Marine By-products: Raw Material use for Functional Ingredient Development

Maria Hayes [1] & Tomas Lafarga[1]

[1] Food Biosciences, Teagasc Food Research Centre, Ashtown, Dublin 15

Outline

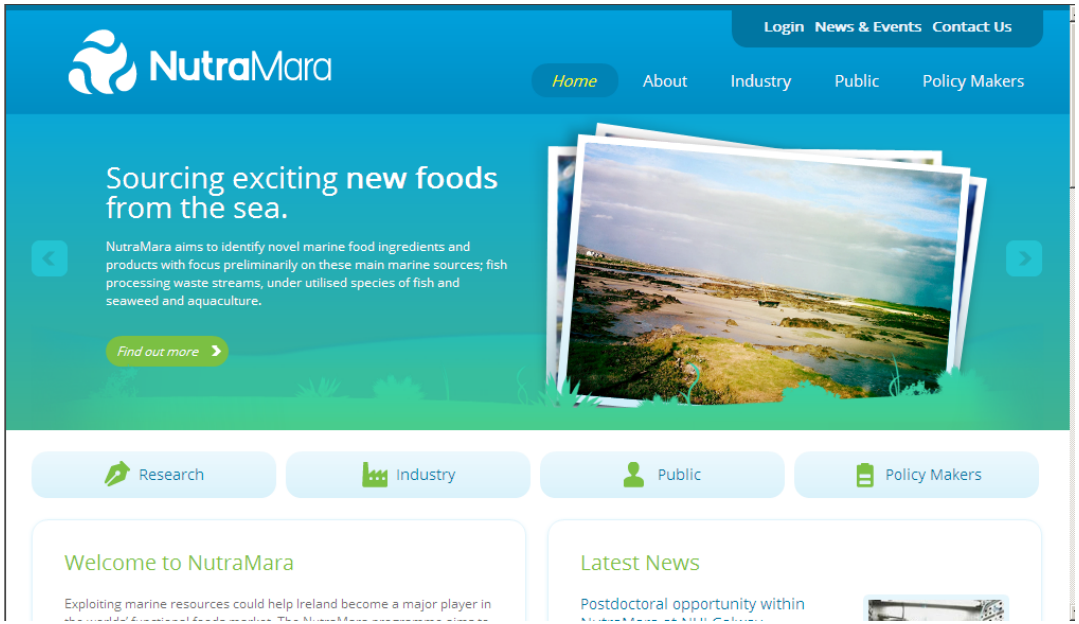
- Introduction
- Why By-products could be used for ingredient production?
- What by-product ingredients can be generated?
- How these are generated? Processes
- Applications?

Changes to the EU Common Fisheries Policy

- February 2013, the European Parliament approved a package of major reform to CFP.
- Introduction of an Eco-Friendly system based on Maximum Sustainable Yields (MSY).
- By-products may be viewed as a valuable source of bioactive compounds including Functional and Technologically relevant peptides and carbohydrates.

The Marine Functional Food Research Initiative

NutraMara



Marine Institute
Foras na Mara

Department of Agriculture, Food and the Marine
An Roinn Talmhaíochta, Bia agus Mara

University of Limerick
Ollscoil Luimnigh

National University of Ireland, Galway
Ollscoil na hÉireann, Gaillimh

University of Ulster

UCD Dublin

Teagasc
Agriculture and Food Development Authority

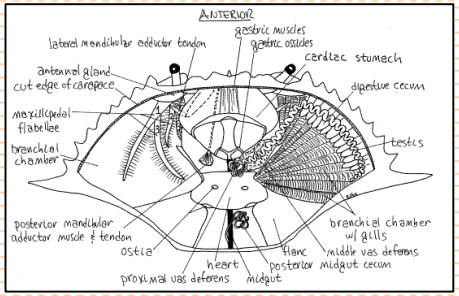
The Irish Agriculture and Food Development Authority

