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Effect of Processing Methods on Antinutritional Factors in Horse Gram

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ABSTRACT

Horse gram (*Macrotyloma uniflorum*) is a drought-hardy pulse that is eaten in many dry regions of South Asia and is valued for its protein and its traditional use as a cheap food, yet it is also known for being hard to cook and for carrying a heavy load of antinutritional factors. This narrative review asks what the published literature says about how soaking, dehulling, germination, cooking, roasting, fermentation and combinations of these affect the main antinutrients, namely protease inhibitors, phytic acid, polyphenols and tannins, lectins, flatulence-causing oligosaccharides and oxalate. It draws on work on horse gram where that exists and on better-studied pulses where it does not, and it reports no new data. Heat and germination act on different factors, dehulling removes mostly the compounds held in the seed coat, and leaching depends as much on water volume and temperature as on time. A reduction in one factor frequently comes with a loss of something worth keeping, and the polyphenols that lower mineral absorption are the same ones credited with antioxidant activity. Studies of horse gram vary in cultivar, method and units, and few measure absorption in people, so a lower laboratory value is not the same as a better-nourished eater. The review concludes that sequences of simple household steps are the most defensible approach and sets out what a comparable study of processing should report.

Keywords: Horse gram; *Macrotyloma uniflorum*; Antinutritional factors; Food processing; Phytic acid; Tannins; Trypsin inhibitors; Narrative review

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Article Class: Narrative Review

1. Introduction

Horse gram, *Macrotyloma uniflorum*, is a small, hard-seeded pulse grown on poor soils with little water, and it has long been eaten by farming households in dry parts of South Asia (1, 2). It rarely appears in the foods that urban consumers think of as

pulses. It is cooked as a whole-seed dish, a soup or a ground flour, and it is often associated with cheap, plain eating. That reputation understates a seed that is rich in protein and carries a good deal of phenolic material (3, 4).



The same seed is not easy to eat well. Like most pulses it carries compounds that interfere with digestion or with the absorption of minerals, and these are usually grouped as antinutritional factors (5, 6). The group includes protease inhibitors, phytic acid, polyphenols and tannins, lectins, oligosaccharides that ferment in the large bowel, and oxalate. None of these is peculiar to horse gram, but the seed coat of horse gram is thick and strongly coloured, and the cooking time is long, so processing matters more for it than for many softer pulses.

Households have always dealt with this by doing something to the seed before it reaches the pot: soaking it, splitting or dehulling it, letting it sprout, roasting it, fermenting a batter. The question this review asks is what published work can tell us about what each of those steps does to the antinutrients, and what it costs in other nutrients. We report no local data; the title refers to processing methods in general, and the evidence is drawn from the published literature on horse gram where it exists and on related pulses where it does not. The aim is a fair account of what is known and what is only repeated.

The article is a narrative review, and it says so plainly. It does not claim a systematic search, and it avoids quoting figures from individual experiments that we could not check against their primary papers. What it offers instead is a structured reading of how the main processing steps behave, where the evidence is thin, and what a study would need to report to be useful to the next reader.

2. Literature Review

2.1 Horse Gram as a Food Crop

The older reviews of horse gram describe a crop of marginal land that supplies protein and minerals to households with few

alternatives, and they note its use as a fodder as well as a food (1). It belongs to the wider group of underutilised legumes whose nutritional potential has been argued for much more often than it has been measured (2). Pulses in general supply protein, fibre and a range of micronutrients, and they are valued for their place in diets where animal foods are costly (7, 8), and their proteins have attracted interest in their own right (9).

Work on the phenolic content of horse gram has grown in recent years, much of it from an interest in antioxidant activity. Extracts of the seed show marked free-radical-scavenging activity in laboratory assays (3), the phenolic compounds are unevenly spread across the milled fractions of the seed (10), and comparisons across commonly eaten Indian legumes place horse gram among the more active (4). Those papers are about benefit, and the wider phytochemical literature on pulses frames the same compounds as health-relevant (11). The nutritional literature reads the same compounds as a cost, because phenolics can bind protein and iron (12, 13). Holding both readings at once is the central difficulty of this topic.

2.2 What the Antinutritional Factors Are

Protease inhibitors are small proteins that block trypsin and chymotrypsin. The soybean literature distinguishes a Kunitz type, which is relatively heat-labile, from a Bowman-Birk type, which is more stable to heat (14, 15). Reviews of antinutrients in food proteins link such inhibitors to reduced protein digestibility and to lower availability of amino acids (5, 16). Lectins, which bind to sugars on the intestinal lining, add to the concern when pulses are eaten raw or undercooked (6).

