2	5	10	15	
P388	Biochar Plant Growth Trials	5	545	435	385	325		2	5	10		

Pkg #	Package Description	TAT		Per Sample Price (\$)					# Samples in Order			
		(wks)	P1	P2	P3	P4	P5	L2	L3	L4	L5	
<b>Biochar Bulk-Analysis Packages</b>												
P390	Biochar Physical Properties Deluxe	2	495	435	380			10	20			
P391	Biochar Physical Properties Ultimate	2	715	655	575			10	20			
P393	Biochar Thermal Properties Deluxe	2	395	325				10				
P394	Biochar Thermal Properties Ultimate	3	875	775				10				
P396	Biochar Soil Amendment Deluxe	4	985	885	815	740		2	10	20		
P397	Biochar Soil Amendment Ultimate	7	1,925	1,645	1,425	1,265		2	5	10		
P399	Biochar Complete Evaluation Package	8	2,965	2,635	2,415	2,195	2,085	2	5	10	20	
<b>European Biochar Certificate (EBC) - Equivalent Tests</b>												
P2	Dry Matter	1	30	25	20			10	30			
P370	Water Content (40 C)	1	30	25	20			10	30			
P33, P372	Total Carbon, H:C <sub>org</sub> , C <sub>inorg</sub> , Nitrogen, Sulphur, Oxygen, Ash	2	108	84	72			25	50			
P53	Bulk Density	1	25	20	15			10	30			
P382	Water Holding Capacity	2	105									
P381	Electrical Conductivity	2	55	45	35			10	20			
P38	Major and Minor Elements	2	150	125	100			20	40			
Free	pH	1										
P384	Polycyclic Aromatic Hydrocar. (PAHs)	2	325	275	215	165		10	20	30		
P364	Surface Area & Pore Size Distrib.	2	360	325	250			10	30			
P51	Particle Size Distribution	1	42	30				10				
	<b>Combined Package:</b>	3	1,050	935	795	745		10	20	30		
	Dioxins/Furans & PCBs *Subcontracted	3	540									
	<b>Combined (Incl. PCB, PCDD/F)</b>	3	1,590	1,475	1,335	1,285		10	20	30		
<b>IBI Test Category A</b>												
P2	Moisture	1	30	25	20			10	30			
P33	Organic Carbon (Corg) (Incl. P372)	2	108	84	72			25	50			
P33	H:Corg (Incl. P372)	2	108	84	72			25	50			
P33	Total Nitrogen	2	54	42	36			25	50			
Free	pH	1										
P381	Electrical Conductivity	2	55	45	35			10	20			
P385	Liming	2	85	65	55			10	20			
P51	Particle Size Distribution	2	42	30				10				
	<b>Combined Package</b>	2	250	215	195			20	40			
<b>IBI Test Category B</b>												
P38	Major & Minor Elements	2	150	125	100			20	40			
P35	Chlorine	2	120	95	85	75		5	10	20		
P384	Polycyclic Aromatic Hydrocar. (PAHs)	2	325	275	215	165		10	20	30		
P386	Germination Inhibition Assay	3	495	435	385	355		5	10	20		
	Dioxins/Furans & PCBs (*Subcontracted)	3	540									
	<b>Combined Package</b>	3	1,465	1,355	1,285	1,195		10	20	30		
<b>IBI Test Category C</b>												
	Total Phosphorus and Potassium	2	98	85	75			20	40			
	Available Phosphorus	2	98	85	75			20	40			
	Total Ca, Mg & Sulphur (Incl. P33)	2	152	127	111			20	40			
	Avail.e Ca, Mg and Sulphate-S (+P33)	2	152	127	111			20	40			
P31	Volatile Matter	2	60	50	40			5	10			
P364	Total and External Surface Area	2	360	325	250			10	30			
	<b>Combined Package</b>	3	795	695	595			10	20			

# 8. Analysis of Process Liquids

## RELEVANT ANALYSIS PACKAGES

### P12 – Sugars in Extract:

Glucose, Xylose, Fructose, Sucrose, Mannose, Arabinose, Galactose, Rhamnose, Xylitol, Sorbitol, Arabinitol, Mannitol, Raffinose, Trehalose

### P13 – Sugars and Oligosaccharides:

As P12 plus amounts of each sugar in oligomeric form.

### P15 – Uronic Acids:

Glucuronic, Galacturonic, Mannuronic, Guluronic, 4-O-Methyl-D-Glucuronic

### P22 – Organic Acids and Furans:

Levulinic Acid, Formic Acid, HMF, Furfural, Acetic Acid

### P23 - Dimers and Trimers from Cellulose:

Cellobiose, Cellotriose

### P24 - Dimers and Trimers from Hemicellulose:

Xylobiose, Xylotriose, Arabinobiose, Arabinotriose, Mannobiose, Mannotriose

### P26 – Xylo-Oligos:

XOS from DP2 to DP6 plus Arabinofuranosylxylobiose, Arabinofuranosylxylotriose, Arabinofuranoxylotetraose.

### P29 - Oligos from Starch:

From DP2 to DP8

### P61 – Sugars in Bio-Oil Water Extract:

As P12 plus Levoglucosan, Cellobiosan, Mannosan, Galactosan

### P62 – Sugars and Oligosaccharides in Bio-oil Water Extract

Biomass conversion processes can produce complex liquids containing an array of products. We have the methods, equipment and expertise to allow you to find the real chemical value in your process liquids.

## MONOSACCHARIDES

Celignis USA quantifies dozens of different monosaccharides derived from biomass.

## SUGAR ALCOHOLS

Analytes we can determine include mannitol, sorbitol, arabinitol, glycerol, and xylitol.

## URONIC ACIDS

We can quantify Galacturonic, glucuronic, guluronic, mannuronic, iduronic, and 4-O-Methyl-D-Glucuronic acids in biomass/liquids.



