

# Production of Carotenoids from Aquaculture Sidestreams as Feedstock Using the Bacterium *Corynebacterium glutamicum*

Ina Schmitt<sup>1</sup>, Florian Meyer<sup>1</sup>, Nadja A. Henke<sup>1</sup>, Sinem Zeytin<sup>2</sup>, Hilke Alberts-Hubatsch<sup>2</sup>, Katerina Kousoulaki<sup>3</sup>, Volker F. Wendisch<sup>1</sup>, Petra Peters-Wendisch<sup>1</sup>

<sup>1</sup> Genetics of Prokaryotes, Faculty of Biology and Center for Biotechnology, Bielefeld University, Germany

<sup>2</sup> Alfred Wegener Institute Helmholtz Center for Polar and Marine Research, Germany

<sup>3</sup> Department of Nutrition and Feed Technology, Nofima, Nofima, Norway

## Contact

Petra Peters-Wendisch  
petra.peters-wendisch@uni-bielefeld.de

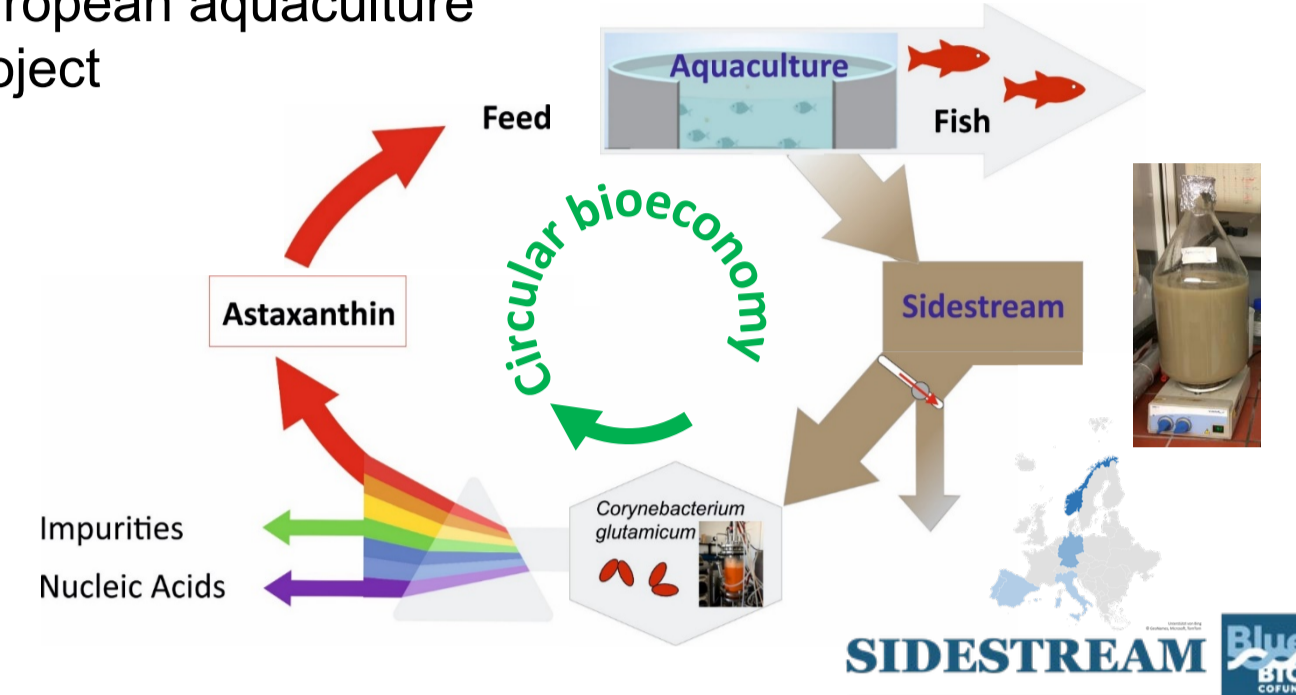


## Introduction

Aquaculture is one of the fastest-growing food production systems, enabling to meet the rising consumer demand for healthy seafood. In order to establish a flexible feedstock concept using aquaculture sidestream<sup>1</sup>, *Corynebacterium glutamicum* that naturally synthesizes the rare yellow C50 carotenoid decaprenoxanthin was metabolically engineered to produce a variety of different carotenoids including the fish feed ingredient astaxanthin<sup>2,3</sup>. **Preprocessed aquaculture sidestream (AQ)** was tested for growth and production of these carotenoids. Notably, astaxanthin production was enhanced by addition of AQ. The process could be scaled up to 2L bioreactor fermentation operated in batch or fed-batch mode<sup>4</sup>. In this project, a step towards a circular economy in aquaculture by using thus produced astaxanthin as fish feed could be demonstrated in a first trout feed trial.

