RESEARCH ARTICLE

Influence of extrusion cooking on physicochemical properties and starch digestion kinetics of *Sphenostylis stenocarpa*, *Cajanus cajan*, and *Vigna subterranean* grains

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Abstract

Thermal degradation of sugars and amino acids, and depolymerization of macromolecules such as starch, proteins and fibre occasioned by high-temperature short-time extrusion cooking modify the physicochemical and functional properties of raw materials. High-temperature short-time extrusion cooking holds promise for the expanded use of non-conventional ingredients as food/feed due to its practicality, increased productivity and efficiency, and ability to retain thermally degradable nutrients during cooking. However, little is known about the effect of the high-temperature short-time extrusion cooking process on the physicochemical properties and starch digestibility of lesser-known grain legumes such as African yam beans (*Sphenostylis stenocarpa*), Pigeon pea (*Cajanus cajan*), and Bambara peanut (*Vigna subterranean*). In this study, we investigate the effect of high-temperature short-time extrusion cooking and extrusion cooking temperature; low (100˚C) vs high (140˚C) temperatures in a single screw extruder, on hydration characteristics, viscoamylolytic properties, *in vitro* starch digestibility and digestion kinetics of these grain legumes. We show that water holding capacity and swelling power increased (*p* < 0.05) with increasing extrusion temperature for *Sphenostylis stenocarpa* and *Vigna subterranean* but not *Cajanus cajan* extrudates. Significant effects of extrusion cooking (i.e unextruded vs 100˚C and unextruded vs 140˚C) and extrusion temperatures (i.e. 100˚C vs 140˚C) on starch digestion kinetics were observed for *Sphenostylis stenocarpa* and *Vigna subterranean* but not *Cajanus cajan* extrudates. Significant effects of extrusion cooking (i.e unextruded vs 100˚C and unextruded vs 140˚C) and extrusion temperatures (i.e. 100˚C vs 140˚C) were observed in peak, trough, final and setback viscosities of all extrudates. Starch digestibility and digestion characteristics were modified with increase in extrusion temperature, however, no effect of extrusion temperatures (i.e. 100˚C vs 140˚C) on starch digestion kinetics was observed for *Sphenostylis stenocarpa* and *Vigna subterranean* except for hydrolysis index (34.77 vs 40.77%). Nutritional and physiological implications of extruded grain legumes in monogastric animal feeding were also highlighted. The Information presented herein will influence expanded use of extruded grain legumes as feed ingredients for intensive monogastric animal feeding.
Introduction

Legume grains are protein-energy rich seeds, members of the dicotyledonous family *Fabaceae* or *Leguminosae* commonly found in the tropical rain forests of the Americas and Africa [1]. Grain legumes have an 11–50% protein content [2], hence the emphasis on grain legumes as dietary sources of protein, especially among low-income populations. The energy yield of grain legumes range from 905–1804 kJ/100g [3] and complex carbohydrate constituents range from 65–72% on dry matter basis. Starch is the most abundant complex carbohydrate fraction (22–45% of legume grains), followed by non-starch polysaccharides ((NSPs) also loosely translated as dietary fibre) representing 10–20% of complex carbohydrates and the balance being sugars [4,5]. Oligosaccharides, polysaccharides (e.g. cellulose, hemicellulose, pectic polysaccharides, and resistant starch) and associated plant substances[6], constitute the NSPs of grain legumes. Oligosaccharides of grain legumes are hailed for their prebiotic activity i.e. their ability to selectively stimulate the proliferation of one or more beneficial bacterial species in the hindgut, as they confer benefits to colonic health to a greater degree than resistant starch [7]. Sucrose galactosides; raffinose, stachyose and verbascose, represent the most researched oligosaccharides and account for 31–76% of total sugars in grain legumes. Carbohydrates of grain legume origin are often termed “low glycaemic” as their consumption produce a smaller rise in blood glucose relative to a reference, often white bread or pure glucose, owing to their high soluble and insoluble fibre, and resistant starch content which range from 5–35% by weight [8,9]. The glycaemic index of foods is influenced by a range of factors which include their botanical origin, varietal differences, type of starch, physical structure of the carbohydrate (e.g. particle size, granular dimensions and presence of associated compounds in the food/feed matrix e.g. lipids, proteins, fibres and acids), extent of processing and storage, and degree of gelatinization of inherent starch. Glycaemic index ranges from 0–100, with grain legumes having a glycaemic index of $\leq 55$ and pure glucose = 100 [9–13].