## SUGAR DEGRADATION

Analyses for various sugar degradation products, such as organic acids (e.g. formic acid, acetic acid, levulinic acid) and furans (e.g. furfural and HMF).



## OLIGOSACCHARIDES

In our labs we can determine disaccharides and oligosaccharides in two different ways:

- Directly – for example, we can determine xylo-oligos up to DP6.
- Indirectly via acid hydrolysis of the liquid to break apart the oligosaccharides and determine their constituent monomers.

## ANHYDRO-SUGARS

Including levoglucosan, mannosan, galactosan, and cellobiosan.

## EXTRACTIVES

We determine fifteen different carbohydrates in water-extracts and fatty acids in organic solvents. Our QTOF-LC/MS system allows us to identify unknown extractive compounds.

## PYROLYSIS BIO-OILS

The bio-oil fraction obtained from biomass can also be highly complex but we have packages to determine the important carbohydrates (including anhydrosugars such as levoglucosan, galactosan, mannosan, and cellobiosan) in the water phase of the oil as well as the oligomeric sugars.

# 9. Evaluation of Pre-treatments

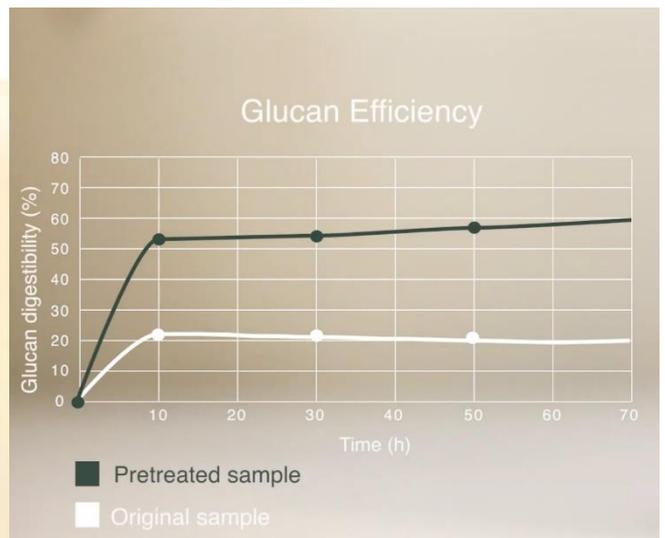
## 1 THE STARTING FEEDSTOCK

We offer several packages for feedstock composition. P10 gives detailed data on extractives content, lignin, cellulose, and hemicellulose. P12, P14, P15, and P16 are complementary, providing data on water soluble carbohydrates, starch, uronic acids, and acetyl. All the above are combined in P19 (Deluxe Lignocellulose Package) which also gives a more accurate lignin determination by correcting for protein (P270). The resulting data will help in selecting the right pre-treatment for the desired product/application. The Celignis team can assess the compositional data and design the pre-treatment process for the selected feedstock.



## 2 LIQUID PRODUCT OF PRE-TREATMENT

Traditionally, liquids from pre-treatments are considered as low value or wastes. However, with advancements in green chemistry and biotechnology, pre-treatment streams are being researched to produce high-value products. For this detailed compositional analysis is needed. Celignis offers analysis packages for sugars, sugar alcohols and oligosaccharides in solution (P13), organic acids and furans (P22) and uronic acids (P15). Additionally, the fermented or chemical conversion products of these streams can be analysed based on custom requirements.



## 3 SOLIDS FROM PRE-TREATMENT

The solids separated from the pretreated slurry contain the biomass fraction that was not deconstructed by the treatment and adsorbed sugars, phenolics, etc. Depending on pre-treatment type the adsorbed fraction can be significant and so needs to be removed for the analysis of structural components. Pretreatment efficiency is calculated by determining the enrichment of required fraction in the solids (e.g. cellulose) and by determining the improved accessibility to enzymes using analysis packages custom-designed by the Celignis team.



BIOMASS FEEDSTOCK ANALYSIS

# 10. Seaweed Analysis



Our team has characterised thousands of seaweed samples as well as the products and side-streams of seaweed valorisation processes.

There has recently been a surge of interest in seaweed as a sustainable feedstock for food, feed, fuels, chemicals and biomaterials. However, since seaweed composition differs greatly from terrestrial biomass, analytical methods should be customised according to its constituents and the complex matrices of the samples. The Celignis team has worked extensively on this since 2020, with several in-house methods developed plus collaborations in international round-robin testing campaigns.

We currently offer over 20 analysis packages focused on seaweed. These cover a diverse array of constituents, including carbohydrates, amino acids, pigments, phytohormones, vitamins, phenolics, tannins, flavonoids, bromoform, phlorotannins, and detailed characterisations of seaweed polysaccharides, including molecular weight analysis.

Our team has also worked with clients on bioprocess projects for seaweed valorisation in different applications, including biomaterials.



## Our Analysis Packages for Seaweed

P71

Seaweed Carbohydrates

Fucose, Mannitol, Glucose, Xylose, Mannose, Arabinose, Galactose, Rhamnose, Total Sugars, Glucuronic Acid, Galacturonic Acid, Mannuronic Acid, Guluronic Acid, Iduronic Acid.

P72

Seaweed Amino Acids

Alanine, Arginine, Aspartic Acid, Cystine, Glutamic, Glycine, Histidine, Isoleucine, Leucine, Lysine, Methionine, Phenylalanine, Proline, Serine, Threonine, Tyrosine, Valine.

P33

Ultimate (Elemental) Analysis

Carbon, Hydrogen, Nitrogen, Sulphur, Oxygen, Ash.

P38

Major and Minor Elements

Aluminium, Calcium, Iron, Magnesium, Phosphorus, Potassium, Silicon, Sodium, Titanium, Antimony, Arsenic, Cadmium, Chromium, Cobalt, Copper, Lead, Manganese, Mercury, Molybdenum, Nickel, Vanadium, Zinc.

