



Variability in nutritional value and safety of *Arthrospira* and *Chlorella* biomass necessitates smart production of microalgae for human spaceflight

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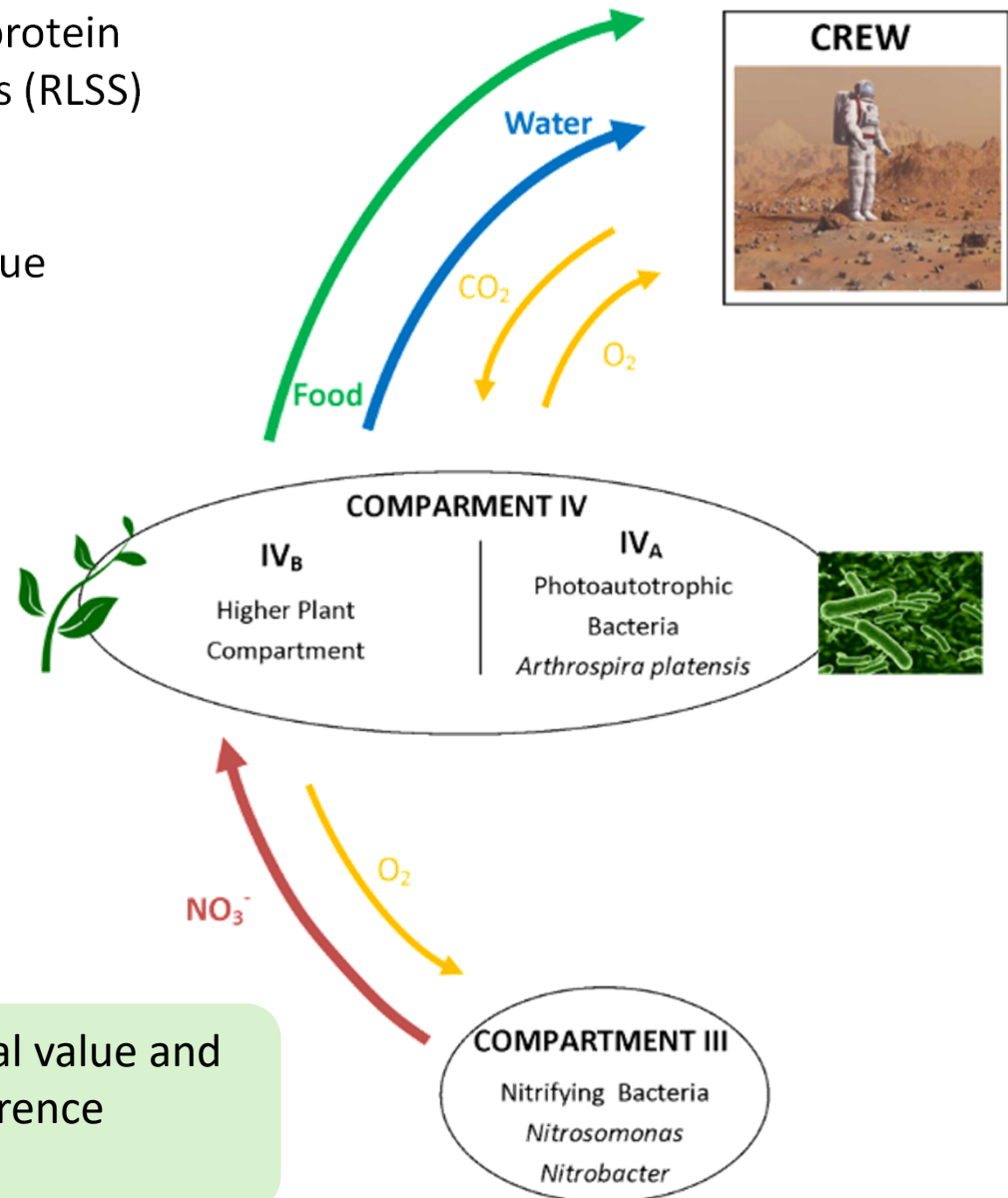
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Microalgae as protein source in RLSS

Arthrospira (Spirulina) and *Chlorella*: key protein source in regenerative life support systems (RLSS)

E.g. MELiSSA concept (ESA)

Arthrospira upgrades nutrients to high-value dietary protein and produces oxygen



GOAL: Biomass production with optimal **nutritional quality** and **safety**!

BUT: Magnitude of variation in nutritional value and safety is largely unknown, as is the preference towards *Arthrospira* or *Chlorella*

Spatial and temporal variability

Research goal:

- Quantification of spatial and temporal variation in nutritional quality and safety of microalgal biomass
- Indication on which common microalgae is favorable in terms of price-quality

Screening of:

Spatial variability = from all over the world

Temporal variability = same brand/different batch

7 commercially available *Chlorella*



5 commercially available *Chlorella*:



