Digestibility of meat analogues and the influence of processing

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Context

Plant-based meat alternatives of high protein quality and digestibility could be a way to reduce meat consumption and, consequently, the environmental impact. However, little is known about their nutritional characteristics and digestion behavior.

Objective

The aim of the present work was to evaluate the protein quality of highly transformed veggie burgers, based on soy or pea-faba proteins, in comparison to a beef burger. The impact of texturizing and grilling on the *in vitro* protein digestibility and the digestible indispensable amino acid ratio (DIAAR) of the ingredients and the finished products was also evaluated.
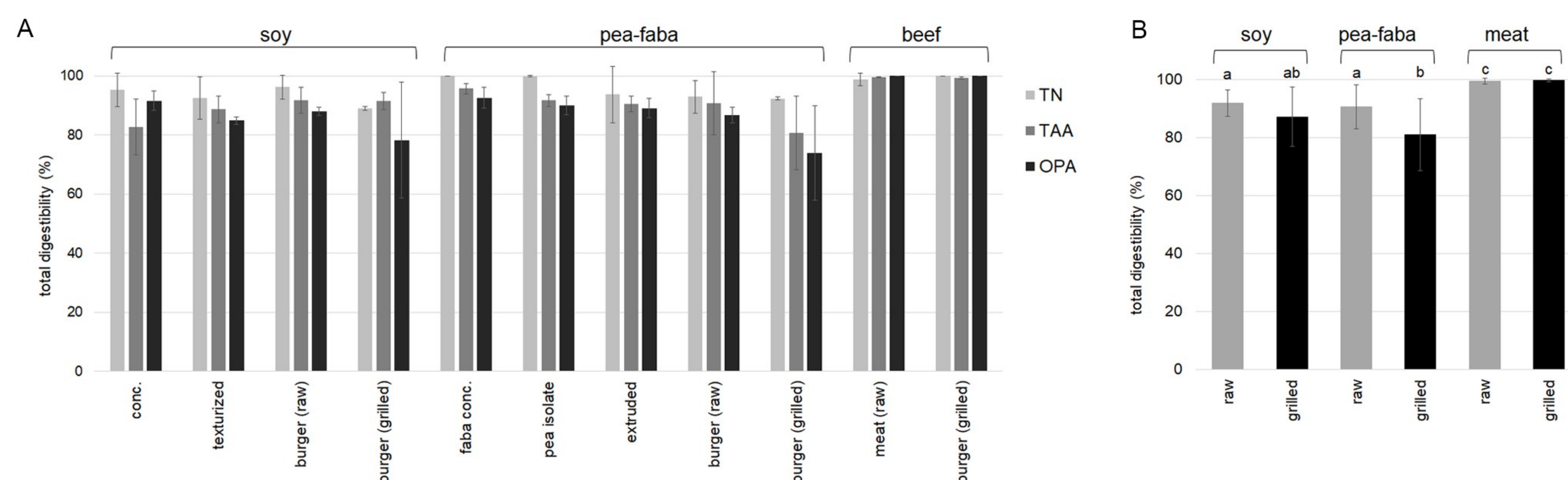
Experimental procedures

- Two highly transformed veggie burgers were digested according to the INFOGEST *in vitro* protocol[1]
- Total protein digestibility, digestibility of individual amino acids, and *in vitro* DIAAS were determined and calculated according to the *in vitro* digestibility protocol[2].