Phytic acid is the main store of phosphorus in the seed, and it binds iron, zinc and



calcium in the gut so that less of each is taken up (17, 18). Trials of iron absorption in people showed that phytate inhibits absorption in a dose-dependent way and that the more highly phosphorylated forms are the most inhibitory (19, 20). Zinc, copper and manganese are affected in a similar fashion (21), which matters most in diets where pulses and cereals supply most of the minerals. Table 1 lists the factors discussed in this review, where in the seed each sits, and the concern attached to it.

Table 1. Main antinutritional factors of horse gram and why they matter.

Factor	Main location	Nutritional concern
Protease inhibitors	Cotyledon	Impaired protein digestion (5, 16)
Phytic acid	Cotyledon and embryo	Binds iron, zinc, calcium (17, 18)
Polyphenols, tannins	Seed coat	Bind protein and iron (12, 13)
Lectins	Cotyledon	Gut irritation if raw (6)
Oligosaccharides	Cotyledon	Flatulence (8)
Oxalate	Throughout	Binds calcium (22)

Note: Qualitative summary. Amounts differ between cultivars and growing conditions, so no values are given. Locations indicate where each factor is chiefly concentrated, not an exclusive site.

Polyphenols and tannins sit mainly in the seed coat and give dark-coated pulses their colour. They bind proteins and digestive enzymes, and they bind iron in ways that depend on their structure (12, 13). The oligosaccharides of the raffinose family are not digested in the small intestine and are fermented in the colon, which is the reason pulses cause flatulence (8). Oxalate binds calcium and is of particular interest where kidney stones are a concern (22). The seed itself has a further property that is not chemical: horse gram is hard to soften, and the long cooking it needs is part of the practical problem (23).

2.3 Processing in Household and Small-Scale Practice

Reviews of traditional food preparation argue that soaking, germination and fermentation are, in effect, low-cost ways of improving the mineral value of plant-based diets (24, 25). The mechanisms differ. Soaking lets endogenous enzymes and water-soluble compounds act and move, germination activates the seed's own phytase, and fermentation brings in microbial enzymes (26, 27). Heat acts on the proteins, which is why it works on inhibitors and lectins but barely touches phytate (28, 29).

A review of another underutilised legume describes the same household methods and the same trade-offs (30). Dehulling is a different kind of step, since it removes material instead of altering it. Because tannins and many polyphenols are concentrated in the seed coat, removing the coat lowers them, but it also removes fibre and some minerals (31, 32). For a pulse with a coat as pigmented as that of horse gram the trade-off is not trivial, and it is a good example of why a lower value on one assay is not by itself a verdict on a method.

2.4 Mineral Absorption and What Counts as Success

The purpose of reducing an antinutrient is to improve what the body gets, and that is a claim about absorption, not about a laboratory assay. Work on iron has shown that degrading phytate can raise absorption, though how much depends on the rest of the meal (33, 34). The presence of ascorbic acid, for example, offsets some of the inhibition (19). A pulse with less phytate on paper may therefore contribute little more iron in practice if it is eaten with tea or without a source of vitamin C.

The wider mineral literature makes a similar point. Reviews of mineral bioavailability from legumes show that composition and absorption are linked only



loosely (35), and that vegetable protein sources differ in how they affect trace elements (18). We take from this a simple rule for reading the literature on horse gram: a processing study without some measure of function, whether digestibility or mineral uptake, has told us what happened to a compound and not what happened to the eater. Read this way, a small change in a laboratory value is not evidence of a nutritional gain.

3. Materials and Methods

3.1 Scope and Question

The review asks one question in two parts. Which processing methods are reported to lower the main antinutritional factors in horse gram and comparable pulses, and what do they cost in other nutrients and in practical effort? We divided it into sub-questions that a study of processing would have to answer: which factor was measured and how, how the seed was prepared and in what units the result is given, whether the processing water and the discarded fractions were analysed, and whether any measure of digestibility or mineral absorption was included.