Grain legumes also provide dietary sources of antioxidants such as polyphenols, flavonoids, isoflavones and anthocyanins, which protect body cells against free radicals by functioning as radical scavengers, reducing agents, potential complexes of pro-oxidant metals and quenchers of singlet oxygen [14]. Other bioactive compounds of grain legumes are enzyme inhibitors (e.g. serine protease inhibitors and amylase inhibitors), phytic acid, haemagglutinins, condensed tannins, and lectins, which possess antinutritional properties. The presence of these antinutritional bioactive compounds in grain legumes impair their extensive use in foods and feeds. However, when grain legumes are subjected to dry or hydrothermal heat processes, heat labile bioactives are deactivated, alienating the antinutritional properties attached to them [15–17].

In high-temperature short-time (HTST) extrusion cooking, a moisture, temperature, pressure and mechanical shear regime is applied to the grains/ingredients [18]. Compared with other hydrothermal/thermal processes such as roasting, boiling, microwaving, and autoclaving, extrusion cooking affords a continuous consolidated multi-operational (involving mixing, heating, pressure cooking and mechanical shearing) process which is energy efficient and produces a high-quality output [19]. Adoption of HTST extrusion by the food and feed industry has resulted in increased production of ready-to-eat snacks and cereals, precooked infant meals, meat and cheese analogues, dry pet foods, fish and livestock ingredients and concentrates using lesser known cereals and legumes [20–27]. This heat processing technique also modifies their molecular structures and nutritional profile. Notable modifications to carbohydrates of grain legumes accruing from HTST extrusion cooking influence their hydration and functional properties, enzyme susceptibility[28], glycaemic index/potential and starch digestibility [29]. Studies investigating the influence of HTST extrusion cooking on enzyme...
susceptibility in grain legumes have reported an increase of 32.05%, 43.70% and 58.27% in starch digestibility of Lentils (*Lens culinaris Medikus*) in twin screw extruders at 140, 160 and 180°C respectively [30] and 21–22% increased enzyme susceptibility in single screw extruded *Phaseolus vulgaris* L. at 150°C [31]. Increase in enzyme susceptibility of materials after HTST extrusion cooking is attributed to macromolecular degradation of their starch granules [32] i.e. reduction in relative molecular weight of starch polysaccharide chains resulting from shearing of glycosidic bonds in amyllopectin and amylose chains during the cooking process [32,33]. The combination of high temperature and moisture employed in extrusion cooking, also disrupts intragranular hydrogen bonds allowing for association of water with free hydroxyl groups and resulting in modified functional and rheological properties [34].

These physical, chemical and nutritional changes in foods and feeds occasioned by HTST extrusion cooking, hold implications for humans and monogastric (single-chambered stomach) animals who consume the bulk of extruded products. The monogastric gastrointestinal tract is involved in mechanical digestion (chewing or mechanical pulverization of food materials in the avian gizzard) and enzymatic digestion, moving digested food thorough the body, and absorbing the products of digestion. The process of extrusion cooking is known to considerably impact nutrient availability[35], palatability, rate and extent of digestion of carbohydrates [36] in monogastric animals, while destroying enzyme inhibitors and toxins, reducing wastage as well as improving food/feed shelf-life and hygiene [35,37].

Lesser known grain legumes such as *Sphenostylis stenocarpa*, *Cajanus cajan*, and *Vigna subterranean* have attracted little research into their response to HTST extrusion cooking, hence limiting their use in foods and feeds even within geographical locations that boast of local production. This study describes the influence of high-temperature short-time extrusion cooking temperatures on the hydration and viscoamylolytic properties of *Sphenostylis stenocarpa*, *Cajanus cajan*, and *Vigna subterranean*, and their susceptibility to enzymatic hydrolysis in a monogastric digestive system model. It further postulates nutritional and physiological implications of these modifications in monogastric animals.

### Materials and methods

**Legume flour preparation and extrusion cooking**

*Sphenostylis stenocarpa* (African yam beans), *Vigna subterranean* (Bambara groundnut) and *Cajanus cajan* (Pigeon pea), were sourced from a local grain supplier, cleaned and milled through a 1mm sieve mesh. Moisture was adjusted to 25g/100g sample and mixed manually before subjecting to high-temperature short-time extrusion cooking in a single screw laboratory-scale extruder. Extruder characteristics were: screw diameter= 18.5mm, screw length=304mm, screw speed—60 rpm, and temperature measured at the outlet die -100°C (low temperature treatment) and 140°C (high temperature treatment), respectively. The choice of extrusion conditions; moisture and extrusion temperatures was based on earlier studies [15,38]. After extrusion, samples were dried in a forced air oven at 50°C overnight [38] and subsequently milled and stored in polyethylene bags at 4°C.

