Impact of dry-fractionation and cooking on antinutrients in yellow pea and faba bean protein fractions

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INTRODUCTION

Pulses have been gaining interest as an alternative and **sustainable plant protein source**. Dry seeds are rich sources of proteins, energy, carbohydrates, fiber, B-vitamins, and minerals. However, pulse seeds also contain minor components, known as **antinutrients**, which include enzyme inhibitors, lectins, alkaloids, phytic acid, and some specific phenolic compounds like saponins and tannins. The existence of these components in the human digestive tract **reduces nutrient absorption, pulses' digestibility, and nutrients' bioavailability** [1]. Nonetheless, some of these antinutrients are reduced or eliminated during soaking, cooking, and processing [2].

Dehulling, air classification and cooking can be applied to pulses with impact on the chemical, physicochemical and nutritional properties of the final fractions.

In the present study, **protein concentrates derived from yellow peas and faba beans**, obtained by **dry-fractionation** [3], and their corresponding raw-materials were characterized for its **condensed tannins content, trypsin inhibition activity and total saponin content**, following spectrophotometric assays.

Aim

Evaluate the impact of processing namely dehulling, dry fractionation (by milling and air-classification) and cooking on specific antinutrients, such as trypsin inhibitors, total saponins and condensed tannins.

METHODS

1. Preparation of protein concentrates by dry-fractionation

Yellow peas (*Pisum sativum* L. var. Ingrid) and faba beans (*Vicia faba* L. var. Kontu) were dehulled, milled and air-classified according to the method described by Saldanha do Carmo et al. [3].

2. Heat treatment (cooking) using a Rapid Visco Analyser (RVA)

Heat-treated raw materials and fine fractions were produced under controlled conditions, using a RVA (RVA-4500, Newport Scientific, Australia), following the AACC Method 76-21.02 (1997). The obtained heat-treated samples were freeze-dried and milled before further analysis.

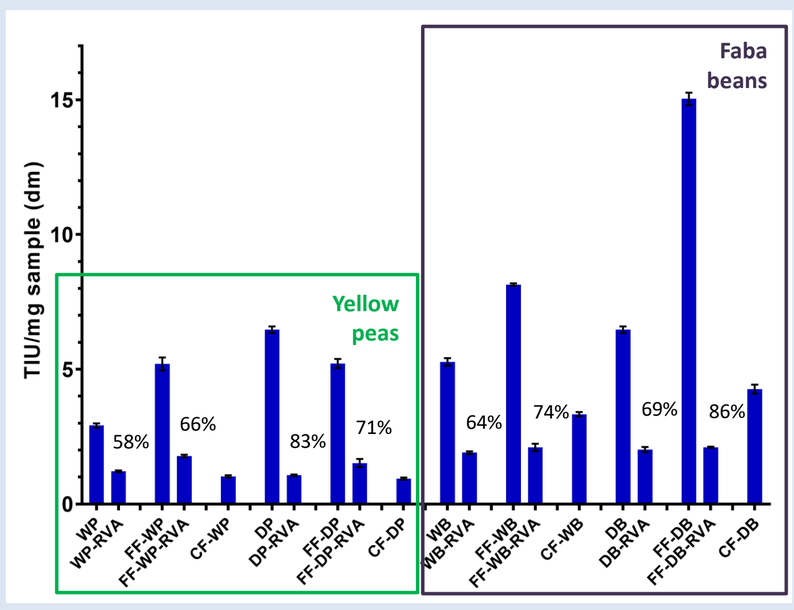
3. Analysis of total saponins, condensed tannins and trypsin inhibitory activity

Table 1. Followed characterization methodologies

Response	Method	Reference
Total saponins (g aescin eq./100g sample)	Spectrophotometric method (absorbance at 540 nm)	Han et al. 2023 [4]
Condensed tannins (mg catechin eq./g sample)	Spectrophotometric method (absorbance at 500 nm)	Çam et al. 2010 [5]
Trypsin Inhibition activity (TIU)	Spectrophotometric method (absorbance at 410 nm)	Liu et al. 2021 [6]