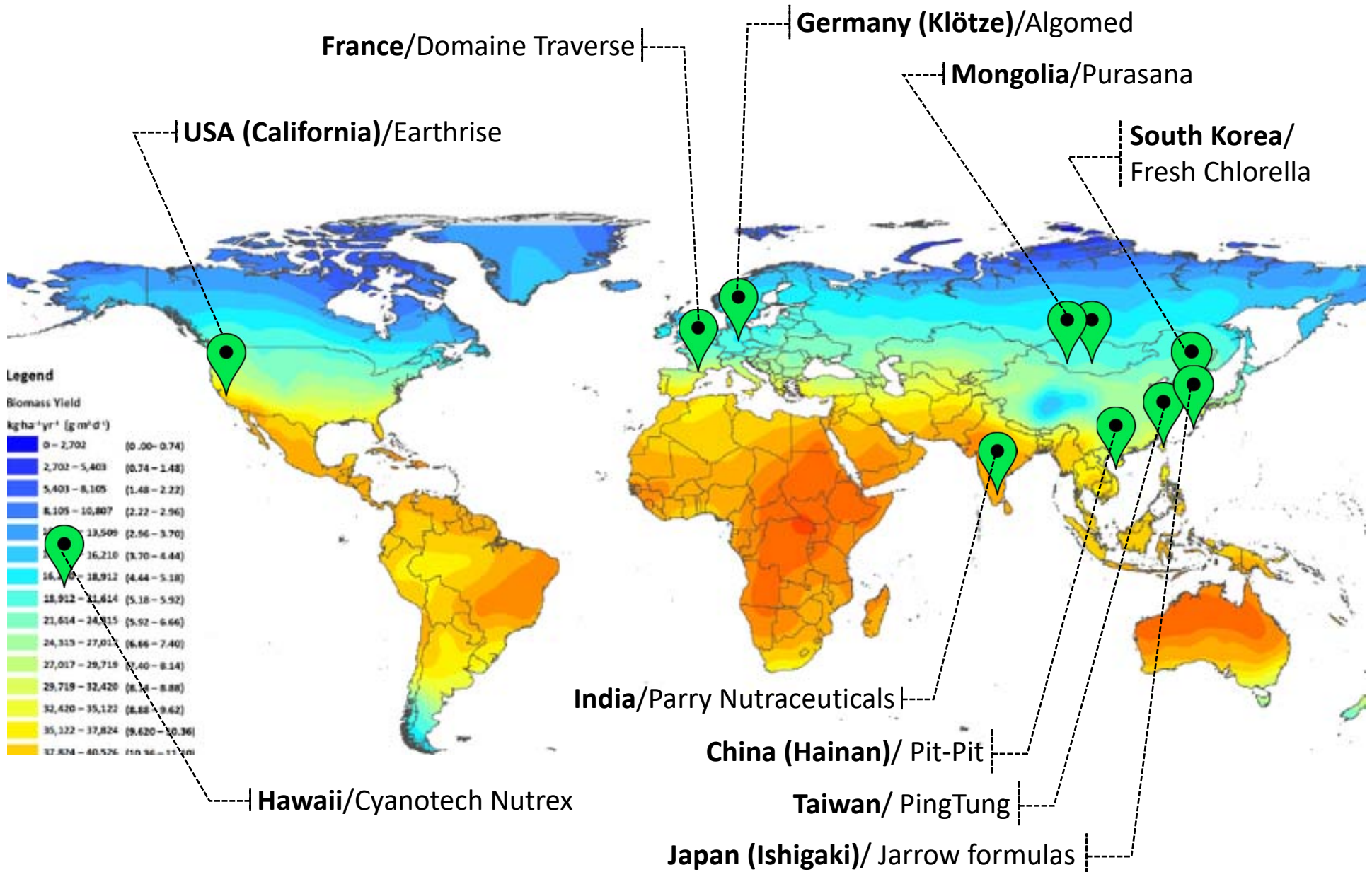
7 commercially available *Spirulina*



5 commercially available *Spirulina*:

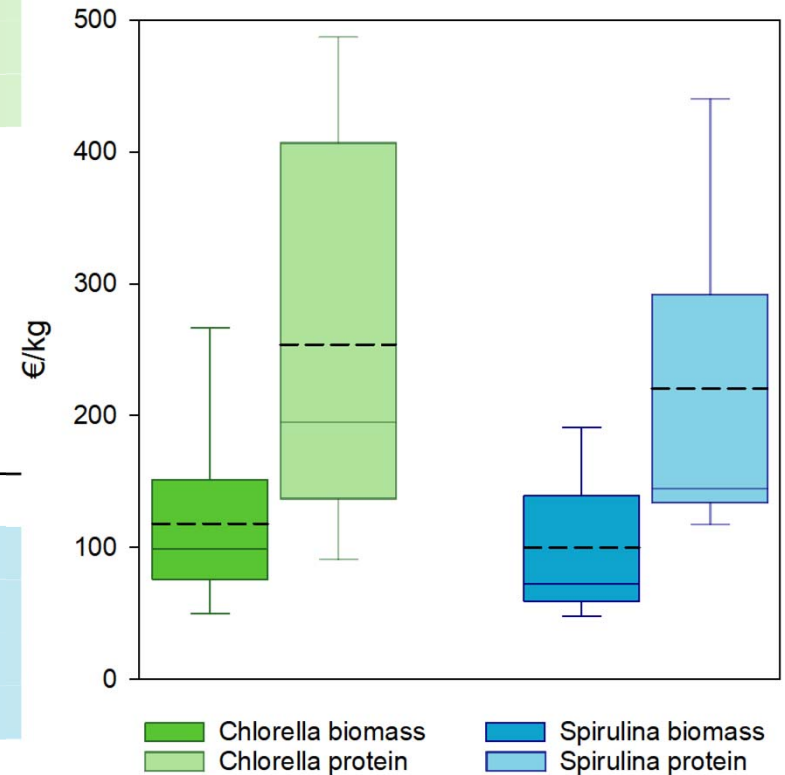


Origin of the microalgal samples



Samples

	Code	Brand	Country of origin	Retailer	Exp. date
Chlorella	C1	Purasana	Mongolia	Origin'O	31/05/18
	C1a	Purasana	Mongolia	Origin'O	31/05/18
	C1b	Purasana	Mongolia	Origin'O	31/05/18
	C1c	Purasana	Mongolia	Bioplanet	30/03/19
	C1d	Purasana	Mongolia	Bioplanet	30/04/19
	C2	Febico	Taiwan Ping-Tung	Febico	04/03/19
	C3	NA	China Hainan	pit-pit	16/09/18
	C4	Jarrow formulas	Japan Ishigaki Island	Jarrow	07/17
	C5	Clean chlorella	South Korea	Health Ranger Select	NA
	C6	Algomed	Germany Klötze/Altmark	Algomed	14/12/18
	C7	Iswari	China Hainan	Iswari	03/17
Spirulina	S1	Purasana	Mongolia	Origin'O	31/05/18
	S1a	Purasana	Mongolia	Origin'O	30/04/19
	S1b	Purasana	Mongolia	Bioplanet	30/04/19
	S1c	Purasana	Mongolia	Bioplanet	30/04/19
	S1d	Purasana	Mongolia	Origin'O	30/03/19
	S2	Febico	Taiwan Ping-Tung	Febico	14/01/19
	S3	NA	China Hainan	pit-pit	11/09/18
	S4	Now foods	India Parry Nutraceutical	Now	11/17
	S5	Nutrex Hawaii	Hawaii cyanotech nutrex	Nutrex	01/19
	S6	Earthrise	USA California	Earthrise	01/19
S7	Domaine traverse*	France Toulon	NA	NA	