P73

Seaweed Lipids as Fatty Acids

Arachidic Acid, Behenic Acid, Decanoic Acid, Erucic Acid, Lauric Acid, Linoleic Acid, Linolenic Acid, Myristic Acid, Caprylic Acid, Oleic Acid, Palmitic Acid, Palmitoleic Acid, Stearic Acid.

P74

Pigments in Seaweed

Fucoxanthin, Astaxanthin, Chlorophyll-c, Chlorophyll-a, Chlorophyll-b, Lutein, beta-Carotene, Neoxanthin, Antheraxanthin, Violaxanthin.

P75

### Seaweed Phytohormones

Gibberellic Acid, Indole-3-Acetic Acid, Indole-3-Propionic Acid, Indole-3-Butyric Acid, 6-Benzylamino-Purine, Kinetin-Riboside, Abscisic Acid, Salicylic Acid, Zeatin.

P76

### Vitamins (Fat-Soluble) in Seaweed

Phylloquinone, Tocopherol, beta-Carotene.

P171

### Molecular Weight Analysis - Alginate

Using size-exclusion chromatography and refractive index detection.

P174

### Seaweed Extract Polysaccharides

A detailed package allowing for the determination of the content and composition of polymers (fucoïdan, laminarin etc.) in various algal species.

P77

### Vitamins (Water-Soluble) in Seaweed

Thiamine, Niacin, Nicotinamide, Pyridoxine, Folic Acid, Riboflavin, Pantothenic Acid, Ascorbic Acid, Biotin.

P79

### Seaweed Phenolics Profiling

Acids (Gallic, Caffeic, Chlorogenic, Ferulic, Coumaric, Protocatechuic), Catechin.

P172

### Total Phlorotannins Estimation

Total Phlorotannins.

P176

### Seaweed Flavonoids

Total Flavonoid Estimation.

P155

### Seaweed Polyamines

Dopamine, Histamine, Serotonin, Phenylethylamine, Putrescine, Cadaverine, Norspermidine, Spermidine, Spermine, Tyramine, Agmatine

P170

### Seaweed Total Tannins

Total Tannins.

P173

### Seaweed Bromoform Content

Bromoform (for suitable species).

P177

### Seaweed Dietary Fiber

Dietary Fiber.

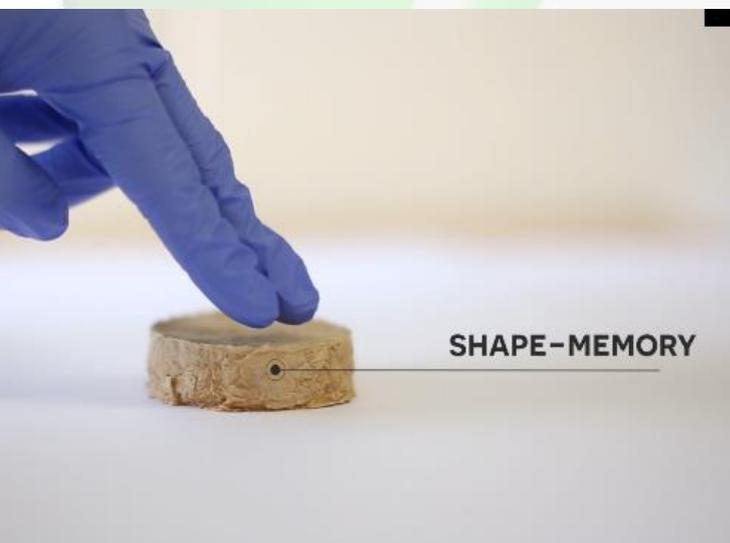
## CUSTOM METHOD DEVELOPMENT

Sajna KV (PhD), our Bioanalysis Developer, has developed new and custom analysis methods, according to our clients' needs, for seaweeds and other biomass. Sajna also took part in the NIST Quality Assurance Program (Seaweed).



## SEAWEED BIOPROCESS PROJECT

**Isolation of Seaweed Polymers for Production of Sustainable Biomaterials:** This project concerned the development of a new sustainable process for the extraction, and subsequent modification, of alginate from seaweed. The process allowed for alginate extraction without the use of harsh chemicals and also considered the valorisation of process side-streams (e.g. fucoïdan, cellulose etc.). We found that tweaks in the extraction and modification stages could influence the physicochemical properties of the resulting alginate, allowing for different types of materials for different end-uses.



## BIOMASS FEEDSTOCK ANALYSIS

# 11. Biomass Combustion

**RELEVANT ANALYSIS PACKAGES:**
**P31 – Volatile Matter**
**P32 – Proximate Analysis:**

Moisture, Ash, Volatile Matter, Fixed Carbon

**P33 – Ultimate Analysis:**

Carbon, Hydrogen, Nitrogen, Sulphur, Oxygen, Ash

**P34 – Calorific Value and Elements:**

Gross Calorific Value, Net Calorific Value, Ash, Carbon, Hydrogen, Nitrogen, Sulphur, Oxygen

**P35 – Chlorine and Sulphur**
**P36 – Major Elements:**

Aluminium, Calcium, Iron, Magnesium, Phosphorus, Potassium, Silicon, Sodium, Titanium

**P37 – Minor Elements:**

Antimony, Arsenic, Cadmium, Chromium, Cobalt, Copper, Lead, Manganese, Mercury, Molybdenum, Nickel, Vanadium, Zinc

**P38 – Major and Minor Elements**
**P40 – Combustion Package:**

Gross Calorific Value, Net Calorific Value, Chlorine, Moisture, Ash, Carbon, Hydrogen, Nitrogen, Sulphur, Oxygen, Volatile Matter, Fixed Carbon

**P41 – Ash Melting Behaviour (Oxidising Conditions):**

Shrinkage Starting Temp., Deformation Temp., Hemisphere Temp., Flow Temp.