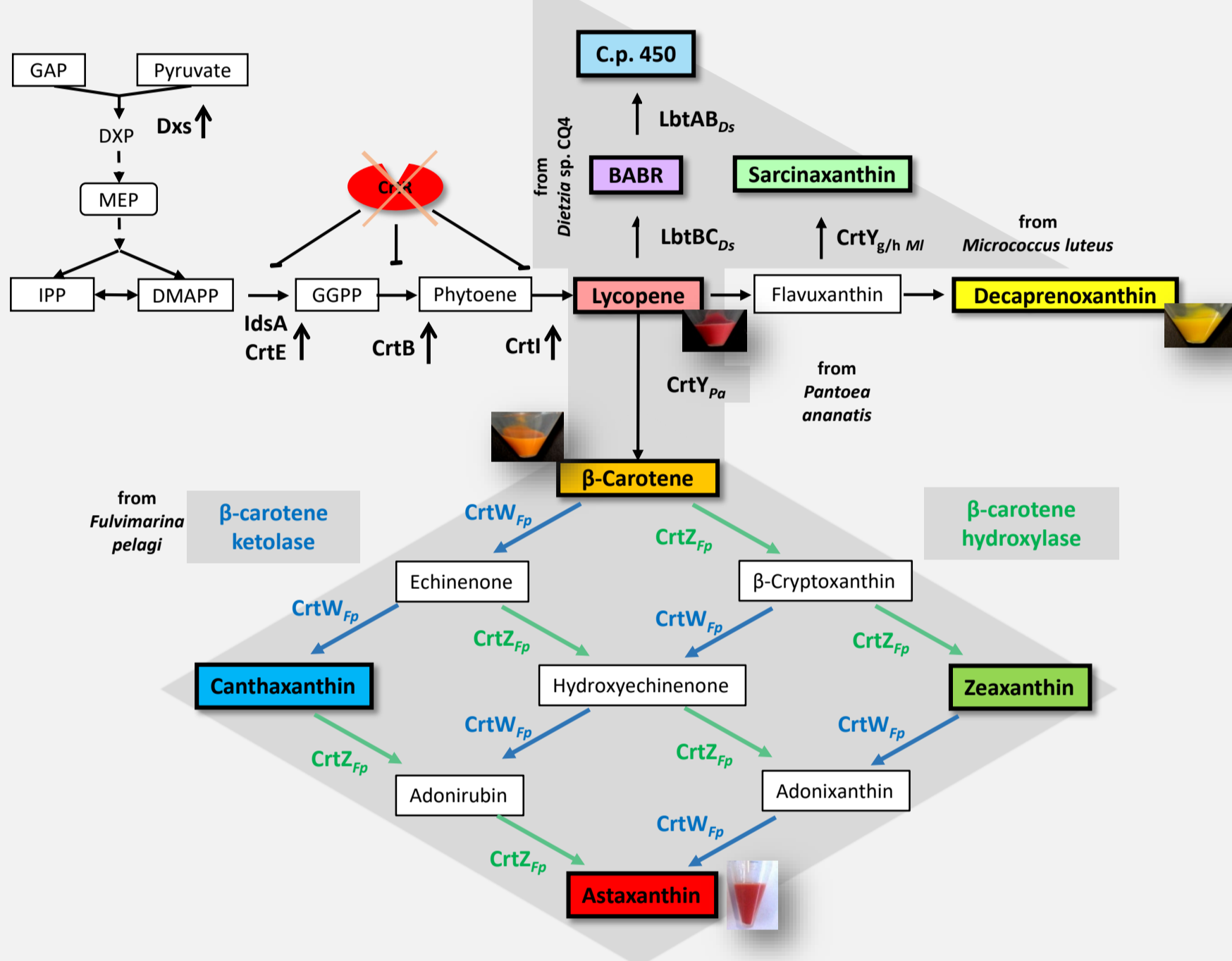
## Summary

European aquaculture project



**SIDESTREAM** BlueBio

## 1. Metabolic engineering of strains for carotenoid production

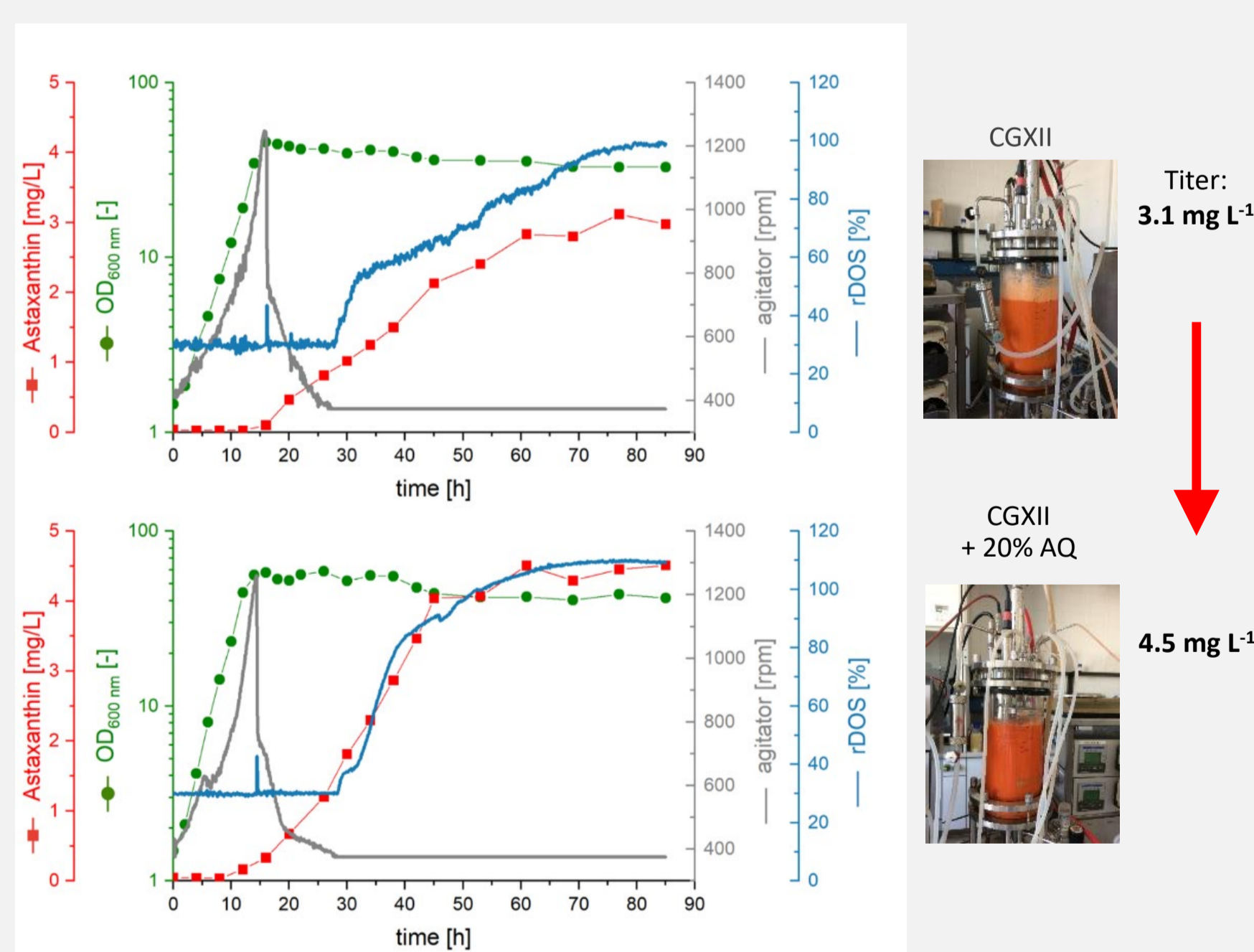


## 2. AQ for production of different carotenoids

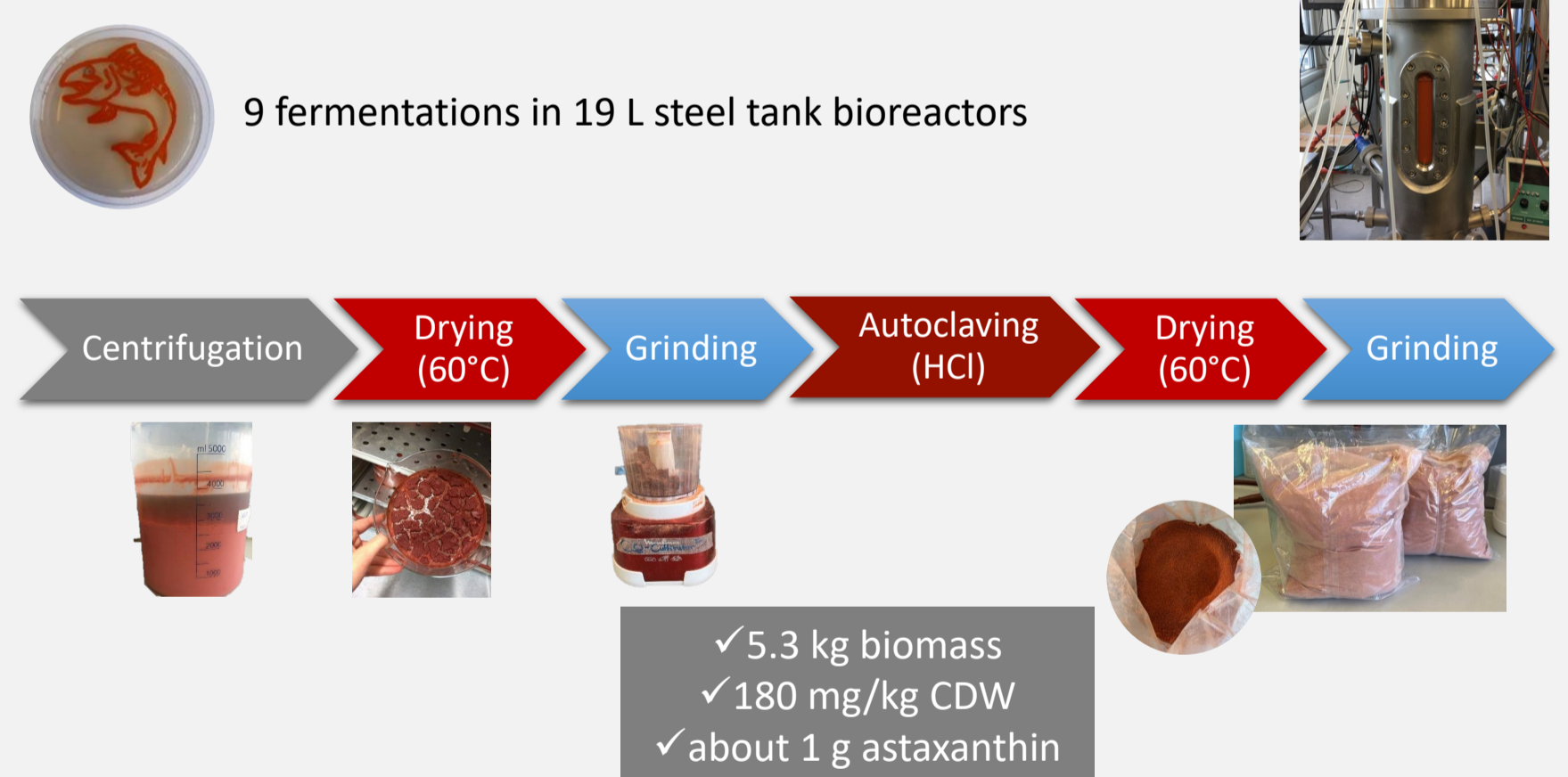
| Strain      | Medium                          | Carotenoid [mg/L] | Carotenoid       |
|-------------|---------------------------------|-------------------|------------------|
| MB001 ΔcrtR | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~100              | Decaprenoxanthin |
| LYC6        | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~10               | Lycopene         |
| BABR1       | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~15               | BABR             |
| CP1         | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~25               | C.p.450          |
| SAX1        | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~10               | Sarcinaxanthin   |
| BETA4       | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~100              | β-carotene       |
| ZEA5        | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~1                | Zeaxanthin       |
| CAN5        | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~5                | Canthaxanthin    |
| ASTA*       | CGXII<br>CGXII + 20% AQ<br>CGAQ | ~10               | Astaxanthin      |