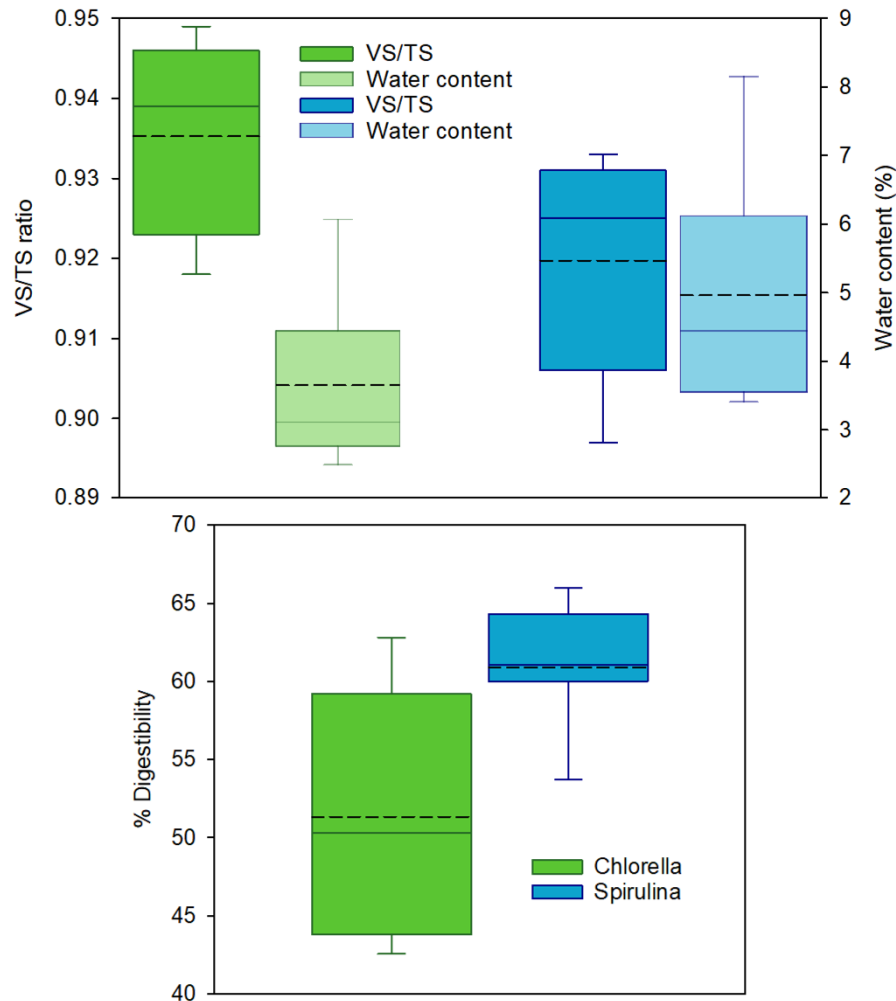


- Huge difference in price for 'the same' product
- Chlorella biomass on average 18% more expensive than Spirulina biomass
- Per kg protein, Chlorella is 15% more expensive