**P42 – Ash Melting Behaviour (Reducing Conditions):**

As P41 but under reducing conditions

**P373 – Thermogravimetric Analysis**

**P50 – Ultimate Combustion Package:**  
P40 plus P38 and P42

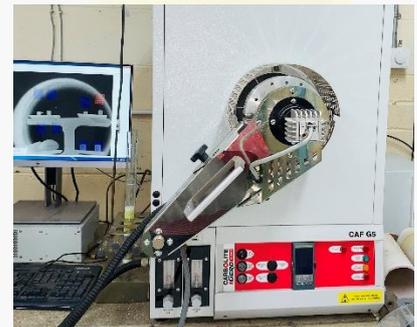
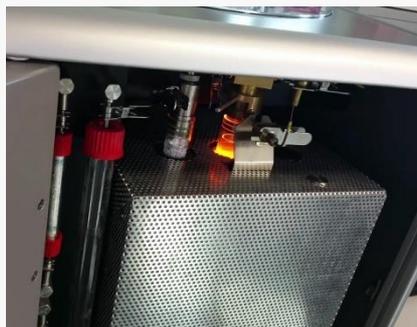


We have a range of packages to help you determine the value of your feedstocks for the production of heat and electricity.

Our laboratories are equipped state-of-the-art equipment that allow us to determine the most important combustion-related properties of biomass.

We recognise that it is important that you have confidence in the analytical data that you receive. That is why we follow internationally-recognised standard analysis methods and undertake most analyses in duplicate, reporting values for each of the replicates analysed, along with the average and the standard deviation. This allows us to repeat the analysis (at no extra charge) if the deviation values are high.

Moisture and ash contents are of crucial importance for combustion. This is reflected in our online, Excel, and pdf reports where data for bioenergy-related parameters are expressed on dry-mass, as-received, and dry-ash free bases, according to standard method ISO 16993:2016.



ACHIEVE A PROFITABLE AND SUSTAINABLE PROCESS!

# 12. Technoeconomic Analysis

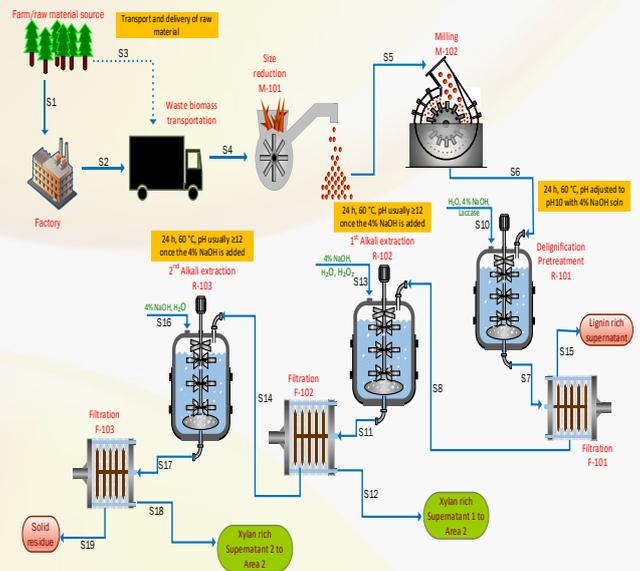
Lab-scale data provide a valuable departing point in the Technology Readiness Level (TRL) spectrum. However, a Techno-Economic Analysis (TEA) is crucial for the successful commercialization of a developed technology. Our staff possess the expertise to develop industrial-scale simulations of lab and pilot scale technologies which are further evaluated to ascertain the economic feasibility of the identified technology.

## METHODOLOGY

Technical modelling and engineering design of pilot and industrial-scale processes are carried out using the experimental data obtained. Rigorous simulation provides reliable mass and energy balance data which constitutes the foundation for equipment design, sizing and specification and utility demand estimations.

The facility design information enables the estimation of capital and operating costs of the proposed production facility using reliable costing models. This is followed by a thorough evaluation of the economic performance of the process. All the technical and economic hotspots in the process flow are identified and modulated to improve the robustness of the process.

A thorough techno-economic evaluation provides more clarity which guides decision making especially in the case of making a significant financial commitment, such as the establishment of a production facility. TEA is valuable in ensuring appropriate resource allocation and identification of main influencing parameters.



## PROCESS SIMULATION

Process simulation enables the model-oriented representation of chemical, physical, biological, and other technical processes as well as unit operations using the appropriate simulation software. This allows for the technical evaluation of a process for the design development, analysis and optimization without having to physically build the process.

With our thorough understanding of chemical, biological, and physical systems, we are able to develop realistic and accurate industrial-scale simulations of lab and pilot-scale processes to find optimal operating scenarios of examined technologies without wasteful expenditure of time and resources. The simulated process largely constitutes the foundation for energy, economic, and sustainability assessment of a technical process or product system.



PIONEERS IN THE BIOECONOMY FOR A DECADE

# 13. HISTORY OF CELIGNIS

1

## Celignis Analytical

Celignis (Ireland) was launched in 2014 as a spin-out from Dan Hayes's PhD on NIR analysis of biomass and his work in the research project DIBANET. The initial focus of Celignis was on compositional analysis services for lignocellulosic feedstocks. Over the years the range of analytical services expanded greatly and the company relocated to larger premises.

2

## Celignis Bioprocess

Celignis in Ireland started offering Bioprocess Development Services (BDS) in 2020 and these have grown greatly since. In 2022, in response to the increased demand for these services at higher technology readiness levels (TRLs), Celignis took a second location, Celignis Bioprocess, focused on BDS projects. This facility has since been populated with an increasing array of bioprocessing equipment up to TRL7 (1 m<sup>3</sup>).