**Hydration properties**

Water retention capacity (WRC) was determined by methods outlined by AACC [39], with slight modifications. Briefly, 10% (w/v) of unextruded and extruded samples was prepared in pre-weighed tubes, vigorously vortexed for 1 min, held at room temperature for 30 mins and subsequently centrifuged at 1590 × g for 15 minutes. The unabsorbed liquid was decanted, absorbed water was calculated by difference and presented as a percentage of the dry sample weight. Water solubility index (WSI) and swelling power (SP) were determined at 70°C by the methods of
Leach et al., [40] with slight modifications. Briefly, 10% (w/v) of unextruded and extruded samples was prepared, stirred for 30 minutes at 70°C and subsequently centrifuged at 7000 × g for 20 minutes. Supernatant was aspirated into a pre-weighed crucible and dried at 105°C for 24 hours, and the dry solids weighed at the end of the drying period. The swelling power of the samples was calculated as the ratio of the weight of moisture imbibed by the granules of the sample to the weight of the dry samples while solubility index was calculated as the ratio of the weight of soluble solids to the weight of the dry sample and presented as a percentage.

**Viscoamylolytic properties**

Viscoamylolytic indices; peak, trough, breakdown, setback and final viscosities, time at peak viscosity and pasting temperature—were determined in a Rapid Visco TM Analyzer RVA-4 (Newport Scientific Pty. Ltd., NSW, Australia) with the software program Thermocline for Windows according to AACC method 61-02-01 [39]. About 2g of unextruded or extruded legume grain flour (corrected to 14% moisture) and 25 mL distilled water were combined and stirred at a paddle speed of 960 rpm for the first 10 seconds then reduced to 160 rpm. Pasting profile adopted involved holding temperature at 50°C for 1 minute, raising to 95°C in 3.75 minutes and holding for 2.5 minutes, then cooling to 50°C in 3.75 minutes and holding for a final 5 minutes.

**Enzyme susceptibility test and starch digestion kinetics**

Digestion was simulated under total tract conditions consisting of a three step hydrolysis mimicking digestion in the mouth, stomach and small intestine within a closed system according to the method of Sopade and Gidley [41], and glucose concentration measured by glucometry (ACCUCHEK active, Roche Diagnostics GmbH, Mannheim, Germany) after incubation at 0, 15, 30, 60, 90, 120, 180 and 240 mins. Zero-hour was defined as the beginning of the small intestine simulation step, as this connotes the main site of starch hydrolysis in monogastric animals [38]. Glucose concentrations obtained were converted to starch by a correction factor of 0.9. Starch digestion kinetics was studied by fitting time point glucose concentration data to a first-order exponential model [42] using the SOLVER add-in function in EXCEL [43]. Area under the digestogram (AUC) and hydrolysis index (HI) for each flour was computed up to 180 mins of incubation, using white bread as a reference product. Predicted glycaemic index (pGI) was also calculated for each flour [44].

**Calculations and statistical analysis**

All determinations were conducted in quadruplicate except the RVA determination which was conducted in duplicate. Data was subjected to one way analysis of variance (ANOVA), contrasts to assess the effects of extrusion (unextruded vs 100°C extrudates and unextruded vs 140°C extrudates) and extrusion temperatures (100°C extrudates vs 140°C extrudates) were done using non-orthogonal contrast (SPSS version 20), and means separated at P<0.05.

**Results**

Proximate composition of the raw and extruded *Sphenostylis stenocarpa* (African yam beans), *Vigna subterranean* (Bambara groundnut) and *Cajanus cajan* (Pigeon pea) products are shown in **Table 1**, and have been discussed in detail in a previous publication [15].

**Hydration properties**

Hydration properties of any plant-based material provides an evaluation of the water affinity of their functional polymeric components which is primarily starch. The hydration capacities
of unextruded and extruded flours of *Sphenostylis stenocarpa*, *Cajanus cajan*, and *Vigna subterranean* are presented in bar charts in Figs 1–3.

Water retention capacity (WRC) measures the ability of food/feed ingredients to bind, imbibe and retain water within their matrix. Extrusion cooking at 100°C and 140°C significantly increased WRC by 52.69% and 116.6% respectively, in *Sphenostylis stenocarpa*, 75.73% and 105.9% respectively, in *Vigna subterranean* and 13.98% and 5.08% respectively, in *Cajanus cajan*. Increase in extrusion cooking temperature from 100°C to 140°C also significantly increased WRC by 41.71% in *Sphenostylis stenocarpa*, and 17.21% in *Vigna subterranean*, whilst decreasing WRC by 7.64% in *Cajanus cajan*. Swelling power measures granular swelling accruing to intergranular and intragranular imbibition of water, and was measured at 70°C in the current study. On the other hand, solubility index indicates the extent of “melting” occurring when extrusion cooking is employed.