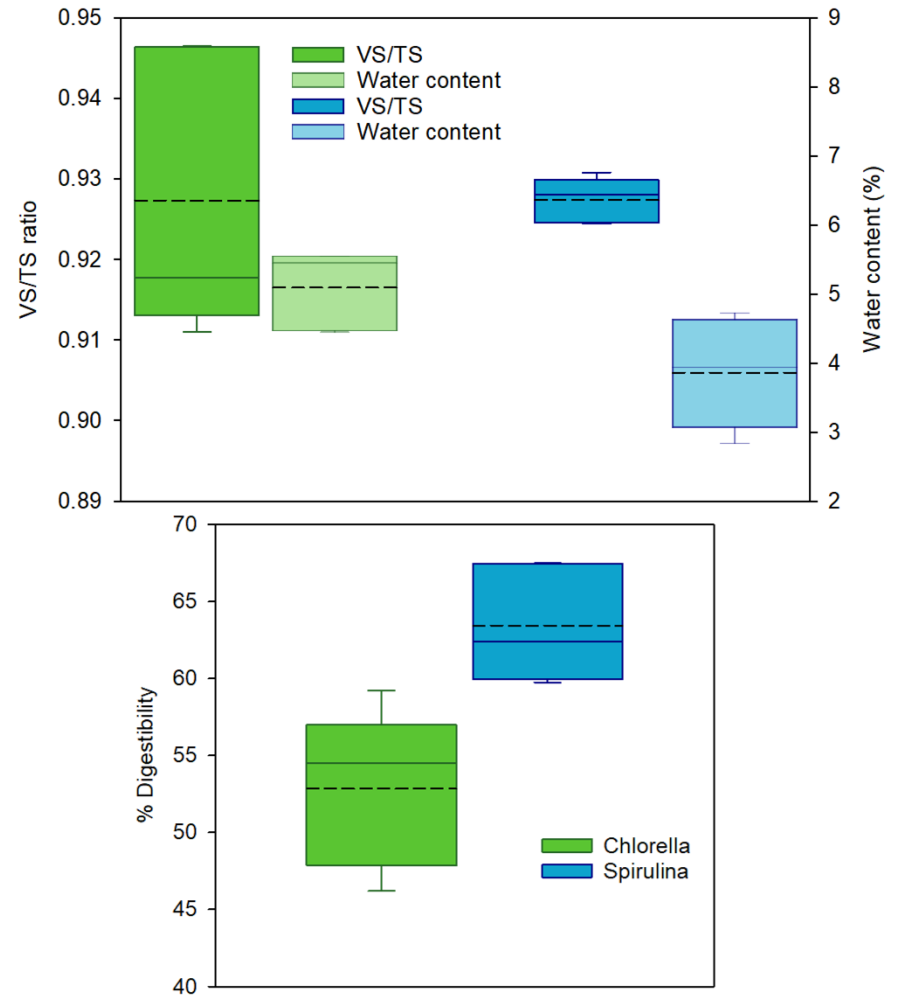
*Pellets; NA: Not Applicable

Spatial and temporal variation in digestibility, VS/TS ratio and water content

Spatial variability



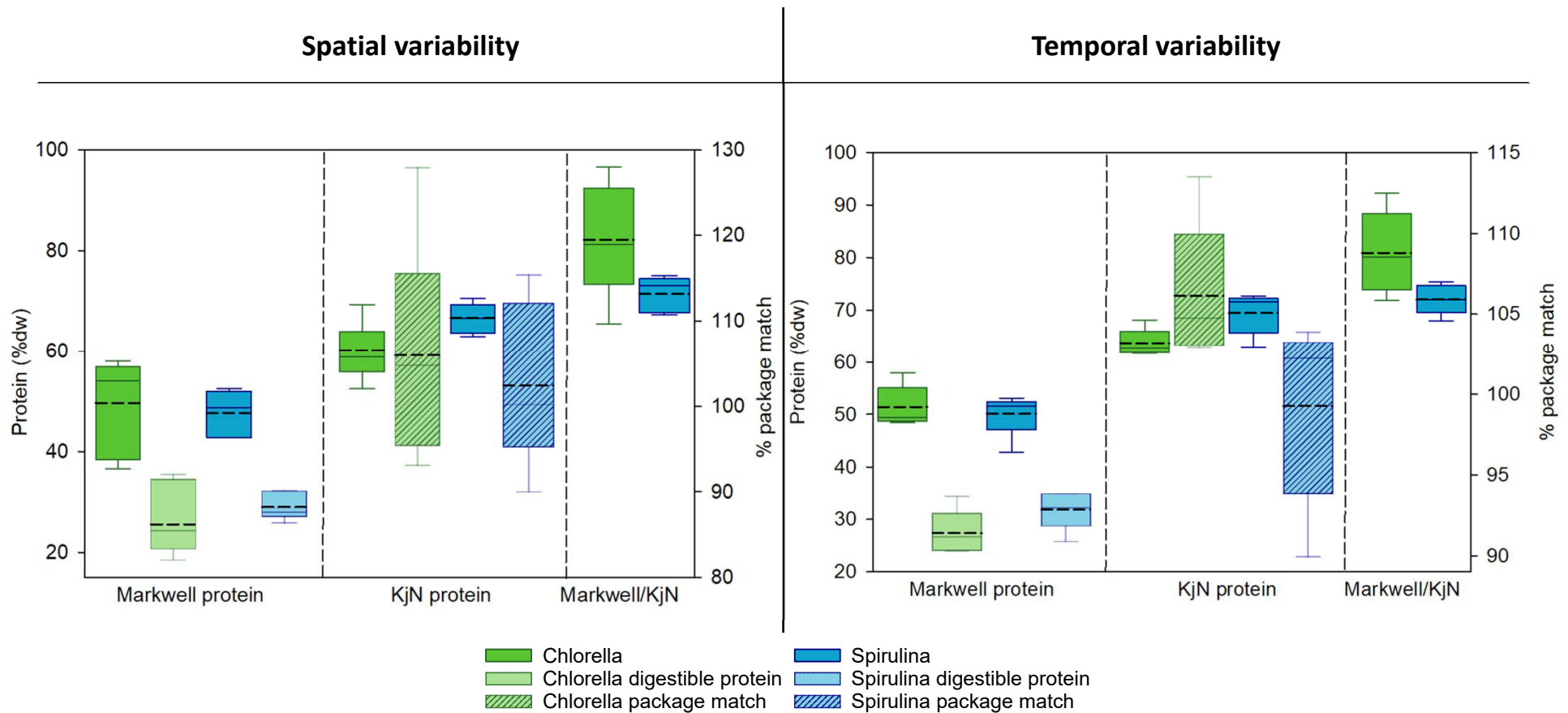
Temporal variability



- Chloroella: 50% lower water content, slightly higher VS/TS ratio and a 20% lower mean digestibility
- Spatial variability in digestibility is larger for Chloroella (broken vs non-broken cell walls?)

- Temporal variability in ash fraction up to 80% for Chloroella => not only spatial effect
- Up to 28% temporal difference in digestibility => significant effect of growing condition on digestibility

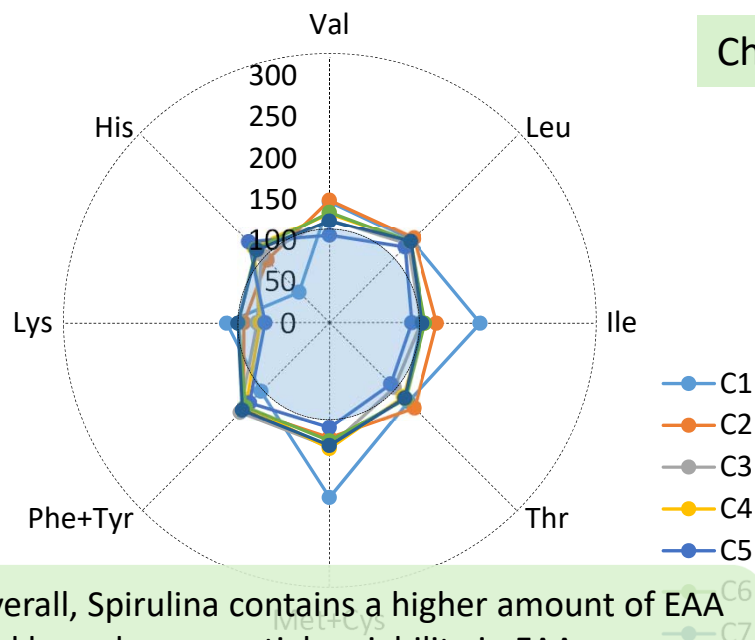
Spatial and temporal variation in protein content (Quantity)



- Chlorella: 58% spatial difference in protein
- Spirulina: 23% spatial difference in protein
- Package protein content is based on KjN
- Up to 37% spatial difference in package match
- Markwell/KjN Chlorella > Spirulina => more non-protein N in Spirulina biomass

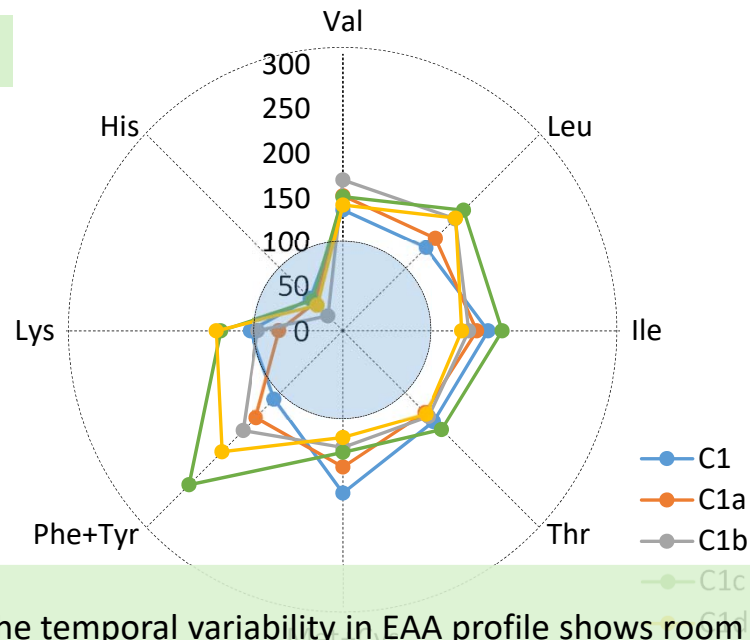
- Both microalgae 20-25% temporal difference in protein content => room for improvement even within one company!