Substrates composition

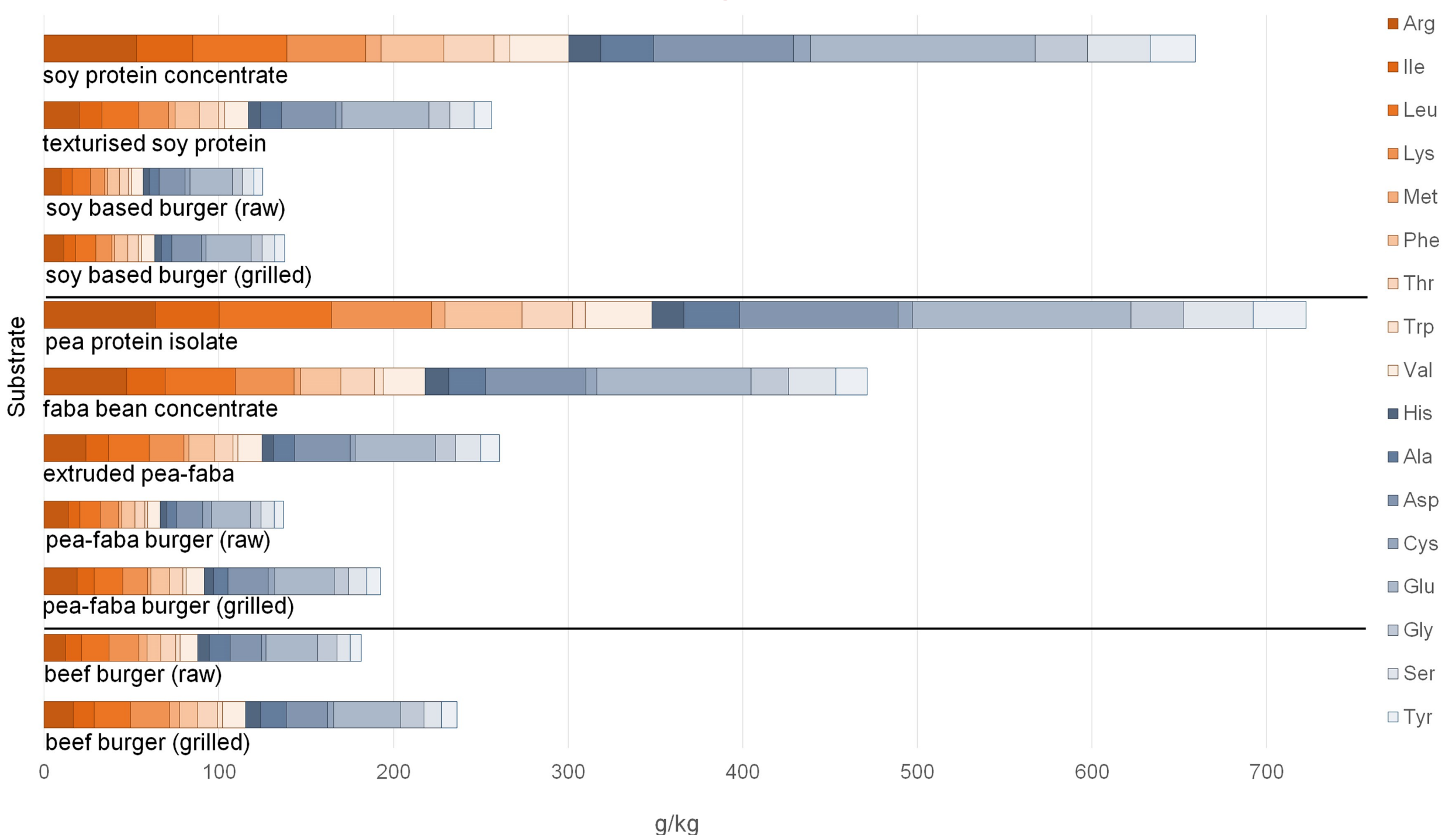| (g/100g)	Protein (TN x 6.25)	Fat (OICC)	Carbohydrates (by difference)	Moisture (Oven)
Faba bean concentrate	54.6	3.3	15.43	7.5
Pea isolate	78.6	9.1	0	5.8
Extruded pea & faba	28.7	3.1	2.2	61.1
Pea & faba burger (raw)	18.5	16.8	4.0	55.9
Pea & faba burger (grilled)	20.3	n.d	n.d	n.d
Soy concentrate	64.4	0.26	0	6.0
Texturized soy	27.3	0.31	1.3	65.6
Soy burger (raw)	12.9	13.3	1.6	65.2
Soy burger (grilled)	13.9	n.d	n.d	n.d
Beef meat (raw)	20.7	n.d	n.d	n.d
Beef burger (grilled)	24.1	n.d	n.d	n.d

Table 1. Composition of substrates in protein, fat, carbohydrates, and moisture.