Sources

Macroalgae

Microalgae

By-product/co-product



Cancer pagurus

Shell and leg which contain meat

Fish scales

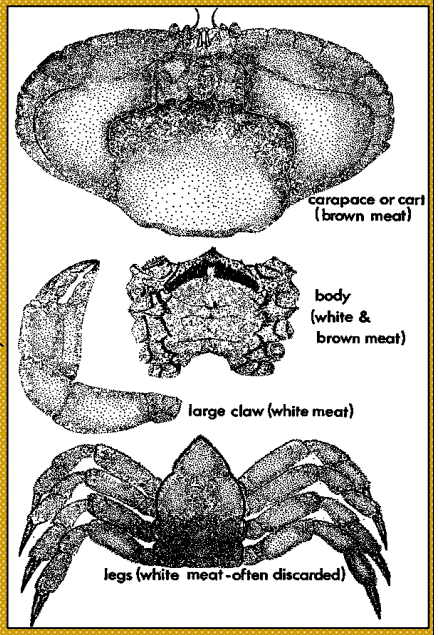
Fish gelatin

Herring Mackerel

Fish fins

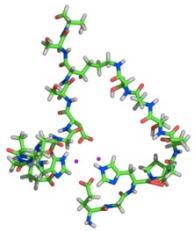
Fish heads

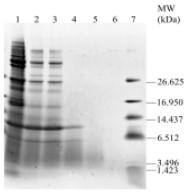
White and brown crab meat



Algal bioactive ingredients

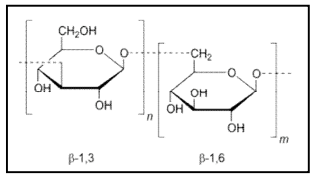
- Antioxidants
- Blood pressure
- Cancer
- Inflammation
- Obesity
- Diabetes
- Mental health

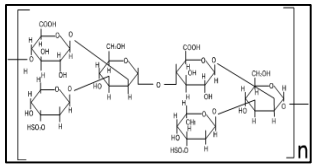




Peptides

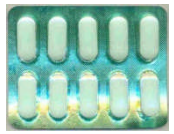
Sugars



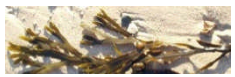


Laminarin

Fucoidan



LIPIDS & small molecules (phlorotannins, carotenoids)



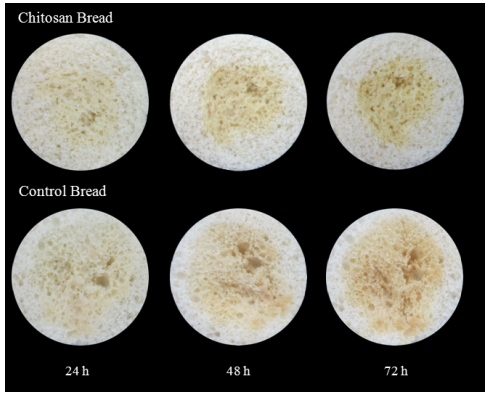
Teagasc
Agriculture and Food Development Authority

Development of Chitosan Bread: Food Safety

(A) Control (B) Chitosan

Figure 4. C-CELL image of white bread (A) and 1% chitosan-containing bread (B).

Inhibitory effects of chitosan bread against *B. cereus* and rope formation when directly inoculated in control and chitosan-containing bread (A)



Bacillus cereus

Potential Hazards…

Waste shellfish prolongs shelf life of sliced pan

Eithne Shortall

TEAGASC, Ireland's agricultural research body, has developed a way of using discarded shellfish to prolong the shelf life of an Irish household staple — the sliced pan.

The research, which was recently published in an international journal, found that by replacing just 1% of a loaf of bread with a substance located in the shells of prawns and crabs, the food could go an extra couple of days without becoming "ropey".

The findings are a promising solution to the 750,000 tons of waste shellfish accumulated in the EU annually which is expensive to discard, and quick spoilage and mould growth affecting Ireland's €500m bread industry.

The researchers extracted a substance known as chitin from the shells and proteins of prawns that were not of a high enough quality to be sold for consumption. This was then injected into bread, which was left to "spoil" at an accelated rate. The spiked bread showed significantly less degradation than untouched bread, and it was estimated it would be safe to eat for up to two days longer.

Teagasc published its findings in the Journal of Agricultural and Food Chemistry, and it has been contacted by several commercial bodies that are interested in bringing the procedure to the mass market.

The research team was led by Tomas Lafarga, a visiting PhD student from Spain. Maria Hayes, a research officer on the project, said they had tested for the "ropey" or "gooey" degradation of bread, which occurs when it is left out in heat. However, they also found that more common mould spotting did not occur when chitin was injected into the product. While further research is needed, Hayes said the findings are promising.