Spatial variability in normalized EAA profile



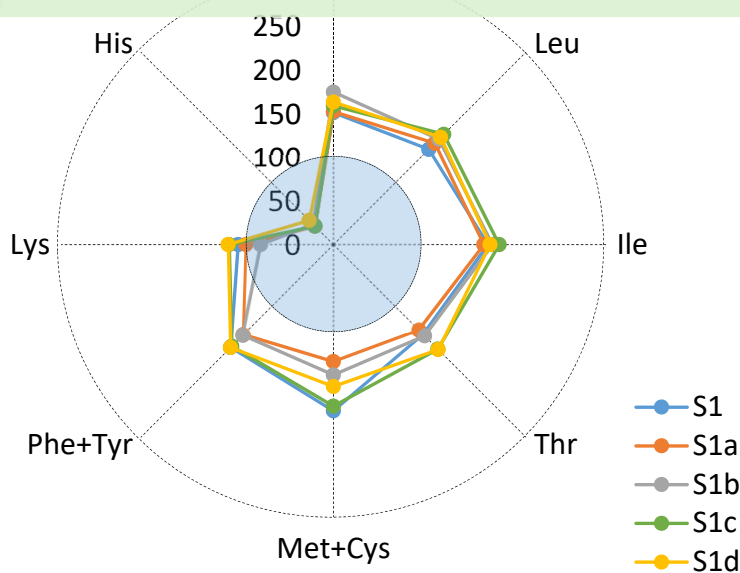
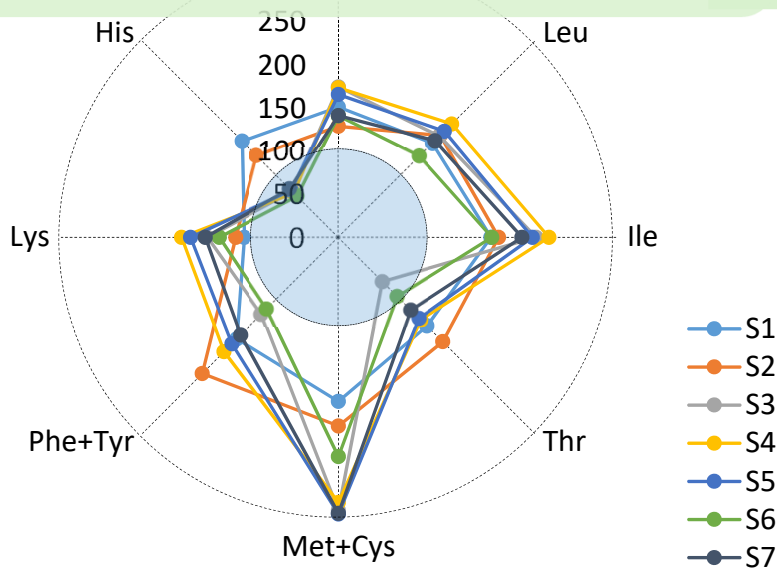
- Overall, Spirulina contains a higher amount of EAA and has a larger spatial variability in EAA
- Chlorocella is mainly short in Lysine
- Spirulina is mainly short in Histidine

Temporal variability in normalized EAA profile



The temporal variability in EAA profile shows room for improvement in one company:

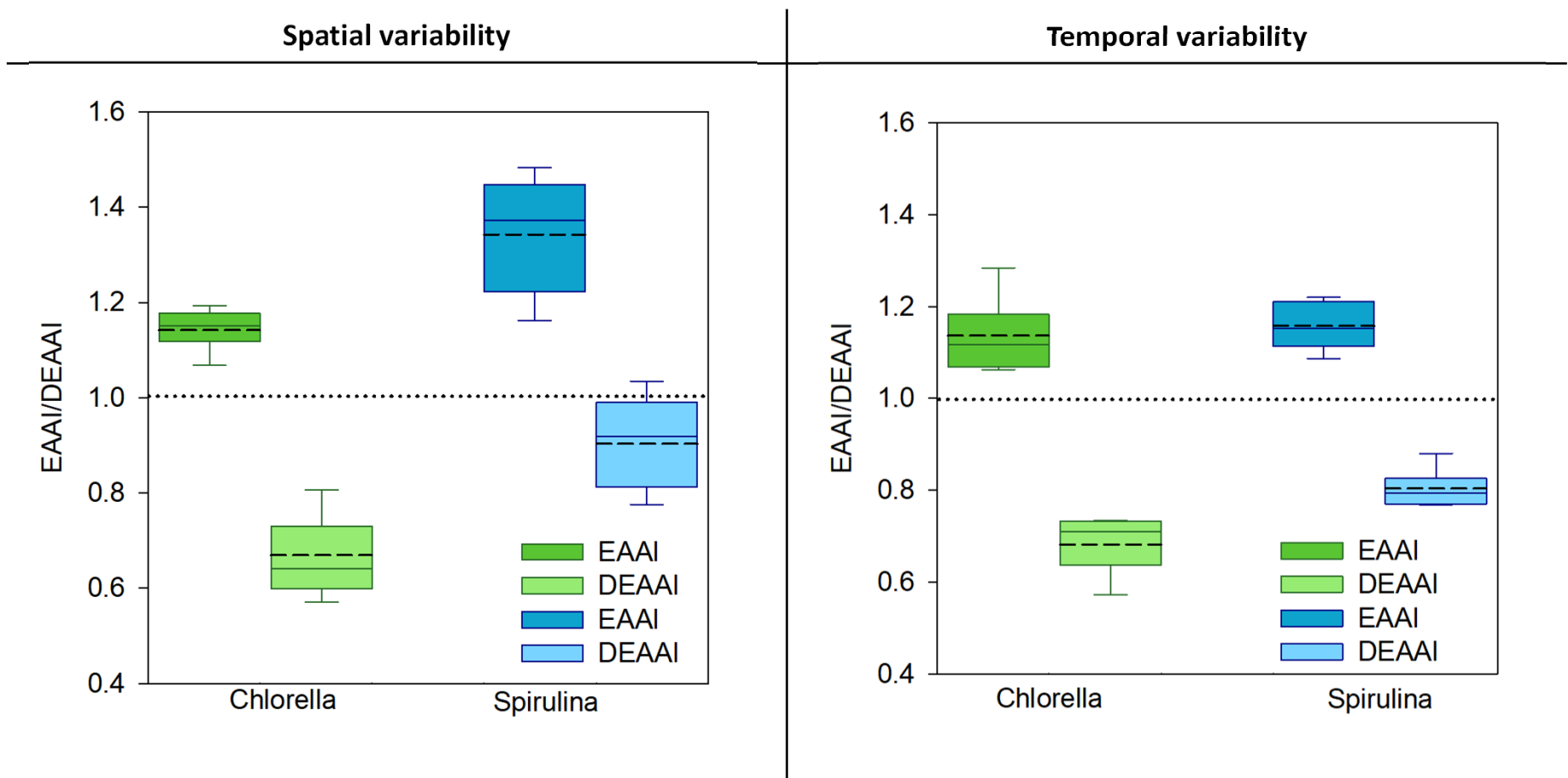
- E.g. Up to 100% difference in Lysine for both algae



Spatial and temporal variation in EAA content (Quality)

$$\text{Essential amino acid index: EAAI} = \sqrt[n]{\frac{\text{EAA1}_{\text{scp}}}{\text{EAA1}_{\text{ref}}} * \frac{\text{EAA2}_{\text{scp}}}{\text{EAA2}_{\text{ref}}} * \dots * \frac{\text{EAA}_n_{\text{scp}}}{\text{EAA}_n_{\text{ref}}}}$$