Amino acid composition of the ingredients and the finished products

Figure 1. Amino acid composition (g/kg of protein source) of the ingredients and the finished products (raw, and grilled samples). Essential amino acids; nonessential amino acids.

In vitro digestibility and DIAAR calculation

a) $\text{in vitro digestibility}[\%] = 100 \times \frac{(Fs-Cs)}{((Fs-Cs)+\max(o;Fp-Cp))}$

b) $\text{in vitro DIAA} = \text{mg of IAA per g of food protein} \times \text{in vitro digestibility of IAA}$

c) $\text{in vitro DIAAR}[\%] = 100 \times \frac{\text{mg of in vitro digestible dieatary IAA in 1g of dietary protein}}{\text{mg of the same dietary IAA in 1g of the reference protein}}$

Figure 2. Formulas used for *in vitro* protein digestibility (a), *in vitro* DIAA (b), and *in vitro* DIAAR (c) calculations, respectively. Fs = Food supernatant || Cs = Cookie supernatant || Fp = Food pellet || Cp = Cookie pellet (max (0;Fp-Cp)) indicates that the amount of amino acids from the protein-free cookie digest was set as minimum.

In conclusion, the grilled beef burger had the highest *in vitro* DIAAS value (Leu 124 %), and the grilled soy protein-based burger reached *in vitro* DIAAS values that could be rated as good (SAA 94 %), according to FAO. The texturizing process did not significantly affect the total protein digestibility of the ingredients. However, grilling led to a decrease in digestibility and DIAAR of the pea-faba burger (P<0.05), which was not observed in the soy burger, an even led to an increase in DIAAR in the beef burger (P<0.005). Furthermore, our results confirm the different effects of cooking is depending on the protein

