- → Professor Margareth Øverland
- → Foods of Norway, Fakultetet for Biovitenskap, NMBU
- → June 18, 2025

# How do we feed our salmon to meet the protein requirement?

Lecture in the immunonutrition NOVA/SDG pilot call cource



### SDG pilot call - INTFELLES Global Research Council



Sharing knowledge and smart technologies for sustainable agro-aquatic production systems

NOVA course - a joint PhD level course financed by the Nordic Forestry, Veterinary and Agricultural University Network (NOVA).





Supporting the development of international scientific networks.

















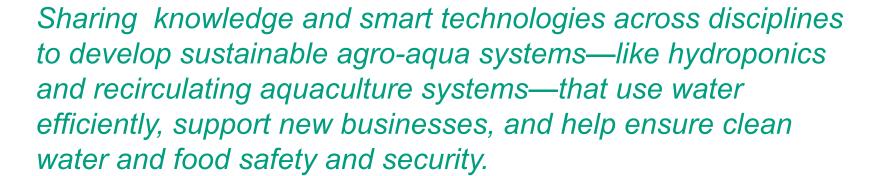




#### SDG Pilot Call Partners:

University of the North, Chile, Project lead, Prof. German Merino Norwegian University of Life Sciences, NMBU, Norway, Prof. Margareth Øverland Bursa Uludağ Üniversity, Turkey, Prof. Nezih Kamil Salihoglu

## Aligning UN SDGs 2, 12, 13 & 17







SDG pilot call



**Responsible Consumption & Production** 



**Climate action** 



Partnership to achieve the goals



### **Outline**

N M H

- Introduction
- History of Atlantic salmon farming
- Why is protein important for fish
- Amino acids needed for growth and health
- How do we measure the protein and amino acid requirements
- How do we measure the protein quaility of feedstuffs
- Amino acid composition and digestibility in common feedstuffs
- How do we feed the fish to meet the requirement to day and in the future
- Concluding remarks





# History of aquaculture at our university

Domestic Atlantic salmon originates from 41 rivers in Western Norway

We have been teaching aquaculture since 1887





Aquaculture lab from 1900
Prof. Harald Skjervold, the farther of farmed salmon

# Development of modern fish feeds



- Methods to determine:
  - nutritional value feed
  - nutritional requirement of salmon

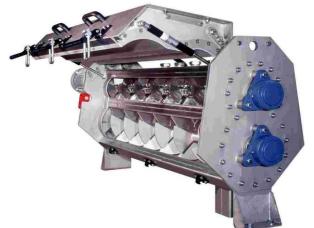
#### Major breaktrough

- Change from wet to dry feed
- Increased fat content in the feed
- New feed processing technology
  - Extrusion
  - Vacuum coating



### Extrusion technology revolutionized the modern fish feed





Conditioner: ~90°C

1983: First commercial extruded salmon feed

1983-1990: Extruded dry feed dominated Norwegian aquaculture

1990: New methods in feed technology, e.g. vacuum coating



Extruder: ~110-140°C



Dryer: ~90-110°C







# All its research aims at finding sustainable alternatives to fishmeal in diets for carnivorous fish

2015: Foods of Norway was established



Ended in 2013

Established in 2003

APC has given:
Important new expertise
New methods to measure amino acid requirements
Knowledge of use and limitations of new ingredients
New methods for processing of fish feed
New knowledge on health effects of bioactive
components in novel feed ingredients
Integrated expertise important in a global context



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#### Industrial partners





Felleskjøpet

geno

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**ANIMALIA** 































denofa





#### Academic partners and collaborators



Norwegian University of Life Sciences





UNIVERSIDAD DE CHILE





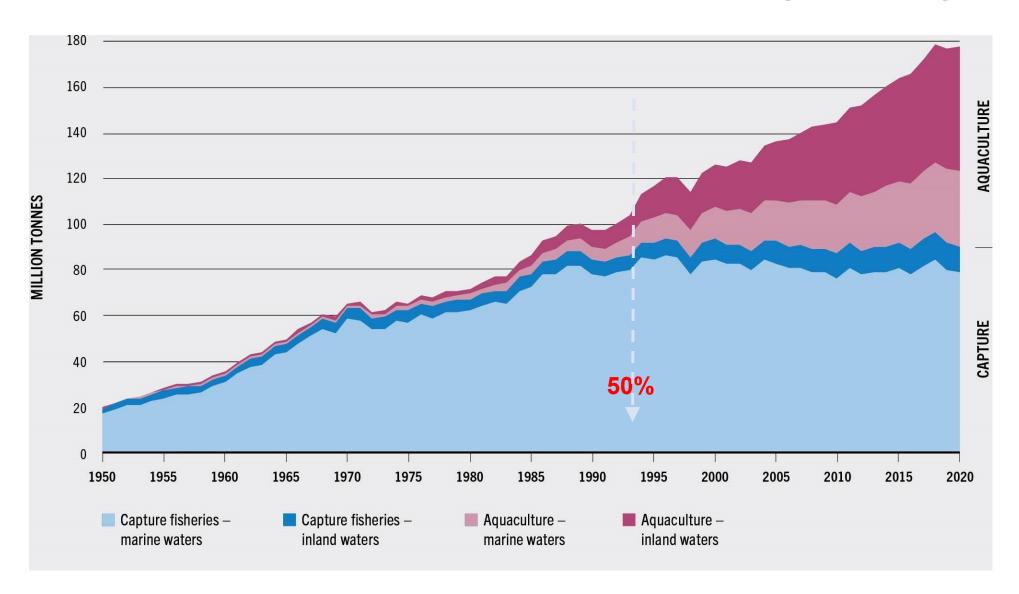








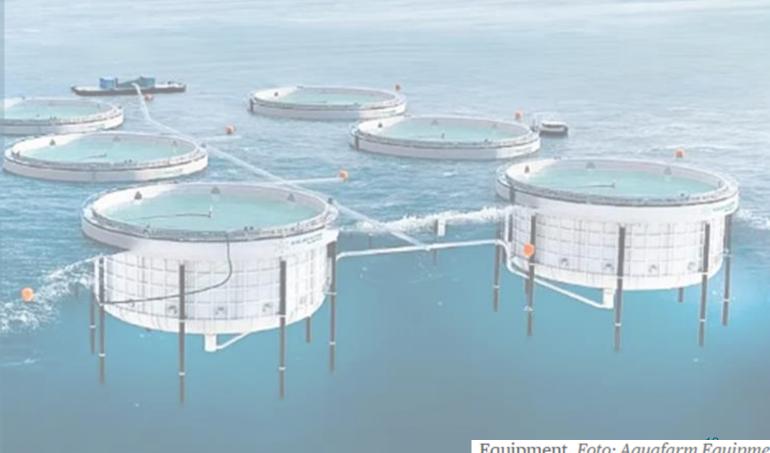
# Aquaculture production keeps growing



# Constraints on growth of the aquaculture industry

 Limited access to high quality feed raw materials

 Increased demand for sustainable raw materials

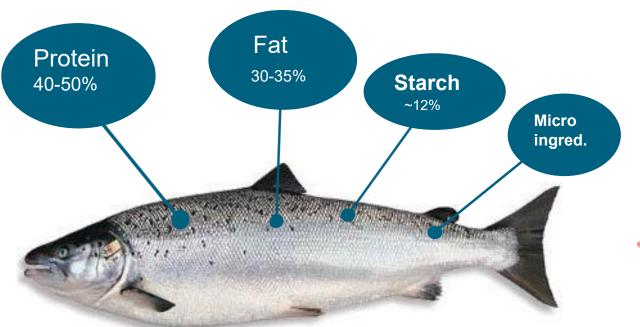


#### **FOODS?NORWAY**

# Meeting the nutritional requirements of the fish

Fish don't need to eat fish to grow, but they require essential nutrients

#### Feeding is about finding the right balance





Feed is the largest cost component in salmon production: 46% of the total production costs. Protein is the most expensive part of the diet and accounts for 50-60% of the total value.