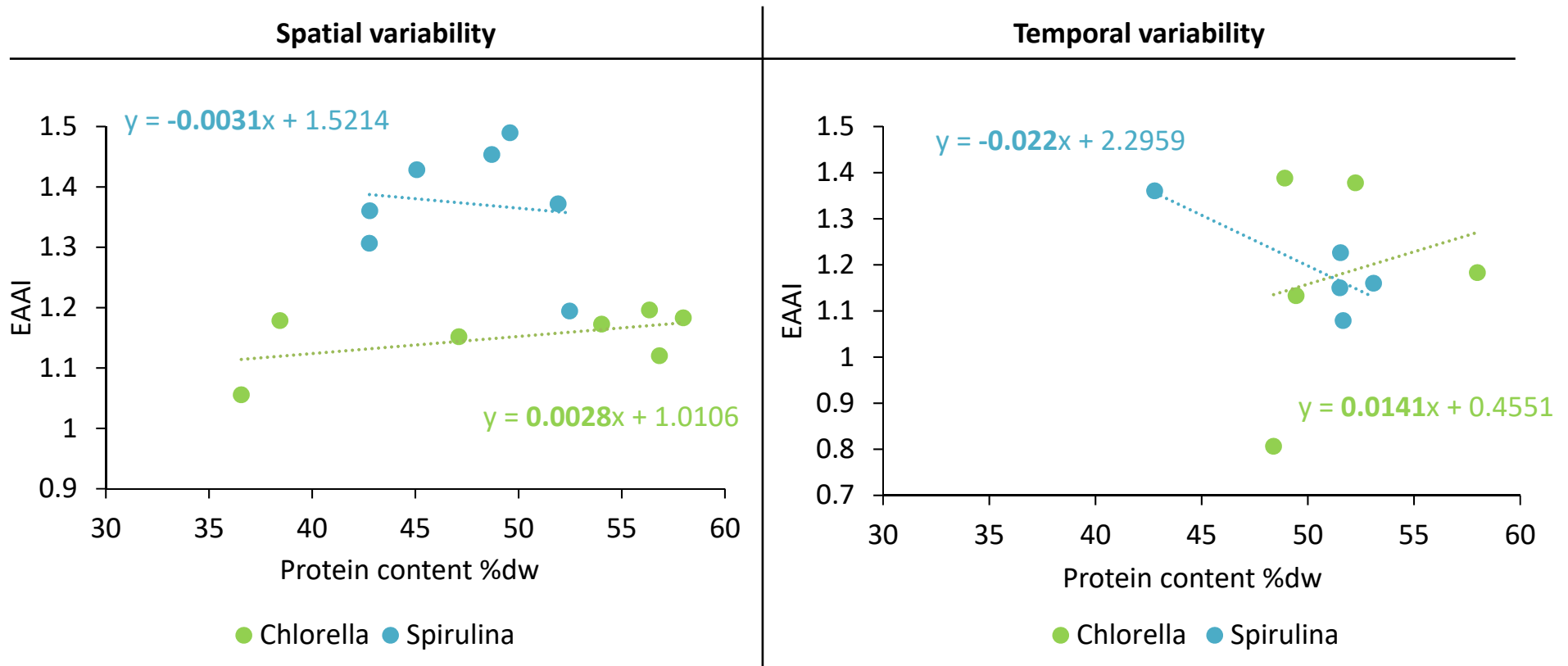
$$\text{DEAAI} = \text{EAAI} * \text{Digestibility (\%)}$$



- The EAAI differs between 13 and 29% spatial and between 12 and 20% temporal
- Spirulina has a more favourable EAAI compared to Chlorella
- Taking into account digestibility the DEAAI drops below 1, indicating an AA shortage compared to the reference protein (WHO)

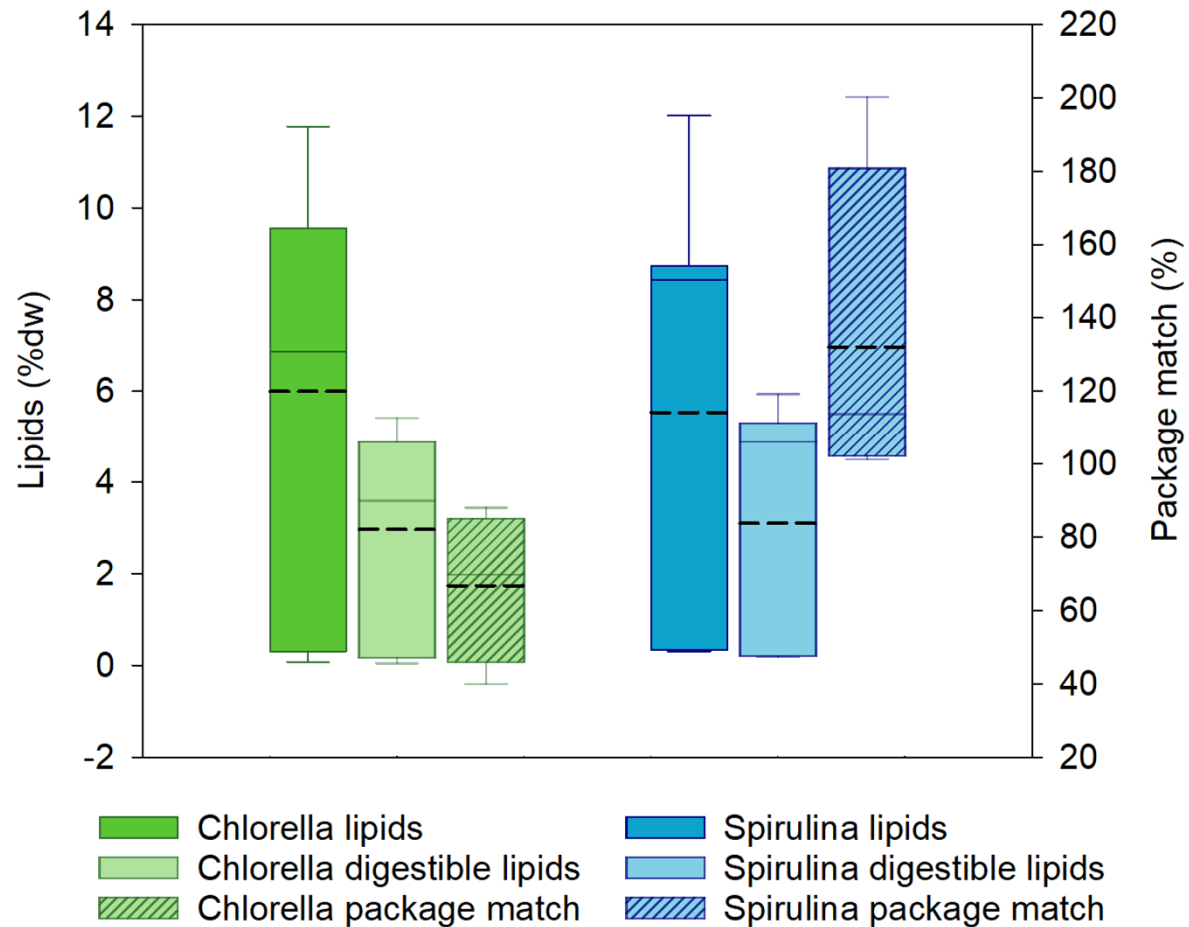
Spatial and temporal variation in EAA content (Quality vs Quantity)

$$\text{Essential amino acid index: EAAI} = \sqrt[n]{\frac{\text{EAA1}_{\text{scp}}}{\text{EAA1}_{\text{ref}}} * \frac{\text{EAA2}_{\text{scp}}}{\text{EAA2}_{\text{ref}}} * \dots * \frac{\text{EAA}_n_{\text{scp}}}{\text{EAA}_n_{\text{ref}}}}$$