3

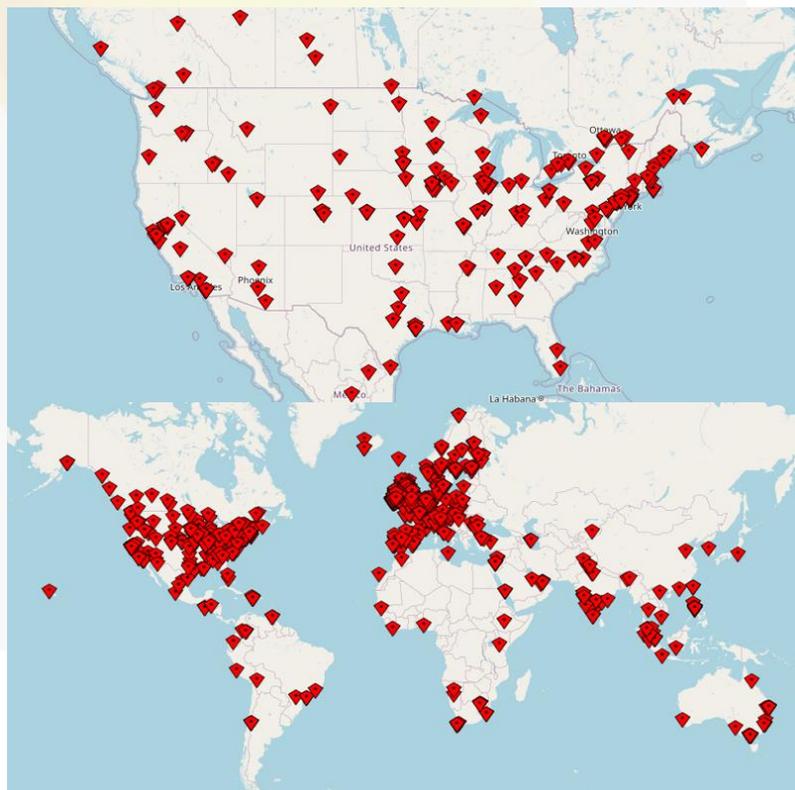
## Growth to Over 1000 Clients

In 2023 Celignis (Ireland) secured its one thousandth client. These customers span the globe and have come to depend on the chemical and process expertise that is prevalent within the Celignis team. Over recent years Celignis (Ireland) also celebrated numerous successes in securing funding for multiple research projects (worth over 6m EUR) to advance the state of the art in biomass valorization.

4

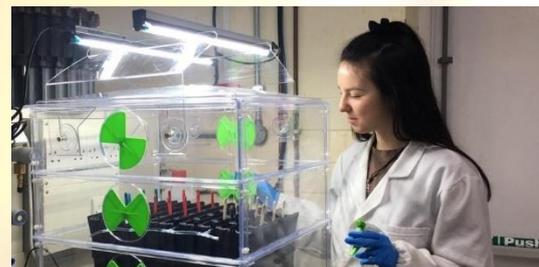
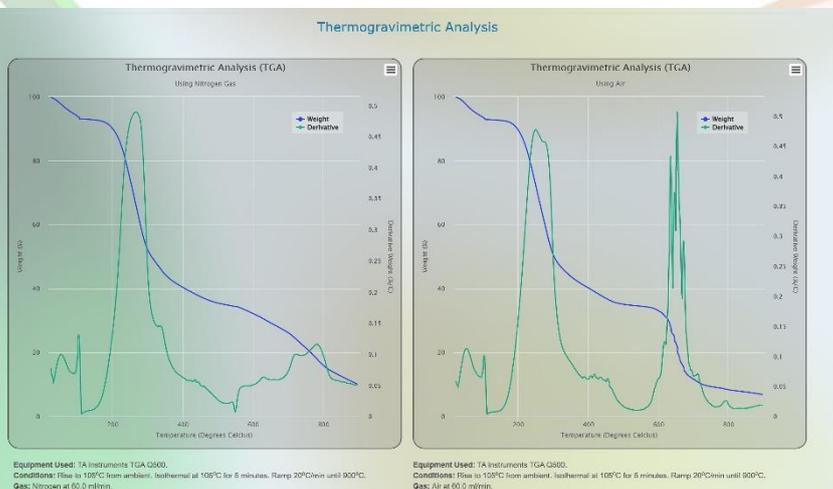
## Launch of Celignis USA

November 2025 marked the launch of Celignis USA. We have the exclusive rights to provide Celignis-branded services to clients throughout the US. Our 10,000 square foot facility is located in Waterloo, Iowa, in the heart of the Midwest. We are looking forward to the closer relationships with Celignis's existing US clients allowed by a national presence in addition to the many new collaborations we will build over the years. Feel free to contact us by email or phone (319-509-3350) and let's work together on developing a vibrant bioeconomy in America!



The Celignis family has provided valued services worldwide since 2014

# 14. SCIENTIFIC RESEARCH



In the bioeconomy if you stand still you end up being left behind. Celignis was born from pioneering research and we are still passionately committed to advancing the bioeconomy.



## 1

### Our Research History

Celignis USA's directors (Lalitha and Dan) have, through Celignis Ltd (Ireland), been involved in over 20 publicly-funded research projects worth over 6m EUR to Celignis. Many of these projects have been large multinational collaborations funded by the European Union, involving multiple research partners.

## 2

### Our Ambitions for the Future

Celignis USA is eager to collaborate in the advancement of the US bioeconomy. We are SAM.gov registered and eligible for full participation in funded projects. We can provide contributions in several domains (e.g. cellulosic fuels, seaweed utilisation, biochar, RNG sector) as a partner focused on analytical or bioprocess development aspects. We can also be involved in the preparation of scientific articles for such projects as well as in the development of proposals for funding.