You need to know the <u>nutrient content</u> and <u>digestibility</u> of the raw materials and how well they are <u>utilized</u> to formulate diets to match the requirements of the fish.

<sup>\*</sup> with current conversion ratio measured in dry weight

## Protein requirement of fish

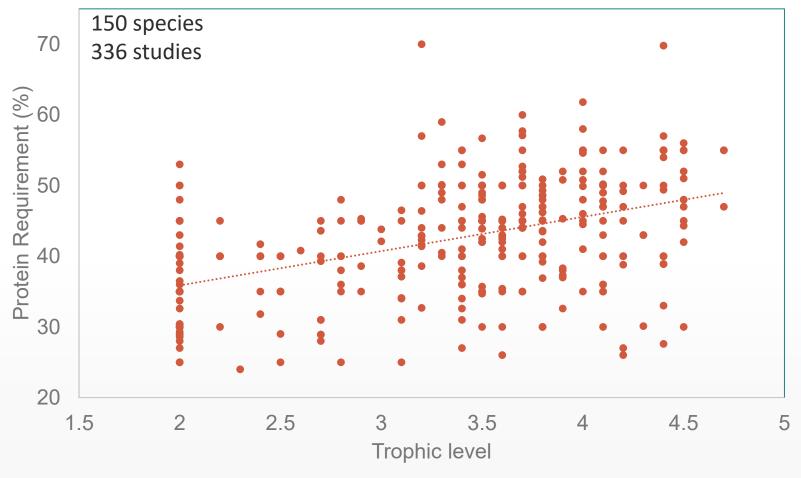
### Fish have high protein requirement

Fish are by far more efficient than traditional farm animals Large differences exist in protein requirements among species

Species	Dietary protein (%)
European sea bass	50
Gilthead sea bream	45
Atlantic salmon	44
Rainbow trout	40
Common carp	38
Nile tilapia	34
Broiler chickens	14-23

NRC 2011

#### Understanding Protein Requirements Based on Trophic Level

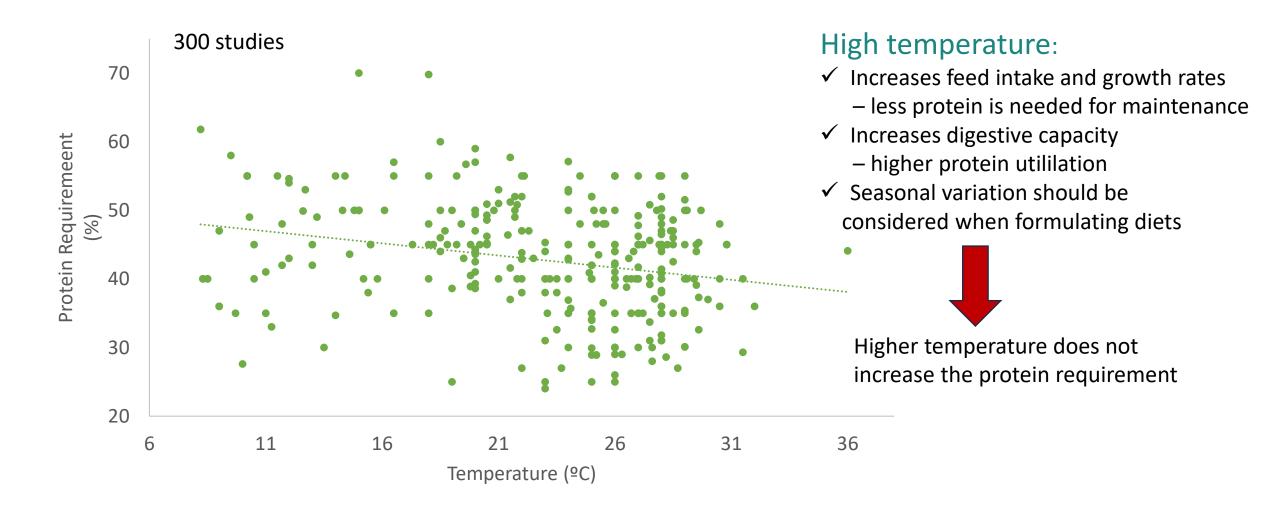


Fish at different tropic level have different protein requirements, this is influenced by:

- 1) Metabolic rates,
- 2) Growth rates,
- 3) Feeding behaviors,
- Adaptation to habitat and ecological conditions

Protein requirements (% diet) & trophic level

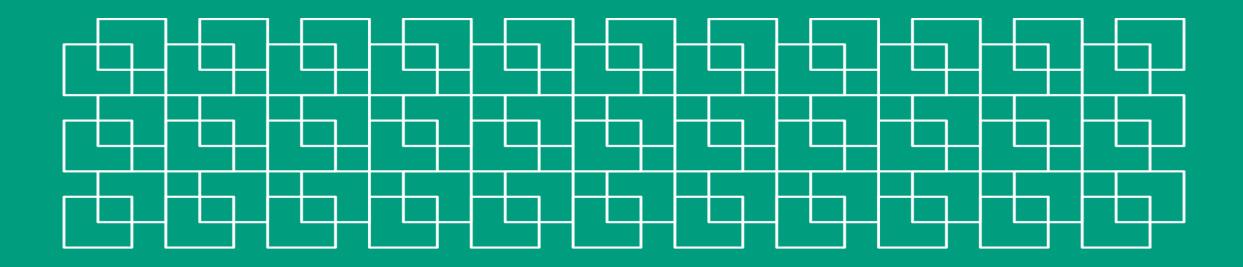
### Effect of Temperature on Protein Requirements in Fish