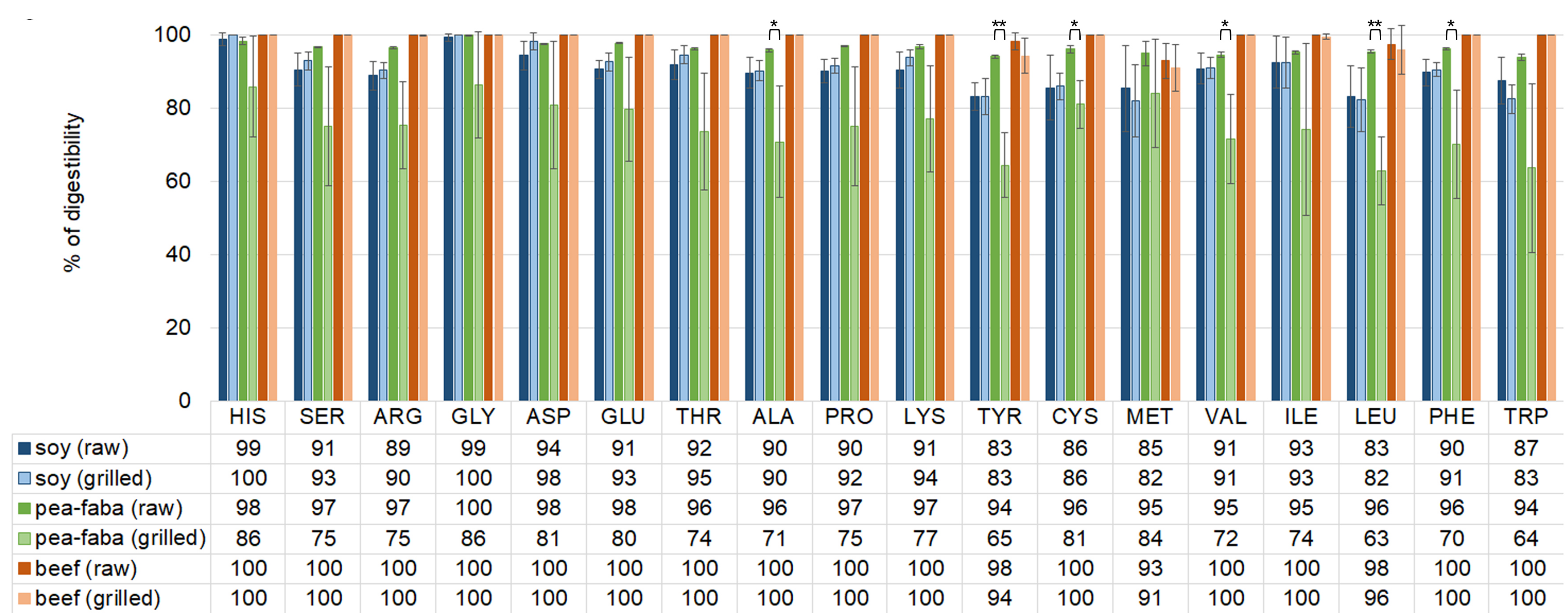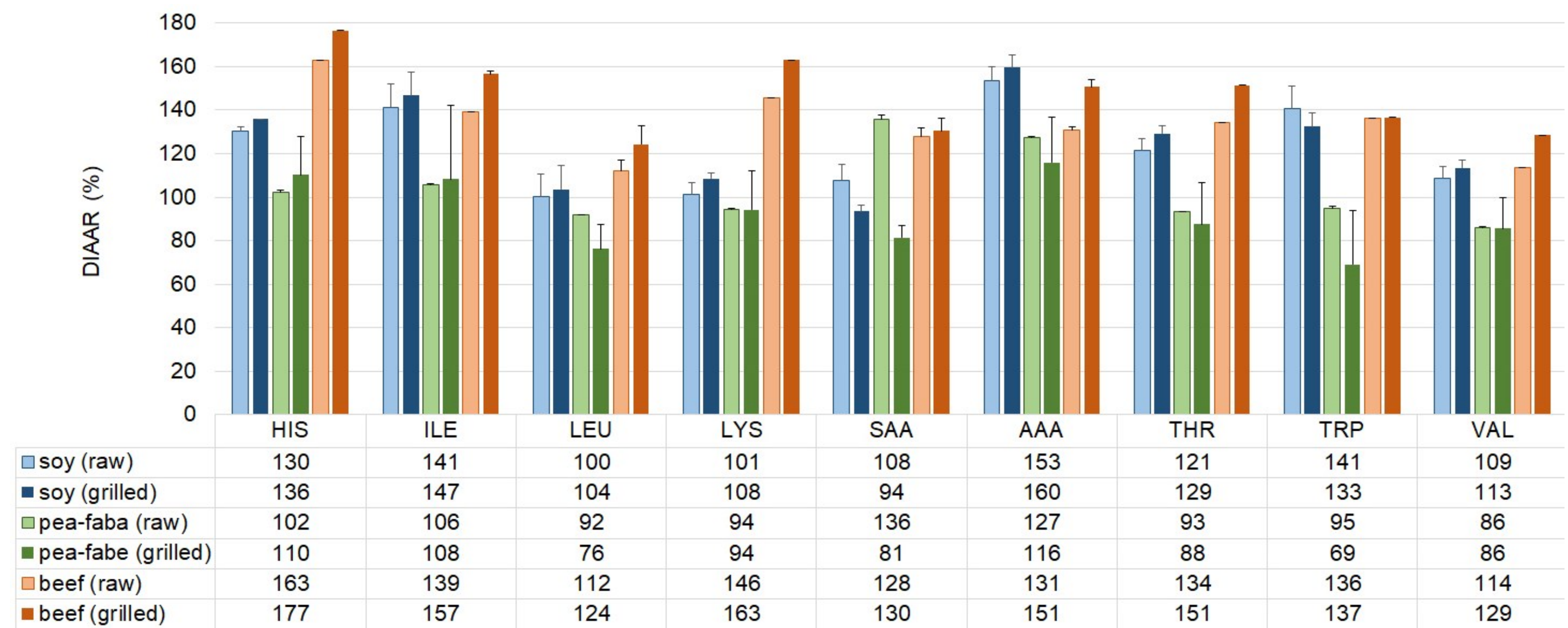## Total digestibility of the ingredients and finished products