✓ AQ supported carotenoid production by various *C. glutamicum* strains

## 3. Addition of AQ improved astaxanthin production in bioreactor cultivation



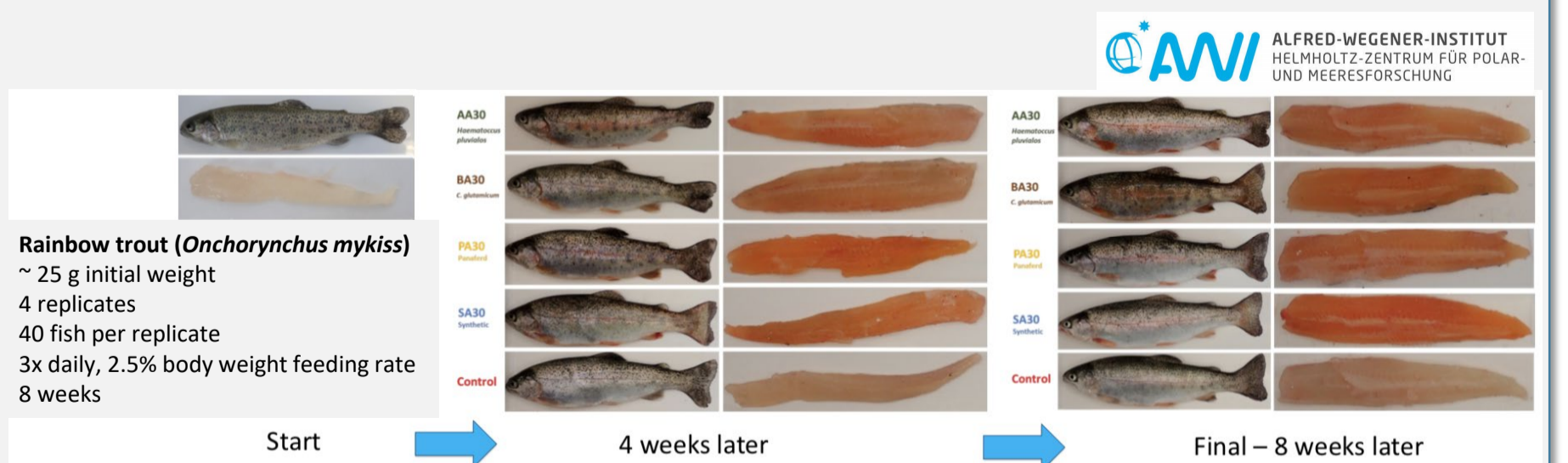
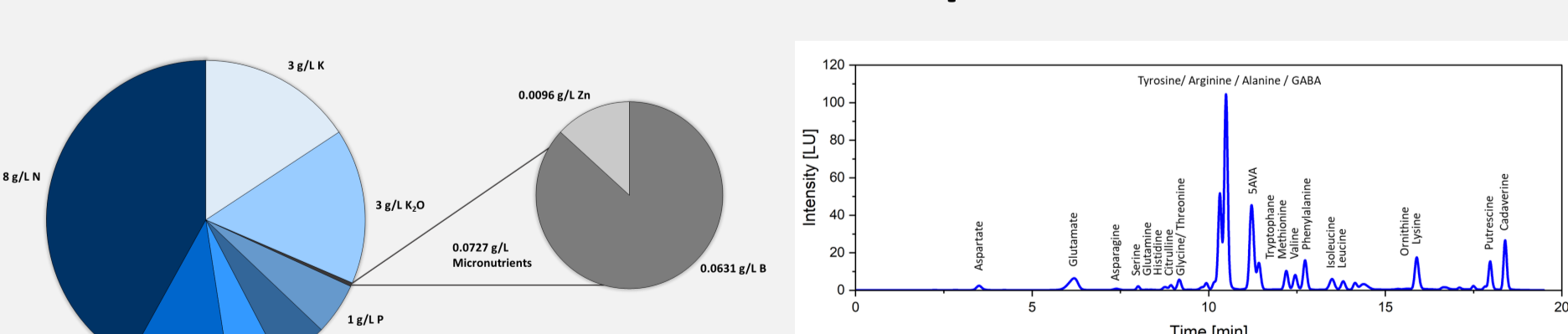
## 4. Astaxanthin production for trout feed trial



### Astaxanthin feed formulation (30 mg/kg feed)

- Nofima
- Haematococcus pluvialis biomass (esterified)
- C. glutamicum* biomass
- Paracoccus carotinifaciens* (Panaferd®)
- synthetic astaxanthin
- controls without astaxanthin

## 5. AQ nutrient and amino acid composition



✓ Comparable trout filet pigmentation with *C. glutamicum* biomass

## References

- Wendisch VF, Nampoothiri KM, Lee J-H (2022) *Frontiers in Microbiology* 13: 835131
- Cankar K, Henke NA, Wendisch VF (2023) *Systems Microbiology and Biomanufacturing* 3: 110-121
- Henke NA, Göttl V, Schmitt I, Peters-Wendisch P, Wendisch VF (2022) *Methods in Enzymology*, 671, 383-419.
- Schmitt I, Meyer F, Krahn I, Henke NA, Peters-Wendisch P, Wendisch VF (2023) *Molecules* 28(4):1996.
- Zeytin S, Alberts-Hubatsch H, Kousoulaki K, Schmitt I, Meyer F, Peters-Wendisch P, Henke NA, Wendisch VF, Slater MJ (2022) Aquaculture Europe Conference Abstract Book.

This research was supported by the ERA-NET BlueBio COFUND Project SIDESTREAM [Grant ID 68].