Source: Oliva-Teles et al 2020

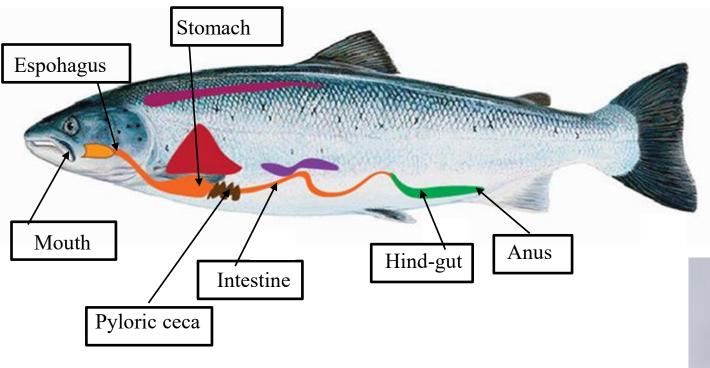


# Protein requirement of the fish





# Gastro-intestinal tract (GIT) of the fish



#### ✓ Gut:body length:

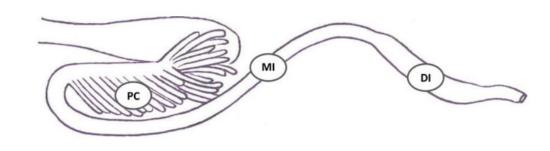
Salmon: 0.8 times its body length

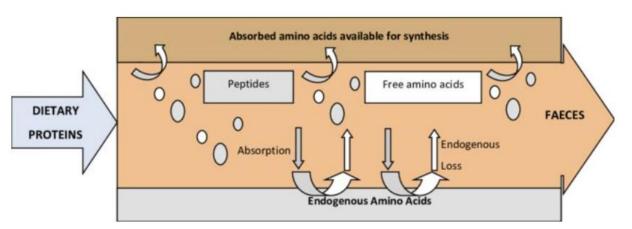
Nile Tilapia: 5-8 times its body length



# Protein Digestion Process in Fish





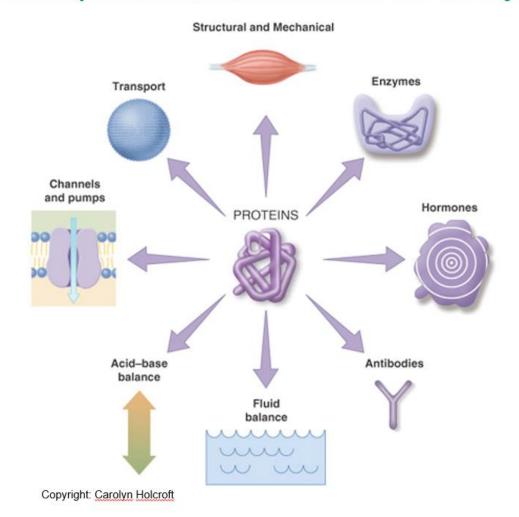


- **1.Stomach (low pH):** Pepsin breaks proteins → smaller peptides
- **2.Pyloric region:** Mixing with bicarbonate, bile acids, digestive enzymes
- **3.Pancreatic enzymes:** Trypsin, chymotrypsin, peptidases break peptides → smaller peptides & amino acids
- **4.Brush border enzymes:** Leucine aminopeptidase and others break peptides → amino acids
- **5.Absorption:** Amino acids and small peptides absorbed via transporters (Na-dependent & Na-independent) into bloodstream
- 3. Amino Acid Absorption at the Brush Border

# Why do fish require protein?



#### Vital protein functions in the body



#### Proteins is required for:

- ✓ Growth & development
- √ Tissue repair
- ✓ Maintaining body tissue
- ✓ Source of dietary glucose and energy
- ✓ Optimal health status
- ✓ Many other vital body functions

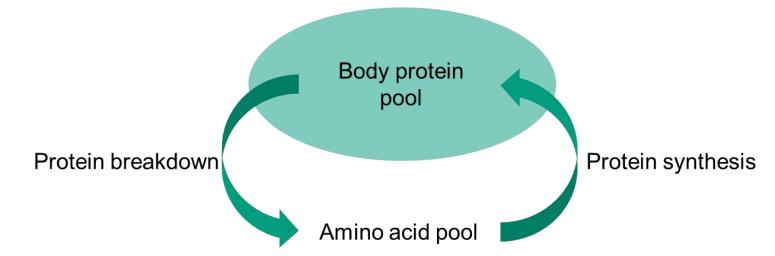
### Protein turnover



After absorbed from the gut, AAs are transported via the portal vein to the liver and enters the <u>AA pool</u> and where they are used for protein synthesis and broken down and reused for other proteins or molecules or <u>deaminated</u> and used as a source of glucose or energy



- Continuous breakdown and
- resynthesis of body protein



# Protein requirement of the fish



Fish **do not require protein to grow**, but they need a mixture of well balanced amino acids, such as essential, semi-essential and non-essential amino acids from the feed.

#### Essential amino acids

- 1. Arginine\*
- 2. Histidine\*
- 3. Isoleucine
- 4. Leucine
- 5. Lysine
- 6. Methionine
- 7. Phenylalanine
- 8. Threonine
- 9. Tryptophan
- 10. Valine

#### Semi essential amino acids

Cystein Methionine

Tyrosine Phenylalanine

#### Non-essential amino acids

- 1. Alanine
- 2. Asparagine
- 3. Aspartic acid (Aspartate)
- 4. Cysteine
- 5. Glutamic acid (Glutamate)
- 6. Glutamine
- 7. Glycine
- 8. Proline
- 9 Serine
- 10. Tyrosine

#### Important physiological function: Ex Arg, Tau, His

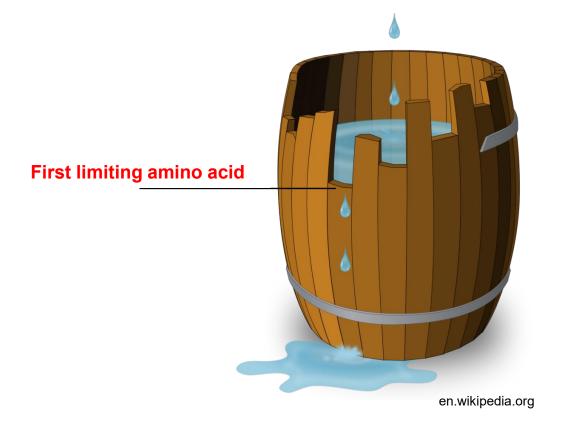
Large biological role: immunity, osmoregulation, antioxidation, cell membrane stability, neurotransmission, vision & many other functions.

# Rain Barrel Concept



To grow, fish need a constant supply of AA from the diet, because protein synthesis will only proceed as long as all the AA are present

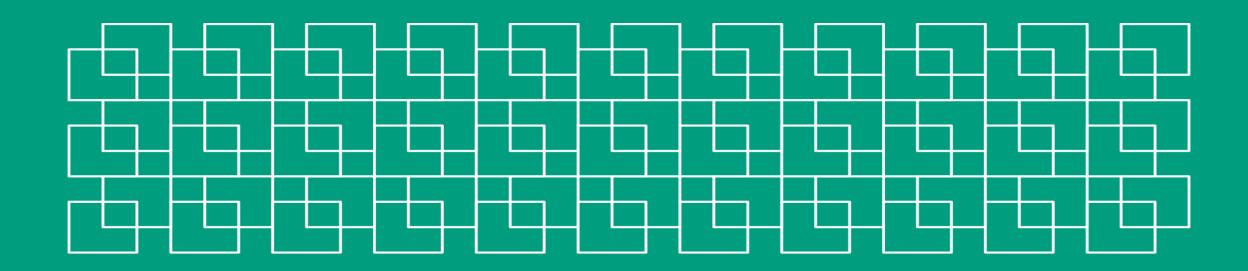
Protein growth requires that the <u>right amino acids</u> are present in <u>the right amount and in the right balance</u> compared to the requirement.



- ➤ The barrel represent the protein growth
- Each stave represent the individual AAs requirement.
- ➤ You can only fill the barrel (growth rate) to the level of the shortest stave, then the water will start to leak
- ➤If <u>one</u> amino acid is limited, the protein synthesis can only proceed to that level and then stop



# Evaluation of protein requirement of the fish



# Dietary amino acid requirement for fish



To formuate the feed we need to know the fish's overall requirement for maintenance and production

Maintenance amino acid (AAs) requirement



Production e.g. growth



The amount required to replace protein & AAs losses:

- ✓ from the gut after a meal
- ✓ from body turnover and oxidation of AAs
- √ from skin & scales
- ✓ Used for synthesis of non-protein products

The amount required to:

- ✓ Support optimal growth
- √ (reproduction)



# Methods to determine maintenance requirement for nitrogen (protein)

#### Maintenance requirement is determined by 2 methods:

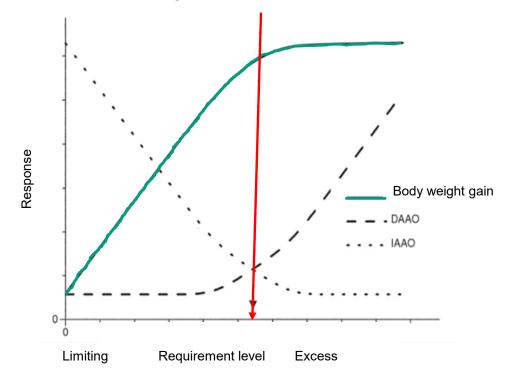
- 1) Direct method measuring N balance during fasting:
  - ✓ by measuring all N-losses in feces, urine, skin and gills (measuring NH<sub>3</sub> and CO<sub>2</sub> in water).
  - the requirement is the amount of N (protein) needed to replace the total losses
- 2) Indirect method by measuring N-retention of fish at end of a dose-response trial when feeding increasing levels of protein.
  - the maintenance requirement is the protein intake at zero weight gain (when the curve flattens out).