Waste shell material is an environmental and economic problem, because dumping it in landfill is expensive and the EU has tightened regulations regarding its disposal at sea. "When you have prawns or crab shell material, there's a grading system, so some of them aren't up to scratch in terms of what the consumer wants. They might not be ultra fresh," said Hayes.

"So instead of dumping the waste back at sea, marine pastures have to store it, and that's a cost."

Hayes said that all evidence suggests chitin is safe to eat, and that it is also considered an "anti-obesity agent".

- May not be suitable for persons with shellfish allergies
- Depends on the purity of Chitosan and if all protein is removed during the production process

Proteins For The Future

- Between 1995 and 2020, about 97.5 percent of the global population increase will be in developing countries, by which time 84 percent of the world's people (an estimated 6.3 billion) will be living in developing nations.
- Need to widen the choice of dietary proteins for consumers
- Up-scaling and sustainable production and use of proteins from alternative marine sources is required

GELATIN IN THE PAST AND PRESENT [1]

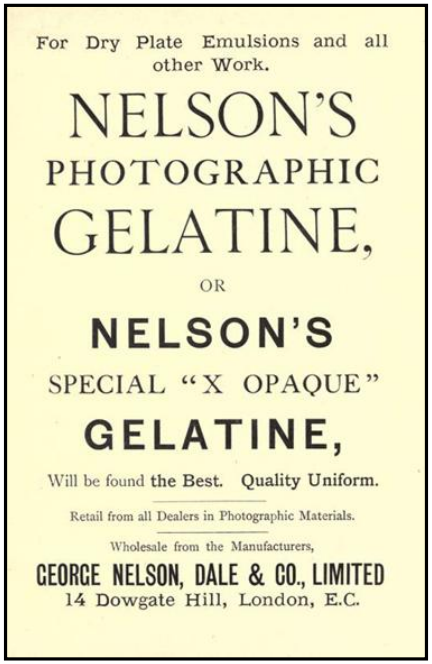
1682 The Frenchman Papin reports on a cooking process in which he tried to obtain a jelly-like mixture from bones

1700 Gelatine (*Latin: gelatus* = stiff, frozen). The word gelatin is first used in Europe around 1700

1754 The first patent in the adhesive sector including gelatin, is granted in England in 1754.

1871 Profound discoveries by the English doctor Richard Leach Maddox lead to decisive breakthrough in photography.

1875 This year is considered to be a milestone in modern gelatin manufacture: emergence of small factories and large quantities of gelatine could be manufactured.



1950 The gelatin industry sees rapid technological developments and makes tremendous progress towards achieving high standards in the production of gelatine and the quality products

1956 First studies and publications regarding generation of gelatine from fish materials

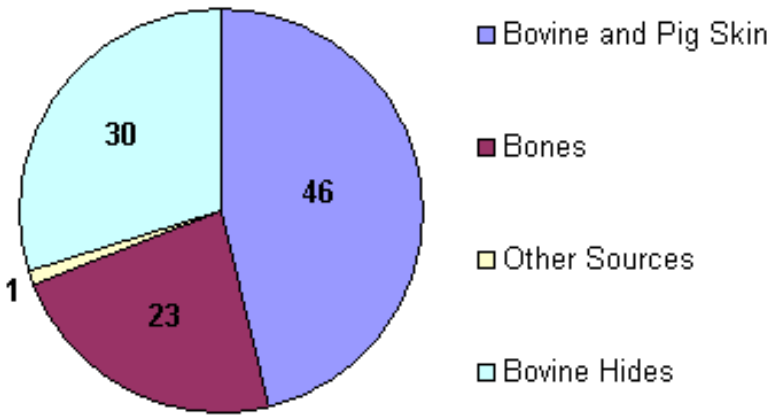
2001 An international study initiated by the European Gelatine Association (GME) in 1999 confirmed once again that raw material regulations and gelatine production processes guarantee maximum safety for consumers.

2013

[1] Source: Modified from All About Gelatin. The Gelatin Manufactures of Europe (GME).

ACTUAL SOURCES OF COLLAGEN AND GELATIN

- Gelatin is derived mainly from bovine and pig skin (**46%** of worlds gelatin output).
- Bones of pigs and cattle represent **23%** of world output.
- Only **1%** of world gelatin output comes from marine sources: skins, scales and bones of fish.



