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Membrane chromatography to fractionate plant proteins from rapeseed and pea seed: a promising approach

Objective: evaluate the feasibility of replacing resin chromatography with membrane techniques for the purification of plant globulins from oleaginous and pulse proteins sources.

7S & 11S globulins from pea

The fractionation of proteins extracted from pea flour (protein content = 21.5%, dry basis) on chromatography resin (HitrapFF Q Cytiva, 1mL) was compared with membrane fractionation (Mustang® Q XT Acrodisc® Pall, 0.86mL) and electrophoresis of the fractions eluted by NaCl gradient allowed the identification of proteins present in each peak.

The scale-up was evaluated using the Sartobind® Q 20mL membrane (Sartorius) with step elution. Multiple cycles were repeated, with cleaning steps between them, to produce larger amounts of protein: from 110g of pea flour, **3g of 7S** and **0.7g of 11S globulins** were produced in **13 runs** lasting approximately 4h.

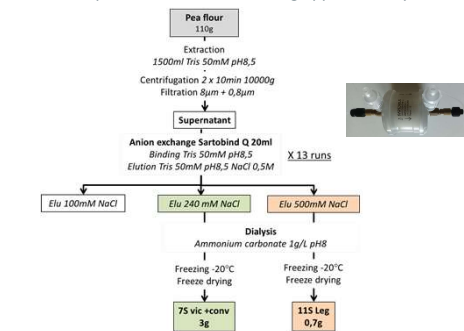
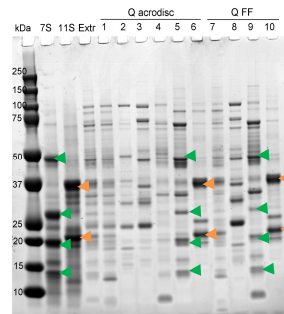
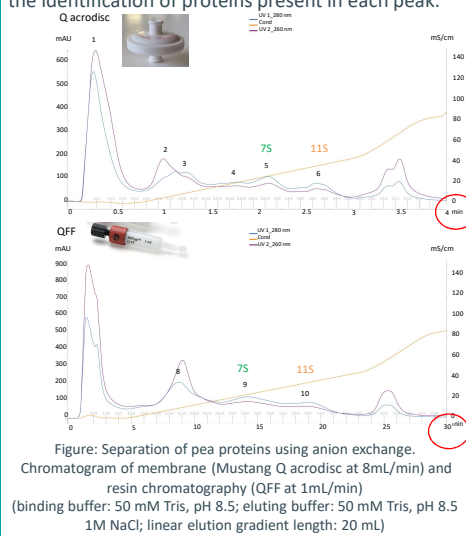


Table: Fixation capacity of pea globulins per hour per mL of column.

	Resin Membrane	
	200	20
Column volume (ml)	200	20
Total purification time (h)	6	4
Purified 7S amount (g)	9	3
Purified 11S amount (g)	3	0,7
7S capacity (mg / h / ml column)	8	38
11S capacity (mg / h / ml column)	3	9

- ↪ Separation of 7S and 11S globulins: 4 min for membrane, 30 min for resin.
- ↪ Binding capacity: 45 mg of protein/mL of membrane ≈ resin.
- ↪ Productivity increased with membrane compared to resin at preparative scale.

12S globulin & 2S albumin from rapeseed

The proteins were extracted from rapeseed meal (protein content = 38.6%, dry basis) then desalted. Their fractionation on chromatography resin (HitrapFF SP Cytiva, 1mL) was compared with membrane fractionation (Mustang® S Acrodisc® Pall, 0.86mL) and electrophoresis of the fractions eluted by NaCl gradient allowed the identification of proteins present.

The **membrane binding capacity** was measured by continuous feeding until saturation was achieved, as determined by a UV trace that asymptotically flattened out → **4 mg of protein/mL of membrane**.

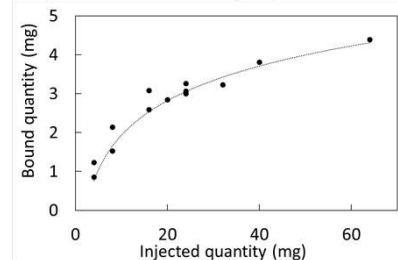
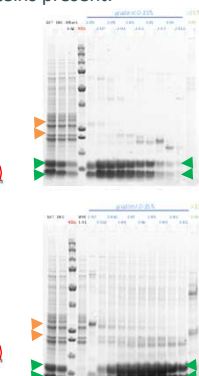
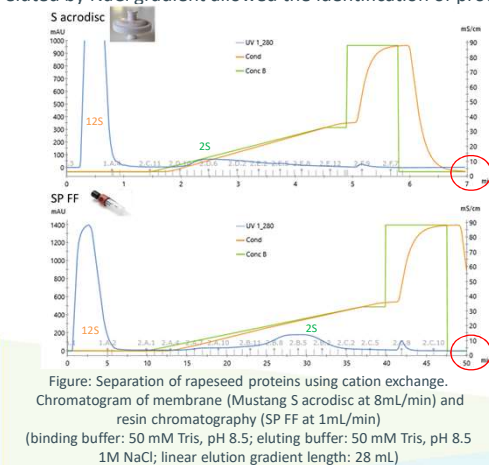


Figure: 2S albumin equilibrium adsorption on Mustang® S acrodisc membrane.

- ↪ Lower binding capacity for 2S albumin in the membrane compared to resin.
- ↪ Upscale on Sartobind® 20mL allowed the production of 5g of 2S albumin and 11g of 12S globulins

Conclusion

Membrane technology advantages: high flow rates, low pressure drop and ease of scale-up.

- ↪ Efficient fractionation of plant proteins in a shorter time compared to resin chromatography, thus increasing productivity.
- ↪ Interest of the process to produce food ingredients at industrial scale.