RESULTS

1. Trypsin inhibition activity

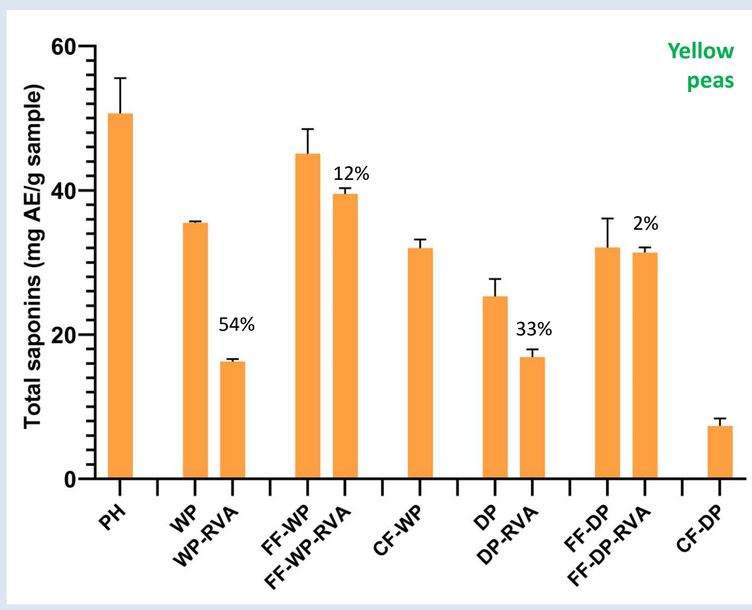


WP: Whole peas; DP: Dehulled peas; FF: Fine fraction (protein-rich); CF: Coarse fraction (starch-rich) WB: Whole faba beans; DB: Dehulled faba beans; RVA: Rapid Visco Analyser (atmospheric cooking program)

Figure 1. Trypsin inhibitory units (TIU) in yellow peas and faba beans before (WP and WB) and after dehulling (DP, DB) and dry fractionation (FF and CF) and respective reduction percentage after cooking.

The trypsin inhibitors were concentrated in the protein fraction from peas and faba beans upon dry fractionation. Moreover, the respective cooked (RVA standardized heating method) raw-materials and fractions were also analysed for trypsin inhibitors. A reduction between 58 and 86% in trypsin inhibitors was observed when the samples were subjected to atmospheric cooking.

2. Total saponins

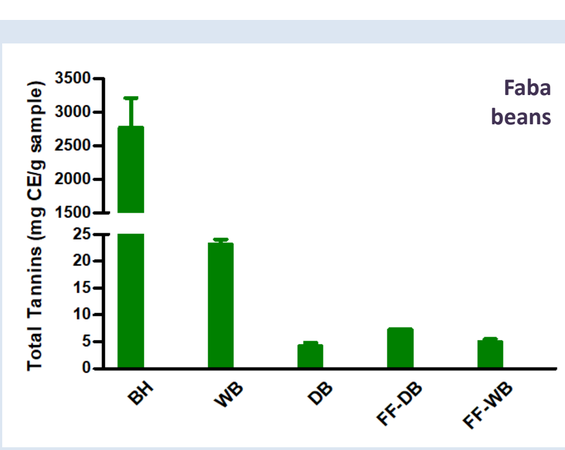


WP: Whole peas; DP: Dehulled peas; FF: Fine fraction (protein-rich); CF: Coarse fraction (starch-rich); RVA: Rapid Visco Analyser (atmospheric cooking program)

Figure 2. Total saponins in yellow peas before (WP) and after dehulling (DP) and dry fractionation (FF and CF) and respective reduction percentage after cooking (RVA).

The saponins were concentrated in the protein fraction from peas upon dry fractionation. Upon cooking, the dehulled and whole peas had a reduction between 33 and 54% in the saponin content. A smaller reduction was observed in the protein fractions derived from DP and WP (between 2 and 12%), respectively.

3. Condensed tannins



The tannins are mainly in the hulls of the faba beans. Thus, the dehulling step is crucial to remove this constituent. Since dehulling is not 100% efficient for the kernels small parts of remaining hulls will have a contribution of tannins to the dehulled kernels. However, when we look at the tannin content of the protein-fractions derived from whole beans and dehulled beans, the tannin content is not very different between these samples since the tannins will mostly/in practice end up in the starch-rich fractions upon air classification. This indicated that the dehulling step is not needed when the interest is focused on a low-tannin protein concentrate.

Figure 3. Condensed tannins in whole and dehulled faba beans, respective protein fractions and hulls

WP: Whole peas; DP: Dehulled peas; FF: Fine fraction (protein-rich); CF: Coarse fraction (starch-rich); WB: Whole faba beans; DB: Dehulled faba beans

CONCLUSIONS

- Trypsin inhibitors and saponins were concentrated in the protein fractions upon dry-fractionation.
- The dehulling step is not needed when the interest is focused on a low-tannin protein concentrate.
- Overall, cooking reduced trypsin inhibitors and saponins.

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