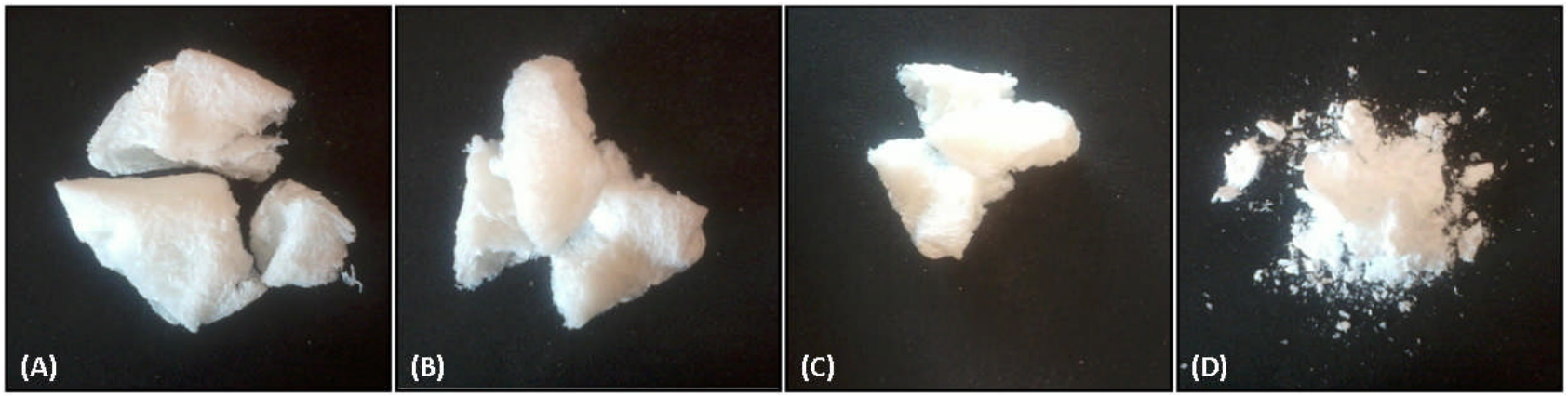
ADVANTAGES OF FISH SKIN COLLAGEN/GELATIN

- Fish Gelatin releases aroma and shows a higher digestibility than animal gelatin.
- Lower gelling temperature: advantageous for certain uses such as in electronical trade or precipitation of emulsions.
- No risk of health-threatening outbreaks of bovine spongiform encephalopathy (BSE) and foot and mouth disease (FMD).
- Acceptable for Hindus and members of the Islamic and Jewish faiths.
- Most gelatine-allergic patients do not react to fish skin gelatin.

[1] Source: Gelatine information, news, history and more. Gelatine Manufacturers Institute of America. Retrieved 2008-09-26.

Gelatin/Collagen from Fish By-Products

Gelatin generation methods from Salmon co-product cuts



Gelatin generated with the NaCl method: (A) Salmon Bones, (B) Salmon Skin, (C) Salmon Heads and (D) Salmon Offal

Underutilised Fish Species: Potential raw material for peptide generation

- White fish including blue white and herring

- *Capros aper* (Boarfish)

Amino acid content of blue whiting protein mince extract and boarfish myofibrillar protein

Amino acid Content	Blue whiting mince extract Concentration g/Kg extract powder	Boarfish Myofibrillar protein extract Concentration g/Kg powder
Cysteic Acid	11.747	0.093
Methionine Sulfone	35.178	
Asp	78.602	2.439
Thre	32.52	0.699
Ser	29.596	0.672
Glu	109.035	3.38
Gly	29.523	0.999
Ala	40.341	1.269
Cys	5.565	1.496
Val *	38.39	0.826
Met		0.496
Ile *	36.31	0.445
Leu *	54.745	1.136
NorLeucine	16400	16400
Tyr	24.538	0.649
Phe	30.685	0.856
His	23.023	0.563
Lys	62.605	1.118
NH3	10.329	117.406
Arg	47.991	2.152
Pro	24.305	0.854