# Protein or AA requirement – by emperical method -Dose response studies



#### • Principle:

- Measure growth when feeding graded level of protein or AAs.
- The requirement is the minimum level of AA required for maximum growth, where the curve reaches a plateau



#### Dose response method

#### Basal diet:

- Feeding a balanced diet, deficient in protein or the test amino acid, which is fed below and above requirement
- Requirement is the break-point
- Fine tuned by the broken line model

#### Nitrogen balance studies at optimal growth:

- Indirect method: Slaughter technique total N intake from feed, total weigh gain and changes in N composition of the fish during the experiment
- Direct method: N intake from feed minus N losses in feces, gills and urine at optimal growth

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# Principles of the factorial method



This is an **experimental approach** to estimating protein / AA requirements. The focus is on the **step-by-step determination** of how much of the protein / AAs is required for: **Maintenance, Growth**, & **Other physiological functions** 

#### **Key Points:**

- •Determine maintenance needs in non-growing fish.
- Determine needs for growth in feeding trials.
- •Evaluate the **efficiency** of protein / amino acid utilization.

#### The total requirement:

- 1. sum of AAs needed for all physiological processes
- 2. Include efficiency of AA utilization how well the AAs are converted into body proteins some are lost in the process
- 3. Include additional amounts needed for other functions such as stress.

Total requirement: = sum of AA deposited in the body, AA used for maintenance, catabolic losses and losses in feces.



# Limitation to the factorial method



- ✓ Often results in estimation of one amino acid at the time
- ✓ Costly and time consuming
- ✓ Assume no interaction among amino acids and other nutrients

#### Limited by both biological and dietary factors:

#### Dietary factors

- 1) Composition of the experimental diets
  - Energy density DP:DE ratio
  - Protein source used in reference diet
- 2) Feeding level
- 3) Taste of the diet

#### Biological factors

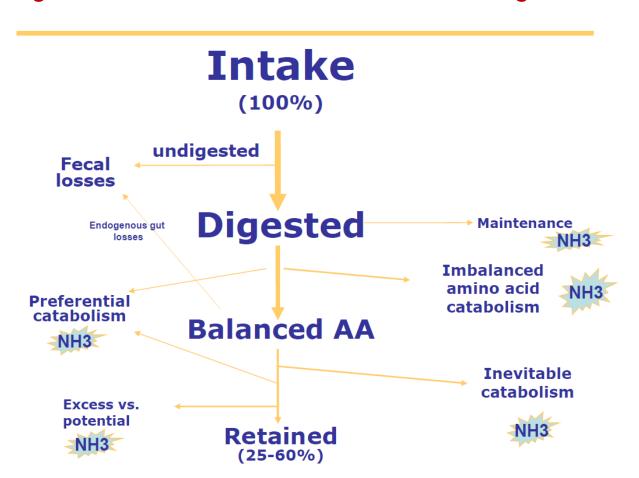
- 1) Weight of fish
- 2) Growth rate of the fish
- 3) Environmental conditions
- 4) Genetic make up
- 5) Stocking density, health status
- Improved model: Takes into account dietary and biological factors that influence EAA utilization
   More precice diet formulations

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# How do we formulate the diets to meet the requirement? Factorial model - Predict how much AAs needs to be **supplied** in the diet to meet net requirements after accounting for losses.



Figure: How amino acid are utilized after ingestion



# Requirement for growth is what is left after accounting for losses

- ✓ The ingested AA are digested,
- ✓ Some are lost in the feces
- ✓ The rest are absorbed
- ✓ Part of the absorbed AA digested is used for maintenance
- ✓ Part will be catabolized
- ✓ The rest will be used for growth



# Ideal protein

Amino acid requirement is a mirror immage of the amino acid pattern in the fish muscle

The requirement of each amino acid is expressed in relation to each other, where lysin is used as the basis

Once the lysine requirment is known, the amino acid requirment is calculated by expressing these in relation to lysine, which is set to 100

Ideal protein concept is also used to evaluate the nutritional value of a protein ingredients

#### Advantages

- Useful when the AA requirement are only partially known.
- Allows feeding a balanced diet that prevents over- or under-feeding of AA
- Maximizes utilization of protein & minimizes N-excretion
- Minimizes the amount of AA/CP needed in the diets, which reduces production costs & increases profitability.



# Ideal protein composition of salmonids.

The ideal AA composition in Atlantic salmon and rainbow trout based on whole body composition, g/100 g.

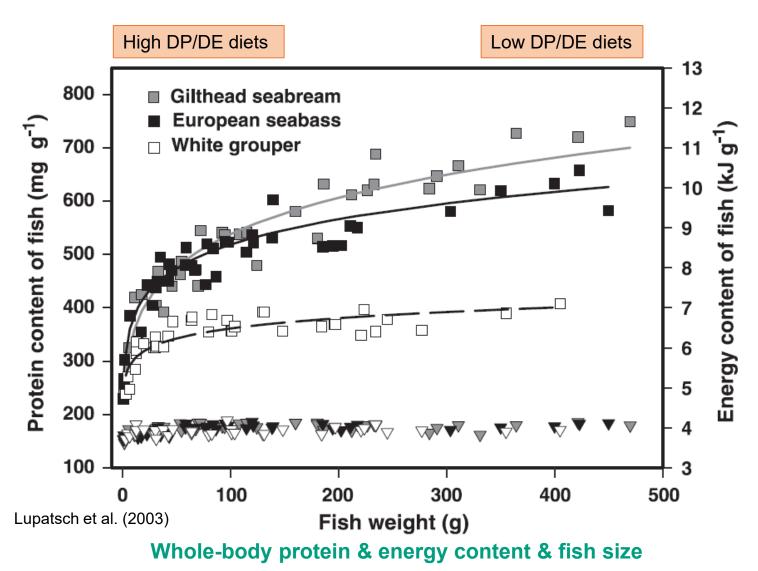
Amino acid	Atlantic salmon <sup>1</sup>	Rainbow trout	4
Ala	6.52	6.57	
Arg	6.61	6.41	
Asp	9.92	9.94	
Cys	0.95	0.80	
Glu	14.31	14.22	
Gly	7.41	7.76	
His	3.02	2.96	
lle	4.41	4.34	
Leu	7.72	7.59	
Lys	9.28	8.49	= 100%
Met	1.83	2.88	
Phe	4.36	4.38	
Pro	4.64	4.89	
Ser	4.61	4.66	
Thr	4.95	4.76	
Try	0.93	0.93	
Tyr	3.50	3.38	
Val	5.09	5.09	

The amino acid requirement of fish is given relative to lysine (100%)