### Table 1. Proximate composition of raw and extruded *Sphenostylis stenocarpa* (African yam beans), *Vigna subterranean* (Bambara groundnut) and *Cajanus cajan* (Pigeon pea) flours†.

|                        | Crude protein (%) | Crude fibre (%) | Ether extract (%) | Nitrogen-free extract % | Ash (%) |
|------------------------|-------------------|-----------------|-------------------|-------------------------|---------|
| African yam beans      |                   |                 |                   |                         |         |
| AYR                    | 20.54 ± 0.05      | 4.38 ± 0.20     | 10.25 ± 1.08      | 52.52 ± 0.95            | 2.95 ± 0.00 |
| AY100                  | 20.21 ± 0.02      | 4.83 ± 0.15     | 4.62 ± 2.04       | 58.74 ± 2.07            | 2.90 ± 0.05 |
| AY140                  | 18.22 ± 0.01      | 4.28 ± 0.30     | 2.15 ± 0.50       | 63.53 ± 0.17            | 3.18 ± 0.13 |
| Bambara groundnut (Vigna subterranean) |                    |                 |                   |                         |         |
| BBR                    | 20.50 ± 0.01      | 4.03 ± 0.05     | 11.45 ± 1.28      | 51.42 ± 1.27            | 2.90 ± 0.00 |
| BB100                  | 20.27 ± 0.01      | 6.13 ± 0.46     | 5.15 ± 0.65       | 55.87 ± 0.87            | 3.63 ± 0.08 |
| BB140                  | 19.91 ± 0.01      | 6.03 ± 0.06     | 0.77 ± 0.18       | 60.36 ± 0.16            | 3.63 ± 0.03 |
| Pigeon pea (Cajanus cajan) |                    |                 |                   |                         |         |
| PPR                    | 21.20 ± 0.02      | 7.72 ± 0.25     | 8.50 ± 3.10       | 48.59 ± 2.74            | 4.88 ± 0.03 |
| PP100                  | 20.07 ± 0.00      | 8.02 ± 0.02     | 3.37 ± 0.16       | 55.58 ± 0.61            | 4.50 ± 0.20 |
| PP140                  | 18.66 ± 0.01      | 9.35 ± 0.13     | 2.03 ± 0.81       | 55.60 ± 0.69            | 5.02 ± 0.12 |

Values are presented as means of triplicate determinations ± standard deviation.

AYR—unextruded African yam beans (*Sphenostylis stenocarpa*); AY100—African yam beans (*Sphenostylis stenocarpa*) extruded at 100°C; AY140—African yam beans (*Sphenostylis stenocarpa*) extruded at 140°C; BBR—unextruded Bambara groundnut (*Vigna subterranean*); BB100—Bambara groundnut (*Vigna subterranean*) extruded at 100°C; BB140—Bambara groundnut (*Vigna subterranean*) extruded at 140°C; PPR—unextruded Pigeon pea (*Cajanus cajan*); PP100—Pigeon pea (*Cajanus cajan*) extruded at 100°C; PP140—Pigeon pea (*Cajanus cajan*) extruded at 140°C.

†[15].

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**Fig 1.** Hydration properties associated with HTST extrusion of African yam beans (*Sphenostylis stenocarpa*) at different temperatures. AYR—unextruded African yam beans (*Sphenostylis stenocarpa*); AY100—African yam beans (*Sphenostylis stenocarpa*) extruded at 100°C; AY140—African yam beans (*Sphenostylis stenocarpa*) extruded at 140°C.

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within the crystalline regions of starch. Swelling power in *Sphenostylis stenocarpa* extrudates increased with increased extrusion temperature and ranged from 11.93–108.9% over the unextruded *Sphenostylis stenocarpa*, with an 87.73% higher SP recorded in 140˚C extrudates compared to 100˚C extrudates of *Sphenostylis stenocarpa*. A significantly higher increase in SP was observed at 100˚C extrusion than at 140˚C i.e. 72.19% and 157.3% respectively, over SP of the unextruded *Vigna subterranean* flour, and a 49.36% decline in SP due to extrusion at the high temperature (BB100 vs BB140; p = 0.001). In contrast, No effect of extrusion cooking at 100˚C or 140˚C was observed on SP (PPR vs PP100 = 0.09 and PPR vs PP140 = 0.65), however, a 143.4% higher SP was recorded for when *Cajanus cajan* was extruded at 140˚C compared to 100˚C extrudates (PP100 vs PP140; p = 0.04). Extrusion cooking or extrusion cooking
temperatures did not affect WSI of *Sphenostylis stenocarpa* and *Cajanus cajan* products. However a marginal reduction (-5.11%) in response to extrusion cooking at 100°C (BBR vs BB100; *p* = 0.03) as well as a marginal increase of +4.84% in response to extrusion cooking temperatures (BB100 vs BB140; *p* = 0.03) were observed, with similar WSI observed in BBR and BB140 (*p* > 0.05).

Viscoamylolytic properties

The effect of HTST extrusion cooking temperatures on viscoamylolytic properties of unextruded and extruded flours of *Sphenostylis stenocarpa*, *Cajanus cajan*, and *Vigna subterranean* is presented in Table 2.