Figure 3. Total digestibility of the ingredients and finished products. All substrates were analyzed using three different methods (TN, NH2, and TAA). All the ingredients (isolated proteins) were digested together with 0.25 g of protein-free cookie to mimic a real meal. At least three independent experiments were performed, and error bars represent the standard deviation (SD) (A); average total digestibility across all three analytical methods (TN, NH2, and TAA, N≥9) for raw and grilled burgers; significant different digestibilities are indicated with different letters (B).

Grilling effect on individual amino acid digestibility

	HIS	SER	ARG	GLY	ASP	GLU	THR	ALA	PRO	LYS	TYR	CYS	MET	VAL	ILE	LEU	PHE	TRP
soy (raw)	99	91	89	99	94	91	92	90	90	90	91	83	86	85	91	83	90	87
soy (grilled)	100	93	90	100	93	93	95	90	92	94	83	86	82	91	93	82	91	83
pea-faba (raw)	98	97	97	100	98	98	96	96	97	97	94	96	95	95	95	96	96	94
pea-faba (grilled)	86	75	75	86	81	80	74	71	75	77	65	81	84	72	74	63	70	64
beef (raw)	100	100	100	100	100	100	100	100	100	100	98	100	93	100	100	98	100	100
beef (grilled)	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100

Figure 4. Effect of grilling on individual amino acid digestibility. Comparison of digestibility of individual amino acids of plant-based burgers from soy (blue bars) and pea-faba (green bars) with beef meat burgers (orange bars) under raw and grilled conditions, respectively. The error bars represent the SD of the triplicate analysis. Significant differences are indicated (*: P≤0.1 and **: P<0.05).

In vitro DIAAR values

	HIS	ILE	LEU	LYS	SAA	AAA	THR	TRP	VAL
soy (raw)	130	141	100	101	108	153	121	141	109
soy (grilled)	136	147	104	108	94	160	129	133	113
pea-faba (raw)	102	106	92	94	136	127	93	95	86
pea-faba (grilled)	110	108	76	94	81	116	88	69	86
beef (raw)	163	139	112	146	128	131	134	136	114
beef (grilled)	177	157	124	163	133	151	151	137	129

Figure 5. The DIAAR values were calculated for the pea-faba burger (green), soy-based burger (blue), and beef meat burger (orange) under raw (darker colour) and grilled (lighter colour) conditions, respectively. DIAAR values were based on total protein (TN*6.25) content and the reference requirement values for preschool children (6 months to 3 years) given by the FAO[3]. The error bars are the SD of at least three analyses.

[1] Brodkorb, A. et al. (2019). INFOGEST static *in vitro* simulation of gastrointestinal food digestion. Nature Protocols, doi:10.1038/s41596-018-0119-1
[2] Sousa et al., (2023). *In vitro* digestibility of dietary proteins and *in vitro* DIAAS analytical workflow based on the INFOGEST static protocol and its validation with *in vivo* data. Food Chem. **404**
[3] FAO. (2013). Dietary protein quality evaluation in human nutrition. Report of an FAQ Expert Consultation. FAO Food and Nutrition Paper, **92**, 1–66

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