Source: Lall and Andresen 2005

# Relationship between protein and energy content and fish size v.s. dietary DP:DE ratio



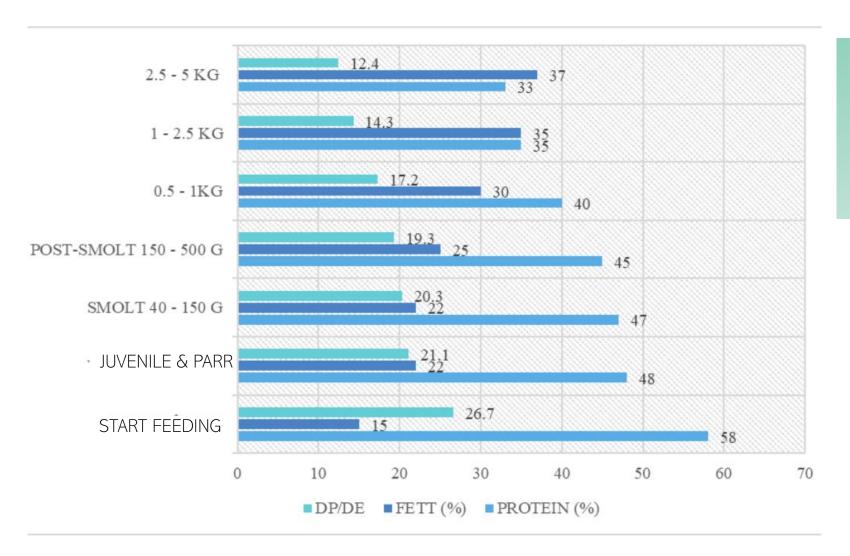


# Dietary DP/DE decreases with fish weight due to:

- ✓ Smaller fish have higher growth rates and require more protein for tissue development and growth.
- ✓ The metabolic rate decreases as fish grow.
- ✓ Larger fish have higher capacity to utilize non-protein energy sources from fat and carbohydrates, leading to a lower DP/DE ratio in their diet.
- ✓ The basis to determine the DP:DE ratio of the feed is to know the protein & energy content & digestibility of the raw materials



## Dietary DP:DE-ratio in salmon at different life stages

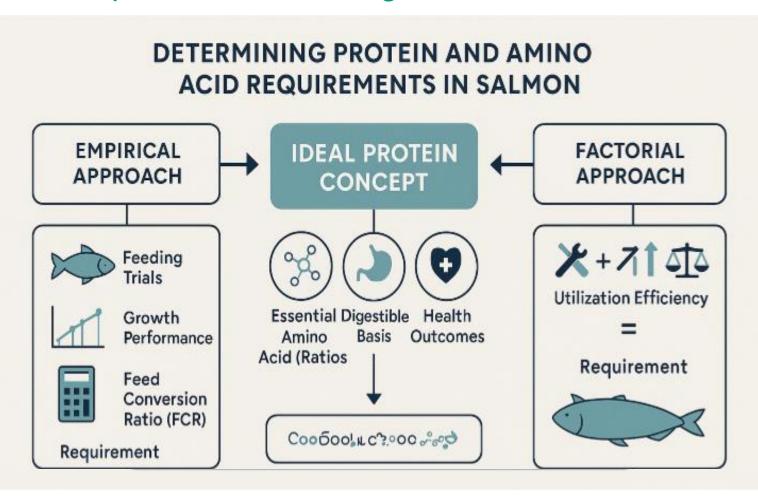


#### The DP:DE ratio for salmonids:

Juvenile salmon: 26,7 g DP/MJ DE Larger salmon: 12.4 g DP/MJ DE

# How to formulate the diets to meet the requirements

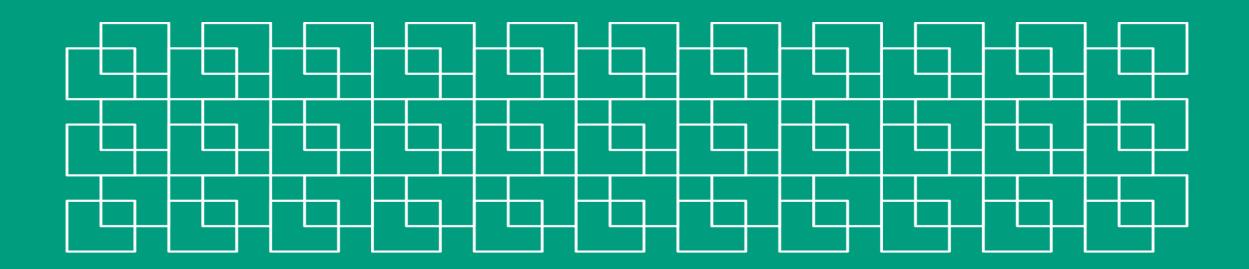
Principles for Determining Protein and Amino Acid Requirements



- 1. Know the amino acid requirement Expressed as mg of digestible AA / kg of body weigh / day
- 2. Formulate diets taking into account:
  - content of digestible protein and AAs,
  - an optimal DP:DE ratio &
  - the ideal protein concept
- 3. To calculate the concentration of the AA to meet the requirement in the diet, requires knowledge on the feed intake of the fish, expressed as %/day
- 4. The diet is then formulated based on the expected amount of feed eaten per day



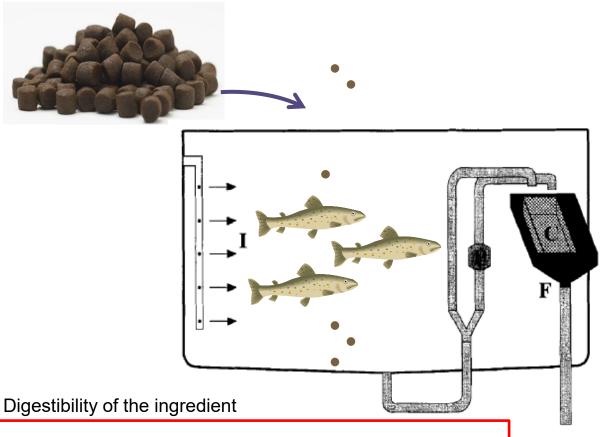
# Evaluation of protein quality in the feed ingredients / diets



# Methods used to determine protein digestibility



• Feeding a reference diet and a test diet where 30% of the test ingredient is mixed with the reference diet & use of an intert marker (e.g Y<sub>2</sub>O<sub>3</sub>)



## **Fecal collection methods**

- ✓ Fecal stripping
- ✓ Setteling column
- ✓ Atomatic collection by screen rotation
- ✓ Dissection