3.2 Literature Assembly

The review draws on published, peer-reviewed work in four areas: the composition and phenolics of horse gram; the chemistry and nutritional significance of antinutrients; studies of soaking, dehulling, germination, thermal treatment and fermentation in pulses; and work on mineral and protein availability. Sources were chosen for relevance, standing in the field and clarity of method. Studies of other pulses are used for what they show about mechanism and limits and are not offered as evidence of an effect in horse gram.

We did not run a formal systematic search and we do not claim one. This is a narrative synthesis, and the choice of

sources reflects the authors' judgement.

3.3 Appraisal Framework

To decide how far a report can be trusted we used four questions. Is the factor measured by a method that captures the compound of concern, or only a related one (36)? Are the results stated per unit of dry matter, so that loss of soluble solids is not mistaken for destruction of the antinutrient? Is the cultivar named, so that variety is not confused with method (2)? And is there any functional outcome beyond the assay (16, 34)? Table 3 sets out the gaps these questions tend to expose and what would close them.

Table 3. Common gaps in studies of processing and antinutrients, with remedies.

Gap	Consequence	Remedy
Units not stated	Cannot compare studies	Dry-matter basis throughout
Cultivar not named	Variety mixed with method (2)	Report cultivar and origin
Only one assay	Misses related compounds (36)	Pair chemistry with function
No absorption data	Lower value, unknown benefit (24, 34)	Measure mineral uptake
Soak water discarded unmeasured	Loss mistaken for destruction (35)	Analyse the water as well
Single method only	Real kitchens use sequences (24)	Test sequences

Note: Gaps are those discussed in the cited literature; they are not attributed to any single study.

4. Results

4.1 Soaking and Dehulling

Soaking is the cheapest step and the most widely used. It softens the seed, lets soluble compounds leach into the water, and gives endogenous phytase a chance to act, so some phytate, some oligosaccharide and some polyphenol are usually lost (26, 37). How much depends on the volume and temperature of the water, on the time,



and on whether the soak water is thrown away; if it is kept and cooked with the seed, much of what leached out is eaten again (38, 39). Table 2 summarises this and the other methods. A seed that has been given time to hydrate behaves differently from one that has not.

Table 2. Processing methods and the direction of effect reported for pulses.

Method	Factors usually lowered	Common caveat
Soaking	Phytate, oligosaccharides, some tannin (26, 37)	Soak water carries solutes away
Dehulling	Seed coat polyphenols (31, 32)	Loses fibre and minerals
Germination	Phytate, inhibitors, oligosaccharides (40, 41)	Depends on duration, temperature
Boiling, pressure cooking	Inhibitors, lectins (28, 39)	Leaching, heat damage to protein
Roasting, extrusion	Inhibitors, lectins (29, 42)	Little effect on phytate
Fermentation	Phytate, oligosaccharides (27, 43)	Needs control of the culture

Note: Directions summarise the cited literature and are not specific to one cultivar; magnitude depends on conditions.

Dehulling removes the coat, and with it most of the tannins and a portion of the polyphenols held there (31, 32). In horse gram, where phenolic compounds are distributed unevenly among milled fractions (10), dehulling should be expected to lower them noticeably. The cost is that the coat also holds fibre and minerals, and the cotyledon, where phytate and inhibitors sit, is untouched. A dehulled seed is thus lower in tannin but not in phytate, protease inhibitor or lectin, and the choice to dehull is a decision about polyphenols rather than about antinutrients in general. This is why the same pulse can

look excellent in one report and unremarkable in the next.

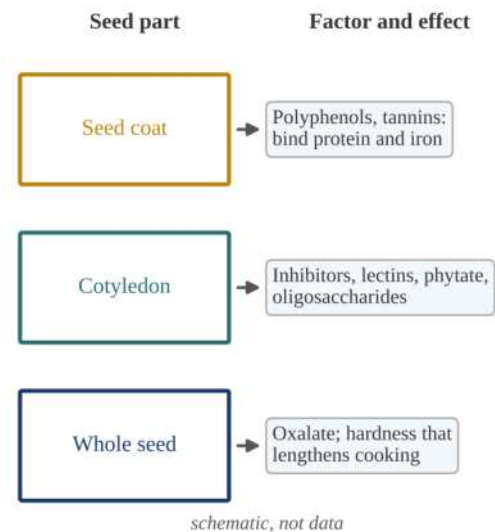


Figure 1. Where the main antinutritional factors sit in the seed and what each does (schematic, not data).

This difference in location is what Figure 1 tries to capture. Factors in the coat respond to physical removal and to leaching, whereas factors in the cotyledon respond to enzymes, to heat and to fermentation. The sections that follow can be read as a sequence of tools, each with a different reach.