Significant decline in peak, trough, final and setback viscosities in response to extrusion cooking temperatures (i.e. unextruded vs 100°C and unextruded vs 140°C) was recorded for all grain legumes investigated, however, no significant effect (*p* > 0.05) of extrusion cooking temperatures was observed for breakdown viscosity. No significant effect (*p* > 0.05) of extrusion temperatures (i.e. 100°C vs 140°C) was also observed for all viscoamylolytic indices (i.e. peak, trough, breakdown, final and setback viscosities, pasting temperature and time at peak viscosity), measured in the current study. However, pasting temperature was significantly

| Viscoamylolytic indices (cP) | Peak viscosity (cP) | Trough viscosity (cP) | Breakdown viscosity (cP) | Final viscosity (cP) | Setback viscosity (cP) | Pasting temperature (°C) | Peak time (mins) |
|-------------------------------|-------------------|---------------------|------------------------|-------------------|----------------------|-------------------------|-----------------|
| AYR                           | 1029.5 ± 33.23    | 909.0 ± 57.98       | 120.5 ± 24.75          | 1282.5 ± 61.52    | 373.5 ± 3.54          | 82.3 ± 0.07             | 4.9 ± 0.19      |
| AY100                         | 100.5 ± 4.95      | 81.5 ± 3.54         | 19.0 ± 1.41            | 187.5 ± 9.19      | 106.0 ± 5.66          | 68.0 ± 2.83             | 6.9 ± 0.09      |
| AY140                         | 189.0 ± 38.18     | 163.0 ± 22.63       | 26.0 ± 15.56           | 282.5 ± 6.36      | 119.5 ± 16.26         | 55.8 ± 7.35             | 7.0 ± 0.00      |

Contrast P-values

| AYR vs AY100                  | *                | *                  | NS                     | *                | ***               | NS                     | *              |
| AYR vs AY140                  | **               | *                  | NS                     | *                | *                | 0.01                   | *              |
| AY100 vs AY140                | NS               | NS                 | NS                     | NS               | NS               | NS                     | NS             |
| PPR                           | 560 ± 8.49       | 546 ± 12.73        | 14.0 ± 4.24            | 760.0 ± 18.39    | 214.0 ± 5.67      | 86.73 ± 0.57           | 6.8 ± 0.28     |
| PP100                         | 42.0 ± 12.73     | 32.5 ± 7.78        | 9.5 ± 4.95             | 71.0 ± 11.31     | 38.5 ± 3.54       | 81.00 ± 0.00           | 7.0 ± 0.00     |
| PP140                         | 62.5 ± 4.95      | 53.0 ± 4.24        | 9.5 ± 0.71             | 93.5 ± 7.78      | 40.30 ± 5.54      | nd                     | 7.0 ± 0.00     |

Contrast P-values

| PPR vs PP100                  | ***              | ***               | NS                     | ***              | **                | NS                     | NS             |
| PPR vs PP140                  | **               | NS                | NS                     | NS               | NS               | NS                     | NS             |
| PP100 vs PP140                | NS               | NS                | NS                     | NS               | NS               | NS                     | NS             |
| BBR                           | 818.0 ± 42.43    | 791.5 ± 47.38     | 26.5 ± 4.95            | 1826.5 ± 70.00   | 1035 ± 22.63     | 85.2 ± 0.57           | 6.8 ± 0.00     |
| BB100                         | 134.5 ± 24.75    | 109.0 ± 19.80     | 25.5 ± 4.95            | 213.5 ± 20.51    | 104.5 ± 0.71     | 74.58 ± 3.47           | 7.0 ± 0.00     |
| BB140                         | 148.5 ± 19.09    | 126.0 ± 15.56     | 22.5 ± 3.54            | 249.0 ± 41.01    | 123.0 ± 25.46    | 70.58 ± 0.60           | 6.9 ± 0.09     |

Contrast P-values

| BBR vs BB100                  | *                | *                  | NS                     | *                | *                | **                     | NS             |
| BBR vs BB140                  | *                | *                  | **                     | ***              | **                | NS                     | NS             |
| BB100 vs BB140                | NS               | NS                 | NS                     | NS               | NS               | NS                     | NS             |

Values are presented as means ± standard deviation.

*** *P* ≤ 0.001

** *P* ≤ 0.01

* *P* < 0.05 NS—not significant, *P* > 0.05.

nd—not determined.

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reduced (p = 0.01) in 140˚C extrudates of *Sphenostylis stenocarpa*, and 100˚C extrudates of *Cajanus cajan* (p = 0.001). Extrusion of *Vigna subterranean* flours at both 100˚C and 140˚C significantly lowered pasting temperature (p = 0.01). Time at peak viscosity was significantly increased by extrusion at either 100˚C or 140˚C in *Sphenostylis stenocarpa* extrudates only.