Sources: Shomorin et al., 2019, Aquaculture , 504; 81-87



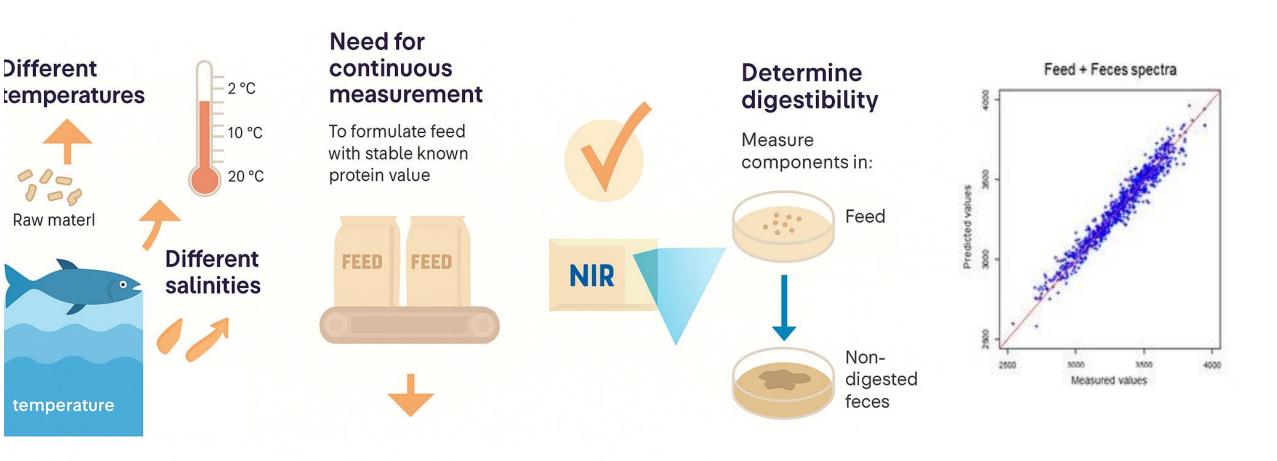
 $ADC_{ingredients}$  (%) =  $ADC_{testdiet} + (ADC_{testdiet} - ADC_{refdiet})$  $\times \left(\frac{0.7 \times Nutrient_{refdiet}}{0.3 \times Nutrient_{testingr}}\right)$ 

Nutrient in feces
Nutrient in feed

# Digestibility varies

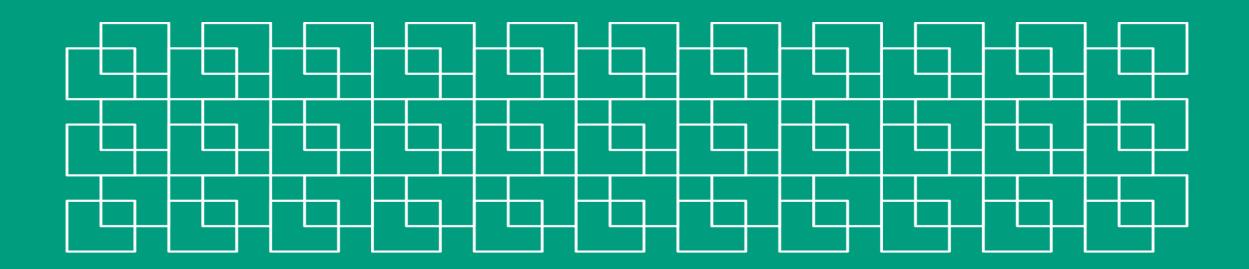


Between raw material, from batch to batch and with water temperatures and salinity





# Nutritional value of common protein ingredients in fish nutrition



# Amino acid composition of common protein feedstuffs, g/100 g protein



	Fish meal <sup>1</sup>	Soy protein concentrate <sup>2</sup>	Pea protein concentrate <sup>3</sup>	Canola protein concentrate <sup>2</sup>	Wheat gluten¹
Crude protein	74,9	70,2	49,6	59,5	89,3
Arginine	5,27	6,41	8,74	5,11	3,26
Histidine	1,87	2,5	2,65	2,42	1,74
Isoleucine	3,69	4,14	4,32	3,62	3,37
Leucine	6,26	6,64	7,21	6,18	6,49
Lysine	6,92	5,53	7,07	4,38	1,57
Methionine	2,42	1,18	0,9	1,7	1,92
Phenylalanine	3,37	4,45	4,73	3,57	5,69
Threonine	3,65	3,52	3,73	3,67	2,54
Tryprophan	0,73	1,14	0,99	1,16	0,76
Valine	4,03	4,06	4,66	4,29	3,71
Total EAA	38,2	39,6	45,0	36,1	31,0

# Digestibility of common protein sources for salmoninds

# Digestibility of protein and amino acid for salmoninds in selected

common protein sources, %

	Fish meal	Soy protein concentrate	Wheat gluten	Corn gluten	Rapeseed meal	Pea protein		Mycelium <sup>2</sup>	1
Crudo protoin			100				┪		┢
Crude protein	89	96	100	92	75	99	-	86.5	₩
Lysine	95	97	95	91	88	100		93.8	
Methionine	97	97	97	92	87	100		91.5	
Threonine	94	93	94	93	90	99		88.4	ı
Isoleucine	97	96	92	91	85	98		90.5	l
Leucine	99	97	97	97	92	97		91.1	ı
Valine	95	95	94	94	87	98		89.7	l
Histidine	94	96	95	96	96	93		92.0	ı
Phenylalanine	93	95	97	95	90	93		-	
Tyrosine	98	99	96	95	90	98		-	

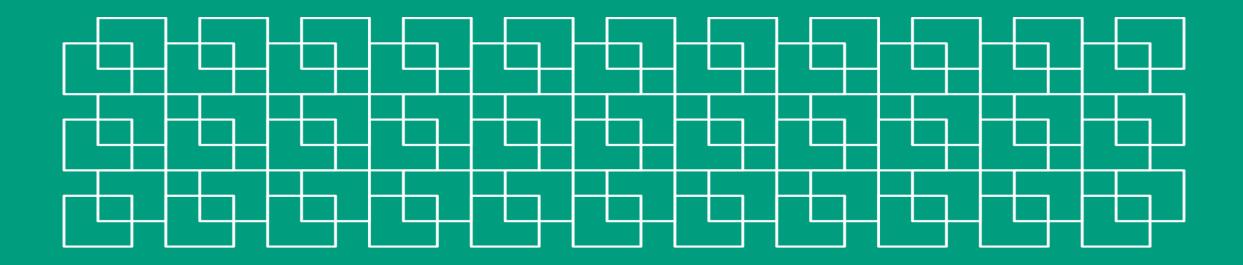
#### Source:

<sup>&</sup>lt;sup>1</sup>After **Barrows et al., 2011**. Database of Nutrient Digestibility's of Traditional and Novel Feed Ingredients for Trout

<sup>&</sup>lt;sup>2</sup>Gaudhaman et al., 2025. Fungal Protein from Non-Food Bioresources in Diets for Rainbow Trout (Oncorhynchus mykiss). Fishes



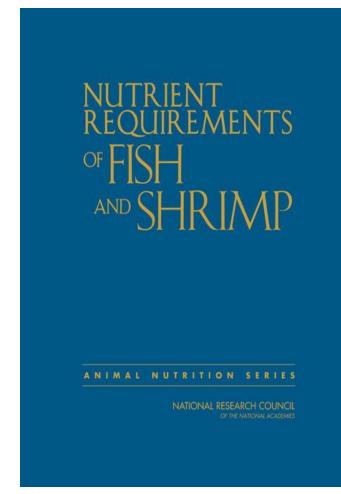
# Protein requirements of fish

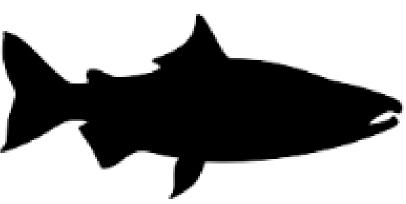


# Atlantic salmon



National Research Council - NRC, 2011:





#### Limitations

- AA requirement for Salmon is not well known.
- Old and not relevant for modern genotypes
- Mainly performed on juveniles
- Established based on LT-fish meal diets, less relevant for plant-based diets
- AA requirement is often based on growth performance, don't take into account other metabolic needs.