4.2 Germination and Fermentation

Germination is the step with the widest effect reported for pulses. Studies on beans, lentils, peas and chickpeas describe decreases in phytic acid, in some phenolic fractions and in oligosaccharides as the seed begins to sprout (40, 41), and work on a range of seeds links this to rising phytase activity during soaking and germination (26). The same studies are cautious about generalising: the outcome depends on the temperature, the duration, whether light is allowed in and which species is under test (32).

Fermentation works through microbial enzymes and acids. Natural fermentation of beans lowered several antinutrients and, in the same study, changed other



constituents that nutritionists value (43). Fermented cereal and legume foods in Africa and Asia show how a mixed culture can serve a household cheaply, and also how variable the result is when the culture is not controlled (27). For horse gram the practical form is a fermented batter or flour, and we found no study that we could cite with confidence that measures its effect on the seed's antinutrients directly. Reviews of this kind are more useful when they say where the evidence stops than when they smooth it over.

4.3 Thermal Treatment

Heat is the most reliable way to deal with protease inhibitors and lectins, and it is also where the processing literature is most consistent. Boiling, steaming, autoclaving and pressure cooking reduced inhibitor activity in food legumes (28, 39), while extrusion and conventional cooking compared on peas and on faba and kidney beans gave different degrees of inactivation depending on conditions (29, 42). The Bowman-Birk type is more resistant than the Kunitz type, so cooking until the seed is merely edible need not remove everything (14, 15).

Heat does less for phytate, which is stable, and what is lost during cooking is usually lost into the water (28, 38). It does something for polyphenols too, although the direction is not uniform: boiling and steaming of cool-season legumes changed total phenolics and antioxidant activity in ways that depended on the species and the method (37). Dry heat, as in roasting, inactivates inhibitors and lectins without leaching anything away, but it leaves phytate in place and can damage protein if pushed too far. Reports on lentils emphasise that cooking and dehulling together alter composition in ways that no single value captures (31). Such losses are small individually, but they add up across a meal and across a season.

Cooking also bears on how horse gram is actually used. Because the seed is hard, a cook will often boil for a long time or pressure-cook it, and in either case the heat treatment is already doing the work needed for inhibitors and lectins. This is a reason to doubt claims that cooked horse gram is nutritionally risky from protease inhibitors alone. It is not a reason to ignore phytate, tannin or oxalate, which survive.

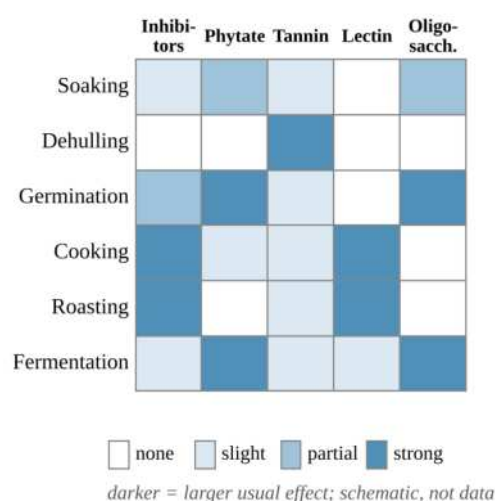


Figure 2. Qualitative matrix of processing methods against antinutritional factors (schematic, not data).

Figure 2 puts the effects side by side in a qualitative way. The pattern is that each method has a home turf, and that none covers all the factors, which is why sequences matter more than any single step.

4.4 Combinations and Sequences

Real kitchens do not apply one method. Soaking is followed by cooking, dehulling by roasting, germination by drying and grinding. Studies that test combinations generally find that the combination does better than any one step on the factors that the steps address separately (24, 32). A sequence that soaks and discards the water, allows a short germination and then cooks until soft addresses the coat, the cotyledon and the heat-labile proteins in turn (Figure 3). Whether the extra effort pays



for itself in nutrition is a different question, and it is the one that most reports leave unanswered. Nothing in this reading argues against processing; it argues for measuring what the processing does.