**Lalitha Gottumukkala**  
Chief Innovation Officer at Celignis

## Selected Projects Involving Celignis USA Personnel



Enxylscope aims at bioprospecting and producing a novel set of xylan debranching enzymes with high catalytic activity and wide operation conditions, thereby demonstrating its ability to make xylan a key ingredient in a variety of consumer products. Celignis (Ireland) is playing a key role in the project, being the technical lead and responsible for the extraction and modification of xylan from biomass.



VAMOS focused on producing and valorising second-gen sugars from municipal solid waste. Celignis (Ireland) was responsible for analysis of feedstocks and outputs and for the development of algorithms to rapidly predict composition using near infrared spectra. We installed NIR equipment at the demo-plant, employing our custom CELDEEP software package.



LIGNOFUN funnels lignin streams into high-value aromatics. Celignis (Ireland) leads WP4, putting crude low molecular weight (LMW) lignin through integrated membrane and solvent fractionation into fractions for bioactivity screening, followed by enzymatic/chemical grafting (e.g., sugars, fatty acids) to tune solubility/surface functionality and stability.



SOLRESS concens the development of an integrated biorefinery to make bio-based solvents from 2G sugars and furans from spent coffee grounds and woody biomass. Celignis (Ireland) roles include: (i) Design and optimisation of a high-density, two-stage Clostridial process for butanol from hydrolysates; (ii) in-situ solvent recovery; and (iii) enzymatic hydrolysis of both feedstocks.



BIONEER aims to widen the range of bio-based building blocks and platform molecules by demonstrating the scaled-up synthesis of a range of novel molecules and polymers from lignocellulosic biomass. Celignis (Ireland) role in the project is demonstrative (1 cubic metre) scale production and modification of xylans and chitosans for the personal care and coatings industry.



PROMOFER address the valorisation of two kinds of feedstock (lignocellulosic biomass and food industry waste) through the improvement of the fermentation processes and downstream purification. Celignis (Ireland) is demonstrating (1 m<sup>3</sup>) xylan extraction and the pretreatment and enzymatic hydrolysis of cellulose residues to glucose for yeast and bacterial fermentations.

## Further Information on Our Research Outputs



### Publications by the Celignis Team

Over 60 peer-reviewed articles by members of the Celignis USA team have been published. Selected articles are listed below:

Rashama, C., [Kuttuvan, S. V.](#), [Gottumukkala, L.](#), Katjouanga, U., [Dobkowski, P.](#), Shiwombolo, J., Hilma, N., Bewer, B., Ben, M., [Hayes, D.](#), Wakefield, D. (2025) Preliminary evaluation of biofuel production potentials for Southern Africa's encroacher and invasive bush biomass, **Bioresource Technology Reports** 31: 102251

Kell, C. J. K., Edison, J., S., [Gottumukkala, L.](#), van Rensburg, E., Tobi, L., Görgens, J. (2025) Anaerobic Co-Digestion of Apple Juice Processing Waste with Manure and Corn Stover; Impact on Biogas and Methane Yield, **Waste and Biomass Valorization** 7(29): 1-9

Ceaser, R., [Bedzo, O. K. K.](#), Donkor, K. O. (2025) Biorefinery approach to producing polysaccharides from seaweed: a focus on hydrocolloids and nanocellulose, **Biomass Conversion and Biorefinery**

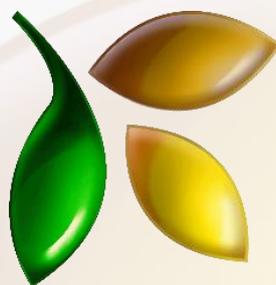
[Bedzo, O. K. K.](#), [Gottumukkala, L. D.](#), Sasso, G. L., Kaminski, K., Schlage, W., Goffman, F., Ivanov, N., Hoeng, J., [Hayes, D. J.](#) (2024) Process development for efficient pectin extraction from tobacco residues and its characterisation, **Biomass Conversion and Biorefinery** 14: 29481-29501

Hamann, M. L., [Bedzo, O. K. K.](#), Diedericks, D., van Rensburg, E., Görgens, J. F. (2024) Autocatalyzed steam pretreatment in a sugarcane biorefinery: optimising for preferred sugar products from bagasse and harvest residues, **Biomass Conversion and Biorefinery** 14: 16107-16120

Gaffey, J., O'Donovan, C., Declan, M., O'Connor, T., Walsh, D., Vergara, L. A., Donkor, K., [Gottumukkala, L.](#), Sybrandus, K., Buckley, E., O'Connor, K., Sanders, J. P. (2023) Synergetic benefits for a pig farm and local bioeconomy development from extended Green biorefinery value chains, **Sustainability** 15(11): 8692

Donkor, K. O., [Gottumukkala, L. D.](#), Lin, R., Murphy, J. D. (2022) A perspective on the combination of alkali pre-treatment with bioaugmentation to improve biogas production from lignocellulose biomass, **Bioresource Technology** 351