High quantity ≠ high quality!

Spatial variation in lipid content

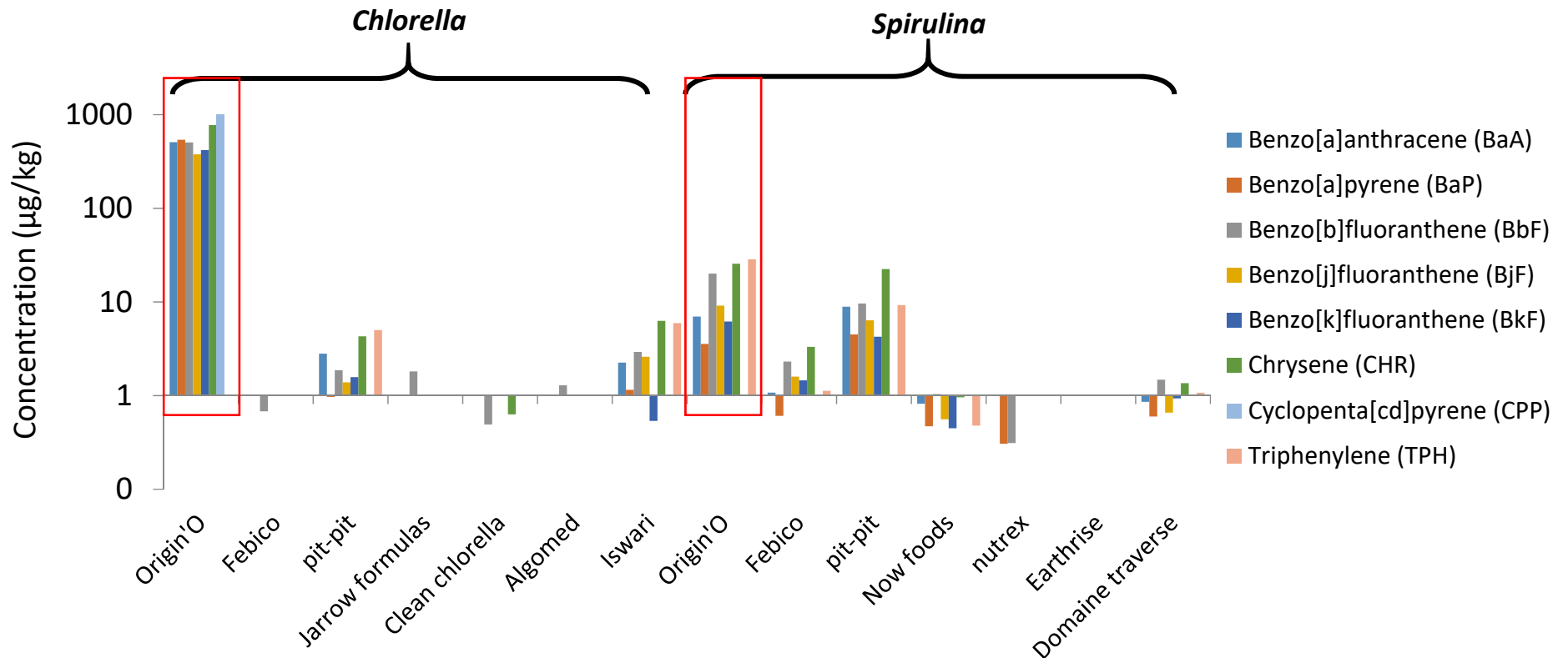


- Both Chlorella and Spirulina have a spatial difference between 0 and 12% in lipid content.
- Chlorella lipid content is rather underestimated on the package, while the opposite is true for Spirulina

Micro pollutants: PAH (Spatial variation)

Maximum levels in food supplements (Commission regulation (EU) 2015/1933 of 27 October 2015):

- Benzo(a)pyrene: **10 µg/kg wet weight**
- Sum of benzo(a)pyrene, benz(a)anthracene, benzo(b)fluoranthene and chrysene ('PAH4'): **50 µg/kg**



For *Spirulina platensis* (Origin'O), slight violation in PAH4:

- Benzo(a)pyrene: **3.55 µg/kg**
- 'PAH4': **56.13 µg PAH4/kg**

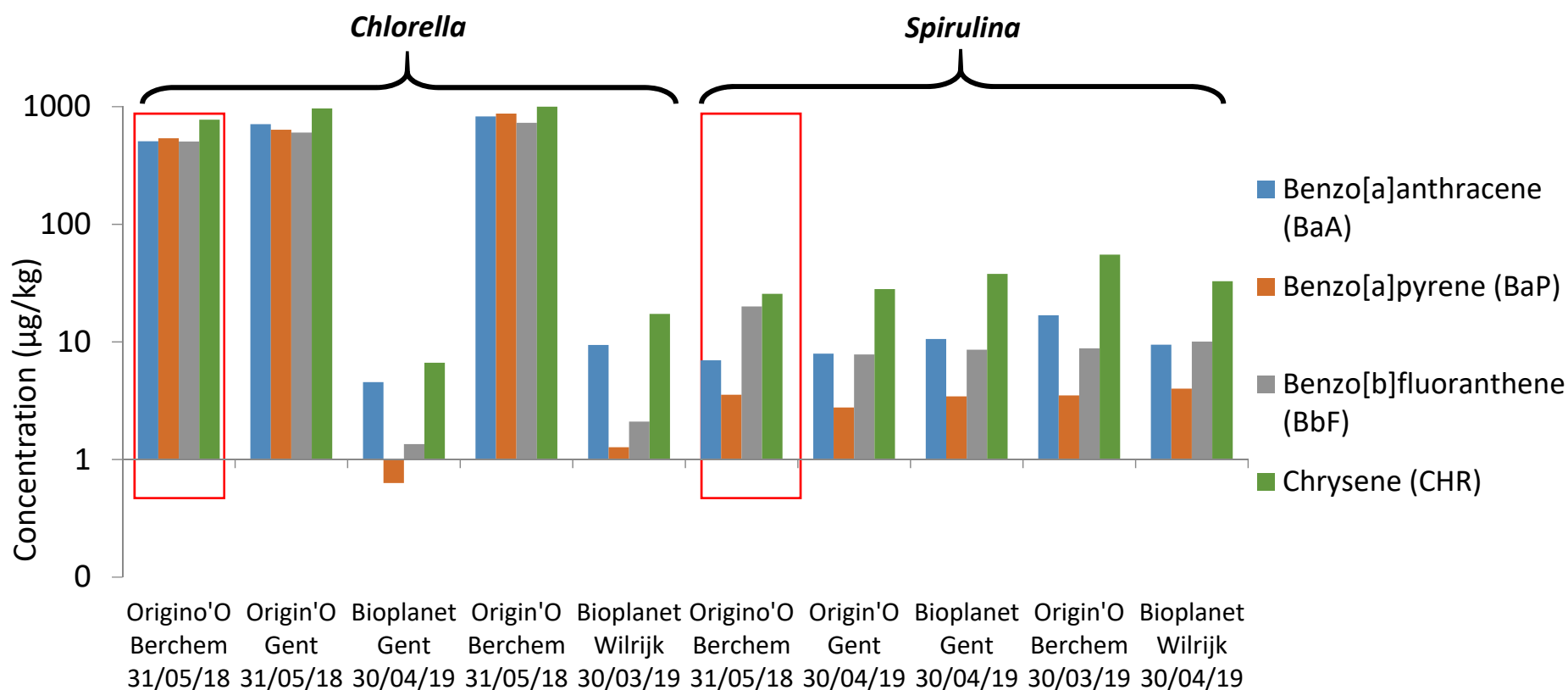
For *Chlorella vulgaris* (Origin'O), strong violation Benzo(a)pyrene and PAH4:

- Benzo(a)pyrene: **538 µg/kg**
- 'PAH4': **2324 µg PAH4/kg**

Micro pollutants: PAH (Temporal variation)

Maximum levels in food supplements (Commission regulation (EU) 2015/1933 of 27 October 2015):

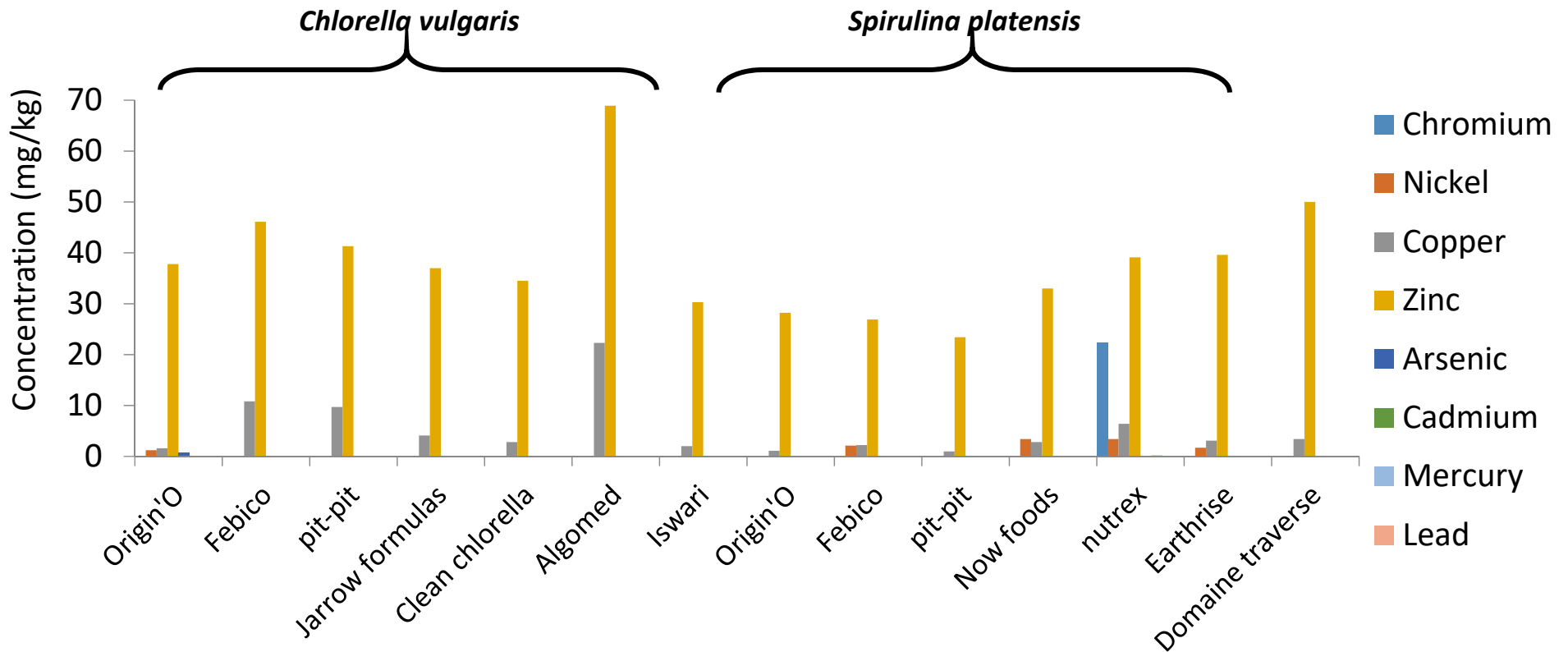
- Benzo(a)pyrene: 10 µg/kg wet weight
- Sum of benzo(a)pyrene, benz(a)anthracene, benzo(b)fluoranthene and chrysene ('PAH4'): 50 µg/kg



- 1) *Chlorella*: Newer batches didn't violate the limits for BaP and PAH4.
- 2) *Spirulina*: Newer batches didn't violate the BaP limit but they all slightly violated the PAH4 limit
=> Violations are not constant through time and are batch specific.

Micro pollutants: Heavy metals (Spatial variation)

Maximum levels in food supplements (Commission Regulation (EC) No 1881/2006):



No violations of heavy metal limits.

Conclusion

- 1) Spatial and temporal variation in nutritional composition of commercially available *Chlorella* and *Spirulina* is existing and not negligible.
- 2) Depending on the nutritional characteristic this variation can be large or rather limited.
- 3) The detected variation within one species of microalgae makes it likely that process conditions are essential in the production process of SCP. This should be further researched.
- 4) Safety of the product is not always guaranteed and should be monitored by the competent bodies.
- 5) From the digestible protein data available at this moment, *Arthrospira* is preferred for RLSS application above *Chlorella*.





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