*** BCAA = Branched chain amino acids**

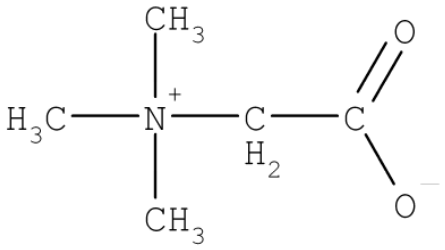
Hayes *et al.*, 2013

PEPTIDES FROM MARINE SOURCES

Company Name	Commercial Name	Source	Bioactivity
Senmie Ekisu	Valtyron (sold as ingredient for supplements or foods)	sardine muscle - dipeptide (Val-Tyr)	ACE inhibitor + anti-hypertensive
Nippon Supplement	Katsuabushi oligopeptide	bonito -oligopeptide converted to bioactive tripeptide (Leu-Lys-Pro)	ACE inhibitor + anti-hypertensive
Natural Factors	PeptACE	bonito -oligopeptide converted to bioactive tripeptide (Leu-Lys-Pro)	ACE inhibitor + anti-hypertensive
Metagenics	Vasotensin	bonito -oligopeptide converted to bioactive tripeptide (Leu-Lys-Pro)	ACE inhibitor + anti-hypertensive
Calpis	Ameal	tri-peptide (Val-Pro-Pro)	anti-hypertensive
Tokiwa Yakuhin	Lapis Support	dipeptide (Val-Tys)	anti-hypertensive
Copalis	Protizen	white fish hydrolysate	stress relief
Yalacta	Stabilium200	atlantic fish autolysate	Stress Relief
Biothalassol	Fortidium Liquamen	white fish hydrolysate (Molva Molva)	stress relief, anti-oxidant, lowers GI
ProperNutrition	SeaCure	fish fillet hydrolysate	IBS, Ulcerative colitis and Crohn's disease
Copalis	Nutripeptin	white fish hydrolysate	lowers postpradial blood glucose
Copalis	Collagen HM	hydrolysed fish collagen	skin & cartilage regeneration
Copalis	Prolastin	hydrolysed fish skins	promotes ligament regeneration + anti-oxidant

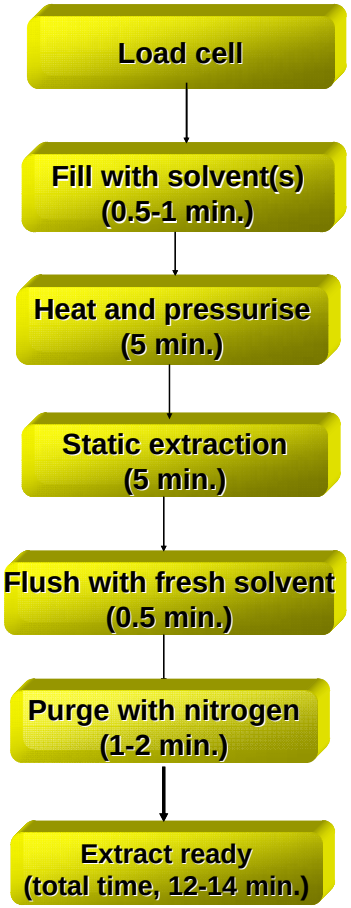
Glycine Betaine & DMSP

- Glycine betaine and DMSP are osmolytic, zwitterionic compounds found in food, microalgae and seaweed.
- Formed by oxidation of bioactive choline in mammals and helps to maintain normal cell volume under osmotic stress.
- The beneficial effects of glycine betaine relate to the maintenance of normal blood concentrations of homocysteine.



- In 2011, the Panel on Dietetic Products, Nutrition and Allergies (NDA) of the European Food Safety Authority (EFSA) established a cause and effect relationship between consumption of glycine betaine and normal homocysteine metabolism and granted it a health claim pursuant to Article 13 of regulation (EC) No. 1924/2006.

Generation of Glycine betaine and DMSP enriched seaweed extracts: Accelerated Solvent Extraction (ASE).