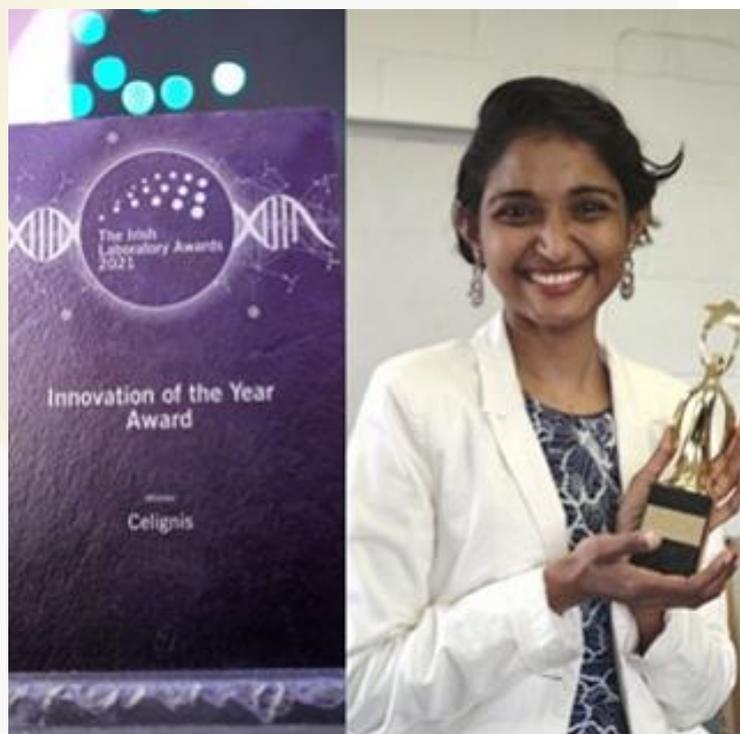
Swart, L. J., [Bedzo, O. K. K.](#), van Rensburg, E., Gorgens, J. F. (2021) Intensification of Xylo-oligosaccharides Production by Hydrothermal Treatment of Brewers Spent Grains: The Use of Extremely Low Acid Catalyst for Reduction of Degradation Products Associated with High Solid Loading, **Applied Biochemistry and Biotechnology** 193: 1979-2003



# Celignis USA

### Publications Including Celignis Data

A literature search has found close to 100 peer-reviewed scientific articles written outside of Celignis but incorporating some data provided by Celignis.



We're scientists with business brains.  
Let us help you find the right path!

WE MAKE IT EASY TO ACCESS AND UNDERSTAND YOUR DATA

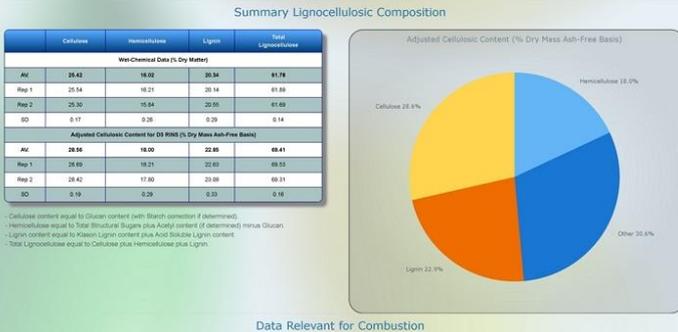
# 15. The Celnignis Database & AI

## Concept

Our slogan, "With Accurate Data the Opportunities are Limitless", reflects the modern age, where data is a powerful resource. We recognise that data is of greatest value when results are presented in a detailed, accessible, manner so that impactful decisions can be made in a rapid timeframe. This is why the Celnignis Database is so popular with our clients. It allows for orders to be placed and monitored and for results to be viewed through a user-friendly online interface. We upload results as obtained meaning that you do not have to wait for an order to be completed to get valuable data. In the case of our biomethane-potential (BMP) tests for the RNG sector, we update our biogas plots daily allowing you to fully monitor the process and make early decisions.

## Detailed Reporting

We present summary results as well as detailed data for each sample with results represented in tabular and graphical forms. We also use the data to estimate potential biofuel yields. Data relevant to thermal processing and biomethane potential are shown on dry-mass, as-received, and dry-ash-free terms. Our Detailed Excel and PDF reports provide data for each of the duplicates analysed, the average, and the standard deviation, so you can see the precision of our work. Our Database also has a real-time messaging system allowing you to directly contact the team with your queries.



Sample Name	Glucan						Xylan						Mannan						Arabinan						Galactan						Rhamnan					
	Av.	R1	R2	SD	Av.	R1	R2	SD	Av.	R1	R2	SD	Av.	R1	R2	SD	Av.	R1	R2	SD	Av.	R1	R2	SD	Av.	R1	R2	SD								
25003	13.52	13.47	13.57	0.07	2.48	2.47	2.51	0.03	1.81	1.81	1.81	0.00	0.19	0.19	0.19	0.00	1.72	1.71	1.73	0.02	0.87	0.84	0.89	0.03												
25004	16.22	16.07	16.37	0.21	3.86	3.84	3.89	0.03	2.24	2.22	2.28	0.03	0.38	0.38	0.37	0.01	1.85	1.82	1.88	0.04	1.41	1.38	1.43	0.04												
25005	19.28	19.15	19.43	0.25	2.72	2.68	2.76	0.05	2.92	2.92	2.91	0.01	0.30	0.30	0.30	0.00	2.31	2.29	2.32	0.03	1.08	1.06	1.10	0.02												

## Celnignis InSightAI

Celnignis InSightAI is a powerful artificial intelligence tool developed by Celnignis and integrated with the Celnignis Database in 2026. It functions as a highly-customised chatbot, focused on providing insights and conclusions from the data displayed on the page. The user can interact with Celnignis InSightAI using a chat window in the screen corner. The tool is provided free of charge to clients of Celnignis USA, with any analysis order placed through the Celnignis Database and can be also used for past orders.



Our Database covers over 170 packages and over 400 analytes

**Explore the results**  
Ask questions. Get clear answers.

**Summary of Analytical Procedure**

This sample was dried and then extracted with water-soluble and ethanol-soluble components, and the remaining solid residue was extracted using 20% ethanol. The original sample was also extracted using 20% ethanol. These operations allowed for the determination of the total contents of water-soluble extractives, ethanol-soluble extractives, and water-insoluble but ethanol-soluble extractives, as well as the total extractives removed under a sequential extraction using water then ethanol.

Following the removal of the water and ethanol soluble components, the sample was hydrolysed using acid in order to determine the lignin (Rovani and acid soluble) and lignocellulosic sugars (glucose, xylose, arabinose, galactose, mannose, rhamnose) contents.