### Starch digestion kinetics and *in vitro* starch digestibility

Table 3 and Figs 4–6 show the *in vitro* starch digestion pattern and starch digestion kinetics associated with HTST extrusion cooking of *Sphenostylis stenocarpa*, *Cajanus cajan*, and *Vigna subterranean* at different temperatures.

Generally, the HTST extrusion process modified starch digestion kinetics of the different grain legumes to varying degrees, hence the distinct differences in starch digestion patterns reflected in the digestograms. A two-fold increase in D₀, gastric digestion of starch (simulated mouth and stomach phases) in response to extrusion at 100˚C was recorded for *Sphenostylis stenocarpa* and *Vigna subterranean* but not *Cajanus cajan* flours. Starch rate constant (K) was unaffected by extrusion cooking at either temperatures, with no difference observed due to temperatures (100˚C vs 140˚C) except for *Cajanus cajan* flours. Area under the curve (AUC), hydrolysis index (HI) and predicted glycemic index (pGI) were significantly modified by

|                | D₀ (g/100g dry starch) | K × 10⁻³ (min⁻¹) | AUC × 10³ (mg dL⁻¹ 240 min⁻¹ | HI | pGI |
|----------------|------------------------|------------------|------------------------------|----|-----|
| AYR            | 1.44 ± 0.14            | 75.44 ± 18.89    | 1.32 ± 0.17                  | 6.94 ± 0.79 | 44.81 ± 0.49 |
| AY100          | 2.82 ± 0.85            | 83.27 ± 21.47    | 7.39 ± 0.46                  | 38.89 ± 3.72 | 70.47 ± 2.79 |
| AY140          | 2.43 ± 0.96            | 75.16 ± 29.87    | 7.07 ± 0.73                  | 37.25 ± 5.00 | 68.73 ± 3.68 |
| AY100 vs AY140 | *                      |                  | NS                           | ***           | ***           |
| AY100 vs AY140 | NS                     |                  | NS                           | ***           | ***           |
| PPR            | 1.37 ± 0.31            | 98.55 ± 26.94    | 1.17 ± 0.27                  | 6.16 ± 1.54  | 44.28 ± 1.27 |
| PP100          | 1.61 ± 0.53            | 94.72 ± 9.25     | 5.18 ± 0.91                  | 27.10 ± 3.99 | 61.25 ± 3.29 |
| PP140          | 3.90 ± 0.97            | 73.78 ± 12.95    | 8.83 ± 0.46                  | 46.42 ± 3.11 | 76.34 ± 3.01 |
| PPR vs PP100   | NS                     |                  | NS                           | ***           | ***           |
| PPR vs PP140   | ***                    |                  | NS                           | ***           | ***           |
| PP100 vs PP140 | ***                    |                  | NS                           | ***           | ***           |
| BBR            | 1.37 ± 0.16            | 71.26 ± 27.15    | 1.16 ± 0.29                  | 6.06 ± 1.45  | 44.07 ± 1.14 |
| BB100          | 2.67 ± 0.68            | 87.42 ± 15.12    | 6.60 ± 0.45                  | 34.77 ± 3.62 | 67.34 ± 2.87 |
| BB140          | 3.55 ± 1.01            | 75.19 ± 11.46    | 7.78 ± 1.06                  | 40.77 ± 4.52 | 71.86 ± 3.67 |
| BBR vs BB100   | *                      |                  | NS                           | ***           | ***           |
| BBR vs BB140   | *                      |                  | NS                           | ***           | ***           |
| BB100 vs BB140 | NS                     |                  | NS                           | ***           | NS            |

Values are presented as means ± standard deviation.

* *** P ≤ 0.001
* ** P ≤ 0.01
* * P < 0.05—NS—not significant, P > 0.05.

D₀—Digested starch at time, t = 0 (equivalent to very rapidly digesting starch [42], g/100g dry starch); K—rate constant (min⁻¹); AUC—area under the digestogram (mg dL⁻¹·240 min⁻¹); HI—hydrolysis index; pGI—predicted glycemic index [44].

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extrusion cooking at 100˚C and 140˚C (p = 0.001) in Sphenostylis stenocarpa, Cajanus cajan and Vigna subterranean flours. However, while a significant effect of increased extrusion temperature (100˚C vs 140˚C) on AUC, HI and pGI was recorded in Cajanus cajan flours, no significant (p > 0.05) response to increased temperature was observed on these indices in Sphenostylis stenocarpa flours. No effects of extrusion temperatures (100˚C vs 140˚C) on AUC and pGI was observed in Vigna subterranean flours. Starch digestibility of Sphenostylis stenocarpa, Cajanus cajan and Vigna subterranean flours in vitro was significantly increased by

Fig 4. Digestion pattern associated with HTST extrusion of African yam beans (Sphenostylis stenocarpa) at different temperatures. AYR—unextruded African yam beans (Sphenostylis stenocarpa); AY100—African yam beans (Sphenostylis stenocarpa) extruded at 100˚C; AY140—African yam beans (Sphenostylis stenocarpa) extruded at 140˚C.