# Major feed companies have developed their own company-specific database



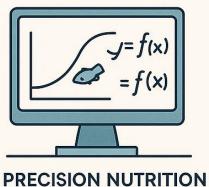
#### **FORMULATING DIETS**



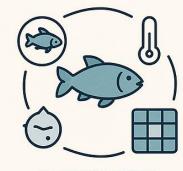
**NUTRITIONAL DATABASES** 



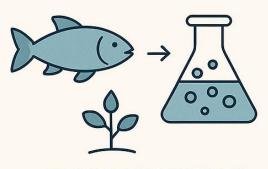
RESEARCH INSTITUTES



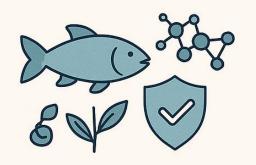
PRECISION NUTRITION MODELS



**CUSTOMIZED** FORMULATIONS



INGREDIENT-SPECIFIC TRIALS

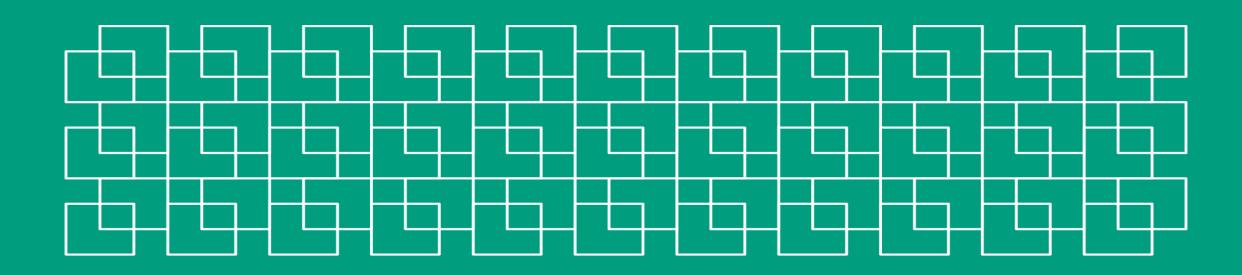


FUNCTIONAL FEEDS AND NUTRITIONAL PROGRAMMING

- 1. Nutrient requirement
  Extensive in-house trials.
  Field trials
  Published scientific literature,
  Precision Nutrition Models
- 2. Regulatory and Market Requirements



# Choice of feed raw materials to meet the protein requirement & future perspectives

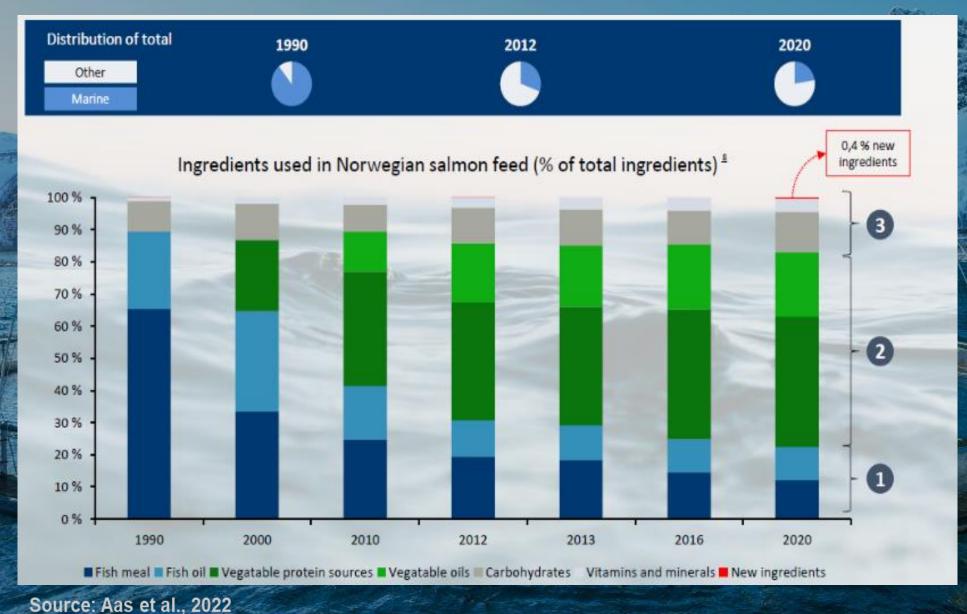


# Fish meal and fish oil





# Changes in the salmon feed composition



# From marine to land – the new salmon feed Common plant-ingredients in fish feed













#### Soy

- 45% crude protein
- 4-6% fiber
- Oligosaccharides
- Protease inhibitors
- Saponins,
- Phytic acid
- Phytoestrogens

#### Rapeseed

- 35% crude protein
- High in fiber
- Glucosinolates
- Sinapine, tannins
- Phytic acid

#### Sunflower

- 38-40% crude protein in dehulled meal
- 15-20% fiber
- Low ANFs & no trypsin inhibitors

#### Peas

- 25% crude protein
- High in starch
- Low level of ANF

#### Field beans

- 30% crude protein
- Fiber-rich hull fraction
- Low level of ANF

Other sources: Wheat and maize gluten (60% crude protein), Plant by-products: brewer and distiller grains

#### **FOODS?NORWAY**





# Plant ingredients

Plant ingredients are abundant and cheap and relieve pressure on wild catch for fish meal production

#### **Strengths**

- Available in large quantitie at a low cost
- Large potential to improve nutritional value through targeted processing
- New advances in fermentation technology provide new opportunities

#### **Barriers**

- Low nutrient density / high fiber content
- Unbalanced amino acids composition
- Low palatability
- Contain a wide range of anti-nutritional factors
- Competition for food, feeds and non-food products such as biofuels
- Increased pressure on land and water resources
- Increased use of pesticides & herbicides
- Social and environmental sustainability



# Norwegian governments' sustainable feed mission

We need to produce enough feed, sustainably, to meet the demands of aquaculture

## Sustainable and cost-effective feeds are essential for advancing aquaculture in the future



92% of the salmon feed raw materials is imported.



Feed accounts for 75-83% of greenhouse gas emissions in salmon production (before transport to market).



Political goals: By 2034, all feed should come from sustainable sources, and preferably produced locally.

Need to adapt a more circular feed system.

#### **FOODS?NORWAY**

# Several promising sources of future feed raw material

Future ingredients can be categorized into harvested novel marine and plant-based ingredients, farmed organisms and underutilized resources

#### **Harvested resources**



#### Farmed organisms



## Underutilized resources





## Novel marine ingredients

- Mesopelagic fish
- Calanus finmarchius
- Krill

#### Plant byproducts\*\*

 Fermented soybean meal/rape seed meal

#### Insects

- Black soldier fly
- Mealworms

### Marine low-trophic species

- Blue mussels\*\*\*
- Polychaeta
- Gammaridae
- Tunicate
- Seaweed

## Microbial ingredients

- Bacteria
- Yeast
- Fungus
- Heterotrophic microalgae

#### Land animal byproducts\*\*\*\*\*

- Poultry
- Pork

**Photoautotrophic** 

microalgae\*\*\*\*

#### Marine byproducts

- Whitefish
- Pelagic fish
- Aquaculture\*

Rethinking on how we feed the fish





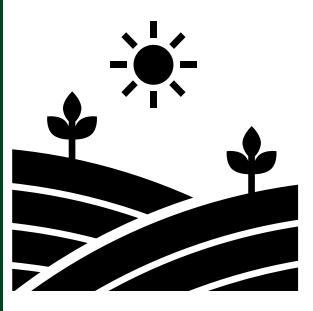
# Sustainable feed is the key to future growth

Shift from linear to circular systems

"Circular feeds" a holistic approach to produce feed that prioritizes sustainability, resource efficiency, and environmental responsibility.