The sample was also analysed for its ash content and its elemental composition (carbon, hydrogen, nitrogen, sulphur, oxygen) with the oxygen content determined by difference.

Its higher (LHV, often referred to as the Gross Calorific Value) and lower (LHV, often referred to as the Net Calorific Value) heating values were then calculated from its elemental composition.

The sample was also analysed for its moisture content.

**Lignocellulosic Data**

**Sugars Data**

**Aldoses**

**Monosaccharides**

**Disaccharides**

**Polysaccharides**

**Cellulose**

**Hemicellulose**

**Lignin**

**Other**

**AI Chat Window:**

Ask about these results

Biogas: The large soluble/extractable fraction (20% extractive; water-soluble 24%) implies an immediately bioavailable fraction that will be readily digested/fermentable.

- Acetone-digestion (AD) or received moisture is suitable for wet AD. The C4 + 21.1 is within a typical acceptable range (optimal < 20-28.1) so no substrate may not be required for C/N balancing. High soluble organics and sugars predict high biochemical methane potential (BMP), but protein content (< 15.6% dry mass) ammonia release could be significant at high loading rates. - consider BMP testing and/or anaerobic toxicity assay (AT) if planning full scale AD.
- For enzymatic hydrolysis/fermentation: glucose is moderate; pretreatment may improve cellulose accessibility because lignin (~14%) and associated structural complexity can limit hydrolysis yields. The soluble extractive fraction means some sugars may be recoverable by aqueous extraction without harsh pretreatment.

Recommended additional tests for biochemistry and:

- Biomethane potential (BMP) and anaerobic toxicity assay (AT) - to quantify methane yield and inhibition risk.
- Sugar in solvent extract (water-soluble sugars) - package to quantify the soluble mono-/disaccharides (P2/P12).
- Starch (P14) if you need to confirm non-structural glucan contributions.
- Define lignocellulose package (P19) if you want the most accurate compositional data including corrections for starch and protein effects on lignin.

2. Thermal conversion (combustion, gasification, pyrolysis, biochar)

- Challenge: High as received moisture (20%) will substantially reduce net energy yield unless dried. Ash (8.4%) is moderate - acceptable in some thermal systems, but ash handling (melting, sintering) is unknown and important for boiler/gasifier operation.
- Element N (2.14%) can lead to NOx formation during combustion. S is low (0.09%).
- For pyrolysis/biochar: significant volatile and extractive fractions can increase condensable tars and affect biochar quality/yield.

WE ARE ALWAYS HAPPY TO CHAT ABOUT BIOMASS!

# 16. Contact Details



## WEBSITE

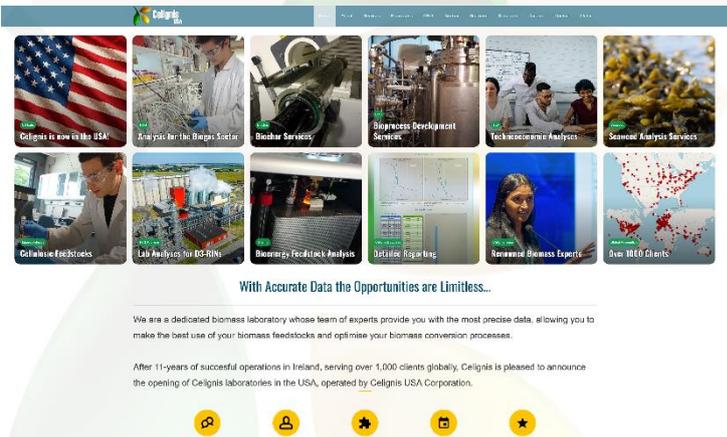
There is lots of information at [www.celignis.us](http://www.celignis.us) on our various analysis packages and the wide range of analytes we determine. We also present more detail on our bioprocess development services and TEA work as well as the many feedstocks we have experience with.

## ADDRESS

You are most welcome to visit us! We are located in a 10,000 square foot facility in Waterloo, Iowa and are able to receive shipments from all 50 US states from USPS and all major courier companies. Our facility is truck-friendly with a standard loading dock (no liftgate required). You will receive automated emails from the Celignis Database when your samples arrive and as data are uploaded.

## PHONE + EMAIL

Our email is [info@celignis.us](mailto:info@celignis.us) or call our analytical labs at 319-509-3350. We're also on social media: LinkedIn (@celignis-usa-corporation) and X (@celignisusa).



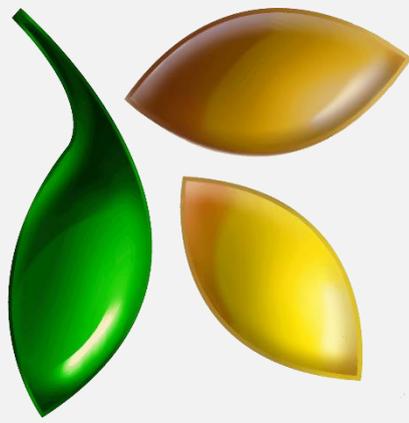
## YOUTUBE CHANNEL

The Celignis YouTube channel (@Celignis) has numerous videos covering our analysis methods, research outputs, and interviews with members of the Celignis family. For example, you can watch a video outlining our procedure for determining the biomethane potential (BMP) of feedstocks and a video detailing an experiment evaluating different pretreatment approaches for the release of sugars from lignocellulosic biomass.

## NEWSLETTER

Celignis routinely issues a newsletter informing our clients on our latest developments. You can sign up for this newsletter at [www.celignis.com/usa\\_contact](http://www.celignis.com/usa_contact).





# Celignis USA

# Bioprocess Development Services

FROM A LAB DEDICATED TO  
ADVANCING THE BIOECONOMY

## CELIGNIS LOCATIONS



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