ASE

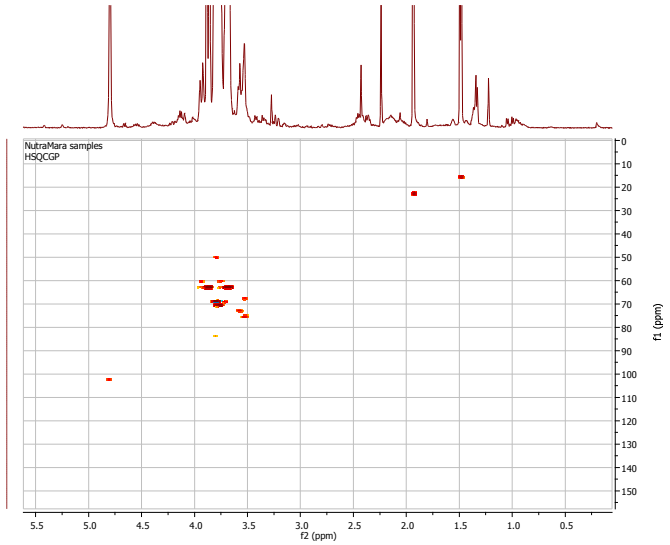
Location and date of collection of seaweed samples analysed in this study.

Code	Name	Location	Date
ISCG0033	*Alaria Esculenta*	Spanish point Co. Clare	03/05/2009
ISCG0365	*Ascophyllum nodosum*	Finnavara Co. Clare	08/02/2012
ISCG0029	*Codium fragile*	Spiddal, Co.	02/02/2009
ISCG0070	*Cystoseira nodicaulis*	Finnavara Co. Clare	30/03/2010
ISCG0442	*Cystoseira tamariscifolia*	Finnavara Co. Clare	04/05/2012
ISCG0283	*Cystoseira tamariscifolia*	Finnavara Co. Clare	28/09/2011
ISCG0259	*Fucus serratus*	Spiddal, Co.	18/07/2011
ISCG0239	*Fucus spiralis*	Finnavara Co. Clare	16/06/2011
ISCG0238	*Fucus vesiculosus*	Finnavara Co. Clare	16/06/2011
ISCG0223	*Fucus vesiculosus*	Golf Course,	08/06/2011
ISCG0072	*Fucus vesiculosus*	Spiddal, Co.	16/04/2012
ISCG0257	*Laminaria digitata*	Spiddal, Co.	18/07/2011
ISCG0463	*Osmundea pinnatifida*	Finnavara Co. Clare	05/06/2012
ISCG0071	*Palmaria palmata*	Spiddal, Co.	04/11/2010
ISCG0355	*Pelvetia canaliculata*	Spiddal, Co.	24/01/2012
ISCG0462	*Polysiphonia lanosa*	Finnavara Co. Clare	05/06/2012
ISCG0028	*Saccharina lattisima*	Finnavara Co. Clare	26/03/2009
ISCG0356	*Ulva Intestinalis*	Spiddal, Co.	24/01/2012

Generation of Glycine betaine and DMSP enriched seaweed extracts: Accelerated Solvent Extraction (ASE).

- Extraction of polar metabolites DMSP and Glycine betaine was carried out using the DIONEX ASE 200 system and water with pressure and temperature of 120 bars & 50˚C; 3 cycles of 5 minutes duration.
- Recovered ASE$^{(R)}$ fractions were centrifuged and aliquots dried under nitrogen and subsequently freeze dried prior to ^{1}H-NMR analysis.