# Shift from Linear to Circular Systems

Optimizing resource use, minimizing waste, and promoting sustainability along the feed value chain

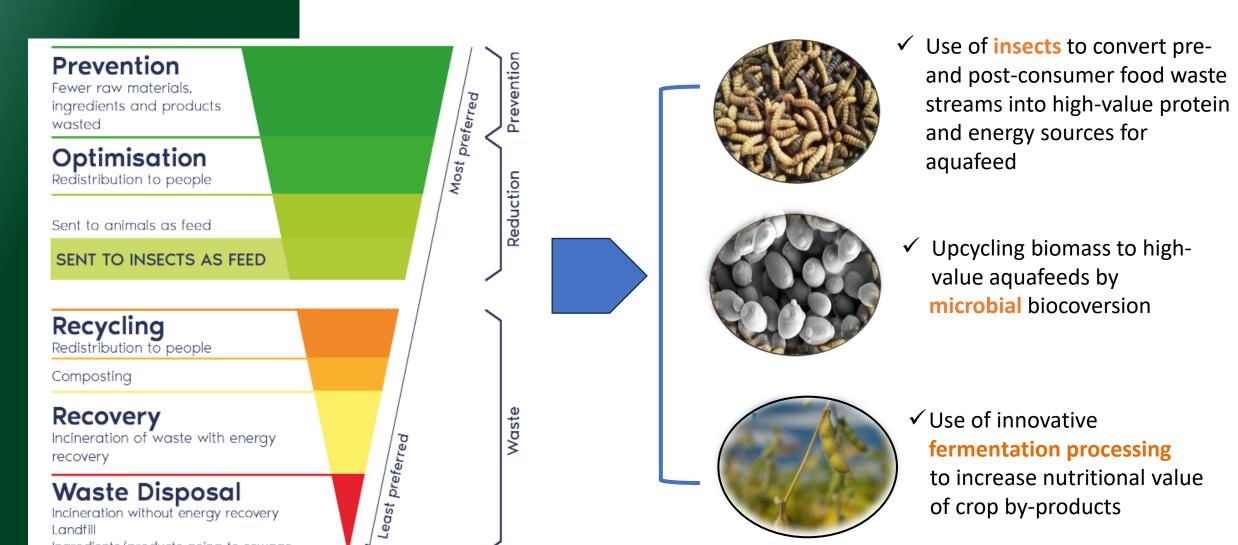


- ✓ Facilitate circular economy by adopting nutrient recycling
- ✓ Use side-streams and low value biomass to maximize circularity

# Moving towards a circular food system

Insects & microbes can add a new layer to the waste hierarchy

Norwegian University of Life Sciences



Landfill

Ingredients/products going to sewage

# From Forest to Fjord: Rethinking Fish Feed

**FOODS**?NORWAY



# Yeast fermentation on small scale at NMBU





# Microbial ingredients in diets for salmon

- 1. Agboola, J.O et al., 2023. Animal Microbiome. <a href="https://doi.org/10.21203/rs.3.rs-1351266/v1">https://doi.org/10.21203/rs.3.rs-1351266/v1</a>.
- 2. Agboola, J. O et al., 2022. Aquaculture 546, 737312
- 3. Agboola, J. O et al., 2021. Scientific Reports 11(4496). DOI: 10.1038/s41598-021-83764-2
- 4. Agboola, J.O et al., 2022. International journal of molecular sciences, 23(3), 1675. https://doi.org/10.3390/ijms23031675
- 5.Grammes, F. et al., 2013. *PLoS ONE* 2013; Volum 8.(12).
- 6. Hansen J.Ø et al., 2019. Aquaculture 511: 1–10. doi.org/10.1016/j.aquaculture.2019.734239.
- 7. Hooft J., et al., 2024/2025. Aquaculture, Aquaculture 589, 740905; Aquaculture 596, 741779
- 8. Morales-Lange, B et al., 2021. Frontiers in immunology, doi: 10.3389/fimmu.2021.708747
- 9. Sahlmann, C et al., 2019. Aquaculture 513:734396 DOI: 10.1016/j.aquaculture.2019.734396
- 10. Øverland, M et al., 2013. Aquaculture, 402–403, 1–7.

# Foods of Norway achieves large-scale production of yeast from trees













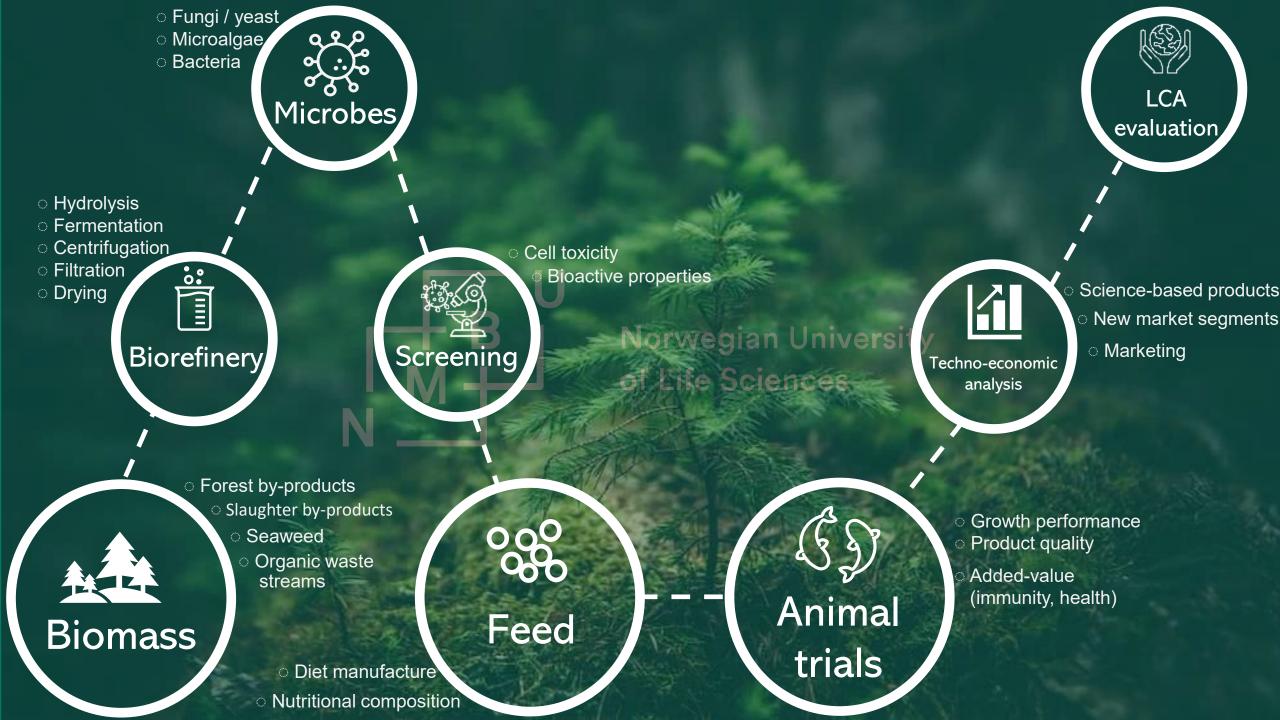












# N@RDIC FEED







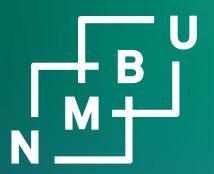








Foods of Norway 2.0 feeds a knowledge platform for sustainable feed solutions



Norges miljø- og biovitenskapelige universitet