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Fig 5. Digestion pattern associated with HTST extrusion of Bambara groundnut (Vigna subterranean) at different temperatures. BBR—unextruded Bambara groundnut (Vigna subterranean); BB100—Bambara groundnut (Vigna subterranean) extruded at 100˚C; BB140—Bambara groundnut (Vigna subterranean) extruded at 140˚C.

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extrusion cooking at 100˚C and 140˚C (p = 0.001). Increased extrusion temperature (100˚C vs 140˚C) also significantly increased in vitro starch digestibility in *Cajanus cajan* and *Vigna subterranean* flours but not in *Sphenostylis stenocarpa* flours (Figs 7–9).

Discussion

Hydration properties have been successfully used in the screening of feed materials for different botanical origin/source [45,46] and varietal [47–49] differences, effects of processing techniques as well as effects of extrusion variables such as processing temperature, moisture [38], and mechanical stress. Increased water absorption potential of legume induced by extrusion have been reported for common bean (*Phaseolus vulgaris*), pinto and navy beans and legume pasta of faba beans, lentils and black gram beans extruded at 80–180˚C [31,50–52] and range
from 30.5–165% in these grain legumes. Similar extrusion-induced decrease in water solubility of extruded legumes as recorded for extrudates of *Vigna subterranean* flour in the current study were also reported for HTC *Phaseolus vulgaris* extrudates as temperature increased from 100–180˚C [53,54]. Extrusion-induced decrease in water solubility of extruded legumes is perceived to be due to decreased protein solubility due to protein denaturation, which ultimately reflects on overall water solubility of the extruded legume product [54,55]. Howbeit in other studies, extrusion cooking seemed to increase water solubility of navy and pinto bean flours by 81.6% and 109.5% respectively [52], and common beans by 15.88–17.91% [31]. Extrusion-induced modifications to hydration properties of legumes have been attributed to dextrinization of the starch granule which is governed by starch and fibre content, swelling power and strength of protein network [50] of the grain legume.

The RVA pasting parameters provide a relative measure of extent of gelatinization, disintegration, swelling and gelling of the starch component of the feed material attributable to extrusion and extrusion cooking temperatures. Similar decrease in RVA peak and final viscosity was
reported for cowpeas in response to extrusion at 124–160˚C [56] and navy and pinto beans extruded at 85˚C, with a decrease in pasting temperature and no significant effect on breakdown viscosity [52]. Setback viscosity which implies degree of re-association, retrogradation and reordering of cooked starch after cooling was also significantly decreased in cowpea extrudates [56] at different extrusion temperatures as in the current study, indicative of little or no deterioration of product starch quality due to extrusion cooking at both temperatures. The combination of heat, mechanical shear and pressure, breakdown intermolecular hydrogen bonding within the starch structure causing “melting” in extruded legume starches at lower temperatures than native legume starches [34,57]. Higher protein content of grain legumes and their availability to form protein-starch matrices are considered responsible for the absence or unresponsiveness of breakdown viscosity in grain legumes [52]. Starch digestibility in vitro is known to correlate well with in vivo digestibility [58], and extrusion-induced increase in starch digestibility reported to increase with temperature and to varied degrees in different grain legumes [28,30]. A study by Masoero et al., [59] on the effect of extrusion on in vitro starch digestibility of peas, faba beans and lupins showed a higher extrusion-induced increase (4.5, 11.4 and 3.4 folds, respectively) in α-amylase susceptibility of the grain legumes at the 30mins time point compared to 120 mins time point (3.4, 7.47 and 1.8 folds, respectively). This is indicative of an increase in rapidly digestible starch fractions due to extrusion cooking conditions, and similar to the marked “steepness” in the digestograms of Sphenostylis stenocarpa, Cajanus cajan, and Vigna subterranea extrudates compared to their raw flours in the current study.

**HTST extrusion of grain legumes: Nutritional and physiological implications for monogastric farm animals**

Besides being protein sources in feeds, grain legumes contain significant complex carbohydrates; starch and NSPs, which have the potential to influence the dynamics of digestion and nutrient absorption in monogastric animals in diverse ways. Typical diets of monogastric farm animals consist of about 50% carbohydrates [60], with starch contributing 80–90% by weight [5] and more than 50% of the apparent metabolizable energy [36,61,62]. Based on in vitro measurements, nutritional research classifies starch into three fractions based on their digestion rate within the small intestine; rapidly digesting starch (RDS), slowly digestible starch (SDS) and resistant starch (RS). Typically, raw grain legumes contain 10.02–25.26% RDS, 21.84–39.64% SDS and 43.57–67.61% RS [63], which makes them poorly digested in the monogastric stomach. However, extrusion cooking has been shown to increases the proportion of RDS to SDS and RS in grain legumes [36] as a result of structural changes that occur in starch during extrusion which include gelatinization, melting and fragmentation [64]. This allows for increased starch digestibility in vivo, higher glycemic index and reduced gas production which is often observed with consumption of diets high in RS and “flatulence-causing” oligosaccharides such as raffinose and stachyose [65].