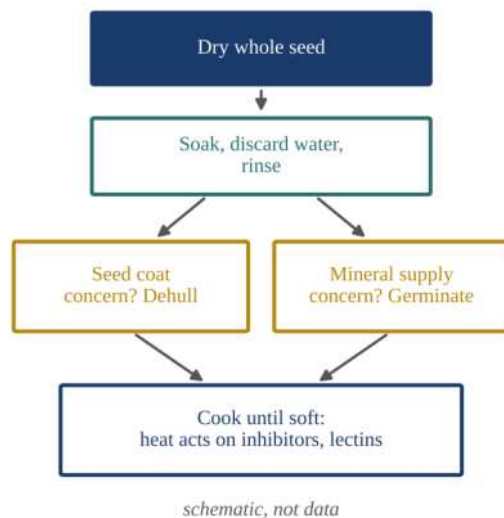


Figure 3. Simple decision sequence for combining household steps (schematic, not data).

Two caveats attach to this. First, the benefit of any reduction depends on the rest of the diet, since a meal taken with a source of ascorbic acid inhibits iron uptake less than one taken without (19, 34). Second, the polyphenols that are removed along with the coat are the compounds credited with antioxidant activity, and removing them to improve mineral uptake means giving up that benefit (3, 4). Neither caveat argues against processing; both argue for being specific about what the processing is meant to achieve. That is a modest conclusion, and it is the one the evidence will bear.

5. Discussion

5.1 Interpretation and Practical Reading

The main finding is a conditional one. Heat and germination act on different antinutrients, dehulling acts on the coat, and the cheap step of soaking helps a little with several. A sequence of simple steps is

therefore more defensible than reliance on one, and it can be done in a household kitchen.

The trade-off between antinutrient and antioxidant is the least comfortable result. A compound that lowers iron uptake in a meal can, in another context, be a useful antioxidant, and the evidence does not let us say which effect dominates in people (12, 13). Where mineral intake is the concern, as in diets low in animal foods, lowering the inhibitory compounds is sensible; where the diet is adequate, there is less reason to strip the seed. A cook reading a laboratory paper wants one thing: will this change what ends up on the plate?

5.2 Limitations

The evidence base has real weaknesses. Horse gram has been studied far less than soybean, common bean or chickpea, so much of what we say is inferred from other pulses, and mechanisms that hold across species may still differ in magnitude. Studies differ in cultivar, in method and in units, and the assays for phytate and for tannin are not always comparable (2, 36). A reduction in a compound is often reported without a measure of whether the seed is better digested or the minerals better absorbed. The point is not that a study is poor, only that its result travels less far than its abstract suggests.

There is also the problem of reading. Laboratory values for antinutrients are commonly expressed in ways that confuse loss of soluble solids with destruction of the compound, and a soak water that is not analysed can hide the fate of what was lost (16, 35). As a narrative review without a systematic search, this article is open to selection bias, and the conclusions should be read in that light. A reduction that vanishes when the units are corrected is not a reduction a household would notice.



5.3 Directions for Future Work

Three needs stand out. First, studies of horse gram that name the cultivar, report on a dry-matter basis and analyse the processing water and discarded fractions as well as the seed. Second, studies that test realistic sequences of household steps and not only single treatments. Third, work that links the laboratory values to function, through in vitro digestibility, mineral dialysability or absorption in people, so that a lower number can be tied to a better outcome (25, 34).

6. Conclusion

Processing can lower the main antinutritional factors in horse gram, but no single method lowers all of them, and the evidence for how much it helps people is thin. Heat deals with protease inhibitors and lectins, soaking and germination help with phytate and oligosaccharides, and dehulling lowers the seed-coat polyphenols at some cost in fibre and minerals. Readers should be wary of any claim of a large nutritional gain that does not rest on measures of digestibility or absorption.

For the household, the message is practical rather than discouraging. Soak, discard the water, consider sprouting, and cook until soft; all four are cheap and each does something different. For the researcher, the opportunity is a set of carefully reported studies of horse gram that name the cultivar, track the discarded fractions and link the laboratory result to a functional outcome. These points apply with equal force to any pulse, not only to horse gram. It is a habit worth keeping even when the answer seems obvious.

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9. Conflict of Interest

The authors declare no conflict of interest.

10. Author Contributions

S.P. conceived the review and wrote the sections on composition and mineral availability. M.H. wrote the sections on fermentation and germination. I.B. wrote the sections on thermal processing and study appraisal. All authors revised the manuscript and approved the final version.

11. Ethics Statement

This is a review of published literature. No human participants or animals were involved and no ethical approval was required.

12. Data Availability

No new data were generated. All sources are cited in the reference list.

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