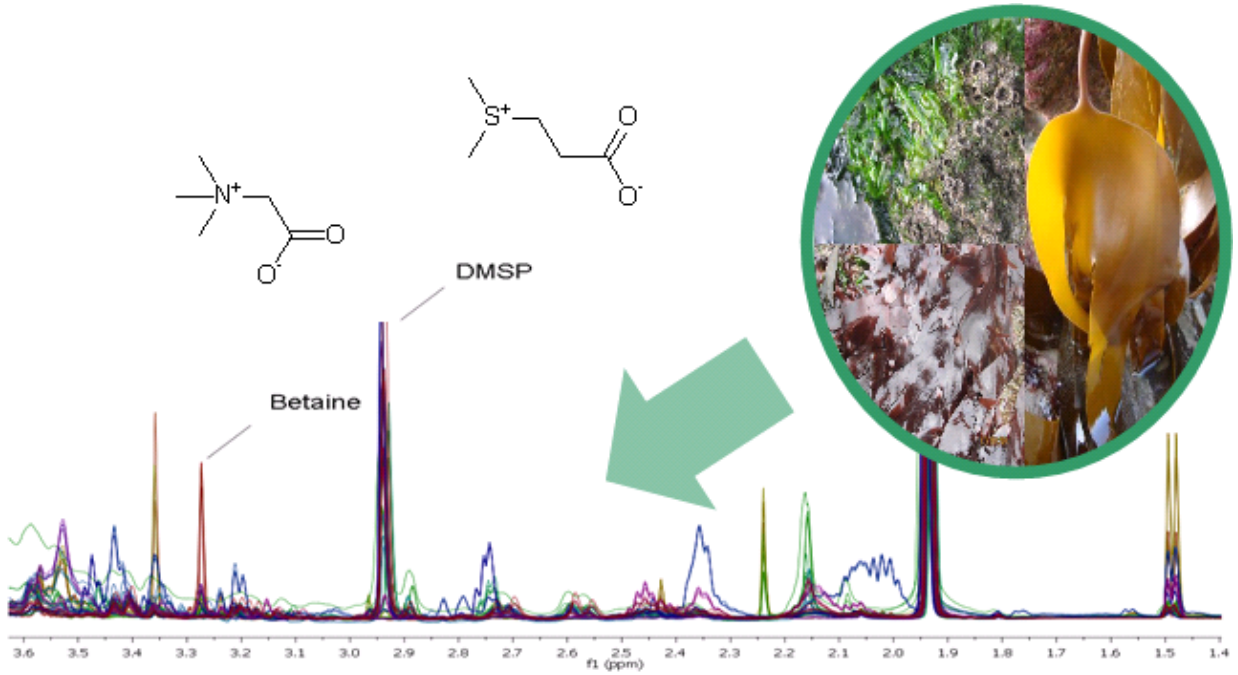
Using ^{1}H-NMR for metabolite identification



500.162 MHz
300 K
5 mm PABBO
Broad Band
Observe probe

3-(trimethylsilyl)propionic acid-d_4 (TSP) sodium salt was used as an internal standard and resonance identification of samples was performed by comparison of reference compounds acquired under similar conditions.