Although individual NSPs possess distinctly different chemical structures and functions, they are indigestible in the stomach and small intestinal segments of the gastrointestinal tract of monogastric animals due to an absence of fibrolytic enzymes that facilitate their digestion [60]. These NSPs are thus able to confer their unique hydration and rheological characteristics on the digesta, influencing the dynamics of digestion and nutrient absorption, and ultimately productivity of the animals. For instance, arabinoxylans and β-glucans are water soluble non-starch polysaccharides of significant importance in the nutrition of monogastric animals because they are present in viscous cereals, significantly wheat, rye, oat and barley [66–68]. When viscous cereals are used as the primary energy sources in the diets of chickens, the hydrocolloid forming ability of the inherent arabinoxylans and β-glucans modify the rheology of the digesta within
the gastrointestinal tract. The resulting viscous digesta within the gastrointestinal tract of monogastric animals fed diets high in soluble NSPs has been implicated in their reduced productivity [69] by: (a) depressing feed intake, decreasing interaction between ingesta and digestive enzymes [70], and reducing small intestinal nutrient absorption rates, (b) increasing endogenous secretions, (c) delayed transition of digesta through the gastrointestinal tract, and (d) proliferation of microorganisms in the distal small intestine of poultry and colon of pigs, with resultant gastroenteritis. Studies by Yaghobfor and Kalantar [69] observed a 36.48% and 22.64% increase in digesta viscosity, and an 8.42% and 7.89% decline in productivity (measured as feed conversion ratio) of chickens on wheat and barley-based diets respectively, compared to the control chickens on a maize-based diet. These authors also reported significant proliferation of the bacterial species; *E. coli* and Clostridia and decline in Bifidobacteria populations in chickens on the wheat and barley-based diets. Similar trends in response to water soluble NSPs in poultry and pigs have also been reported by other researchers [71–73]. Grain legumes on the other hand are low in water soluble NSPs [68] with their predominant NSPs being insoluble NSPs; xyloglucans and galactomannans, which do not lend to increased viscosity [70] in the gastrointestinal tract. The process of extrusion cooking further lowers the rheological properties of *Sphenostylis stenocarpa*, *Cajanus cajan*, and *Vigna subterranean* extrudates, alluding to their potential for maintaining sound gut health and productivity in monogastric farm animals.

The tendency for starch retrogradation -i.e. the re-association of disaggregated amylase and amylopectin chains of gelatinized starch [74]- in a starch paste is implied by the setback viscosity index and largely dependent on time and storage temperature [74,75]. Retrogradation is associated with adverse effects on starch digestibility in monogastric animals and sensorial properties [76]. The higher lipid content of grain legumes may provide a supply of free-lipids which could retard starch retrogradation during extrusion and/or storage by constraining the mobility of amylase [77], formation of lipid-amylose complexes and lipid-amylopectin complexes with outer branches of amylopectin [74,78]. The limited tendency of grain legumes to retrograde may be beneficial for feed quality in storage and effectively retain the increased starch digestibility attained by extrusion cooking of grain legumes. Overall, the hydration characteristics, viscoamylolytic properties and *in vitro* starch digestibility of starchy feed ingredients correlate well with starch digestibility within the gastrointestinal tract of monogastric animals [58], with documented evidence of extrusion-induced increase in starch digestibility of grain legumes available for poultry [79] and swine [36].

In summary, extrusion cooking at either 100˚C or 140˚C modified water retention capacity and swelling power of African yam beans (*Sphenostylis stenocarpa*), Pigeon pea (*Cajanus cajan*), and Bambara peanut (*Vigna subterranean*), but not water solubility index of *Sphenostylis stenocarpa* and *Cajanus cajan* extrudates. Extrusion cooking at either 100˚C or 140˚C also significantly decreased peak, trough, final and setback viscosities of extrudates, with no observed differences in viscosity indices when extrusion temperatures were compared. Starch digestibility also increased with extrusion temperature, even as rapidly digestible starch fractions were markedly increased in all grain legumes investigated. Findings from this research encourage further research into the use of “lesser known grain legumes” as food and feedstuffs, which could drive increased cultivation and improvement of these species.

**Novelty statement**

This research sheds light on changes observed in physicochemical and viscoamylolytic properties, and enzyme susceptibility, digestion kinetics plus glycemic index of starches of grain legumes; *Sphenostylis stenocarpa*, *Cajanus cajan*, and *Vigna subterranean*, in response to high-temperature short-time extrusion cooking at different temperatures.
Supporting information

S1 Table. (TIF)

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