Using ^{1}H-NMR for metabolite identification

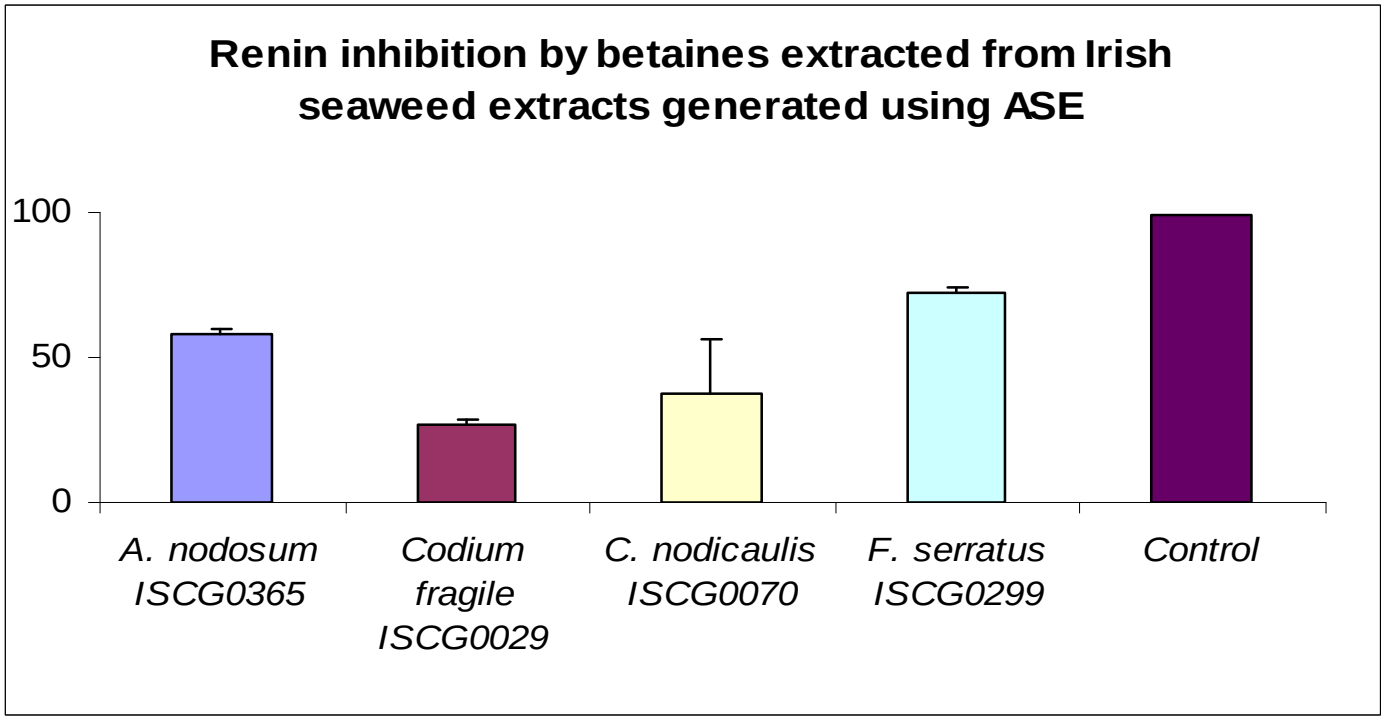


Acquired Spectra were Line broadened manually phased and Baseline corrected using MestReNova v6.0.2
Baseline correction was carried out using Whittaker smoother algorithm.

Estimated quantities of glycine betaine and DMSP found in different macroalgae ASE® extracts in this study.

CODE	Name	Concentration (µmol / g DM)			
		ASE® extract		Water extract	
		Glycine betaine	DMSP	Glycine betaine	DMSP
ISCG 0033	*Alaria esculenta*	8.43 ± 1.92	ND	*	*
ISCG 0365	*Ascophyllum nodosum*	ND	94.51 ± 18.54	ND	63.18 ± 3.15
ISCG 0029	*Codium fragile*	7.44 ± 0.86	130.46 ± 3.31	ND	33.83 ± 20.26
ISCG 0070	*Cystoseira nodicaulis*	13.69 ± 1.82	12.18 ± 1.48	ND	ND
ISCG 0283	*Cystoseira tamariscifolia*	13.6 ± 2.47	ND	ND	ND
ISCG 0442	*Cystoseira tamariscifolia*	6.27 ± 0.97	ND	ND	ND
ISCG 0259	*Fucus serratus*	ND	31.55 ± 7.1	ND	31.70 ± 2.17
ISCG 0239	*Fucus spiralis*	ND	110.67 ± 2.87	ND	89.57 ± 6.56
ISCG 0238	*Fucus vesiculosus*	ND	93.61 ± 13.45	ND	67.75 ± 4.70
ISCG 0223	*Fucus vesiculosus*	ND	56.27 ± 15.36	*	*
ISCG 0072	*Fucus vesiculosus*	ND	74.86 ± 8.3	ND	86.00± 44.77
ISCG 0257	*Laminaria digitata*	ND	ND	ND	ND
ISCG 0463	*Osmundea pinnatifida*	ND	ND	ND	ND
ISCG 0071	*Palmaria palmata*	11.41 ± 3.01	ND	8.80 ± 1.15	ND
ISCG 0355	*Pelvetia caniculata*	ND	119.12 ± 38.02	ND	101.92 ± 5.32
ISCG 0462	*Polysiphonia lanosa*	ND	119.17 ± 10.58	ND	43.30 ± 1.29
ISCG 0028	*Saccharina lattisima*	6.62 ± 0.61	ND	*	*
ISCG 0356	*Ulva intestinalis*	4.38 ± 0.38	77.16 ± 10.56	ND	53.54 ± 10.14

Bioactivities – Renin Inhibition



Potential applications of Betaine extracts…

- Application in baked goods to prevent loss of moisture
- Extracts use in sports performance products
- Cognitive function applications for pregnant women

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* Kilibwa, United States Patent US 6,217,930 B, April 2001

** Pryor, J. L., et al., (2012) Effect of betaine supplementation on cycling Sprint performance, Journal of the International Society of Sports Nutrition, 9, 12.

*** Asia Pac J ClinNutr, 2013, 22, 2, 319-324.

FACTORS OF IMPORTANCE

- Safety of ingredients generated
- Compliance with labelling laws and EFSA regulations for Novel Foods and Nutritional claims

Department of Agriculture, Food and the Marine
An Roinn Talmhaíochta, Bia agus Mara

Education and Culture DG
Lifelong Learning Programme

Thank you for your kind attention

Contact details:

maria.hayes@teagasc.ie

Tel: +353 (0) 1 8059957

Thomas.lafarga@teagasc.ie