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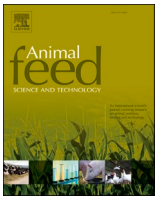

Effects of dry ensiling and toasting on nutrient concentrations, antinutritional compounds, and the formation of Maillard polymers in field pea grains

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ABSTRACT

The effects of dry ensiling of field pea grains and toasting of the ensiled pea grains on crude nutrient concentrations and the energy value, starch morphology, protein fractions and solubility, formation of Maillard polymers, and trypsin inhibitor activity were studied. A total of 27.3 t pea grains, harvested at 786 g dry matter (DM)/kg, was ensiled in a silage bag for 9 months using lactic acid bacteria inoculants. The ensiled grains were toasted using a mobile toaster with a grain temperature graduated between 60 and 110 °C. The dry silage had a pH of 6.1 and mainly lactic acid (3.7 g/kg DM) and acetic acid (1.8 g/kg DM) were produced. The dry silage was stable for at least 7 days under aerobic storage conditions. Dry ensiling led to a reduction of acid detergent fibre, whereas the sugar concentration increased 1.7-fold ($P < 0.05$). Subsequent toasting increased the fibre fractions and decreased sugar and non-fibre carbohydrates concentration, starting at 80 °C grain temperature ($P < 0.05$). The crude protein (CP) concentration remained unaffected. Estimated concentrations of metabolizable energy (ME) for dairy cows and horses, ME for swine (MES), or nitrogen-corrected apparent ME (AMEN) were not affected by dry ensiling. Toasting mainly affected AMEN, which decreased by 1 MJ/kg DM between 60 and 110 °C grain temperature. Dry ensiling and toasting did not or just marginally alter starch granule morphology.

Abbreviations: A, non-protein nitrogen; ADFom, acid detergent fibre; ADL, acid detergent lignin; AEE, acid ether extract; AMEN, nitrogen-corrected apparent ME; aNDFom, neutral detergent fibre; Arg, arginine; B1, TP soluble in borate-phosphate buffer; B2, TP insoluble in borate-phosphate buffer minus TP insoluble in neutral detergent; B3, TP insoluble in neutral detergent but soluble in acid detergent; C, TP insoluble in acid detergent; CA, crude ash; CF, crude fibre; CML, carboxymethyllysine; CNCPS, Cornell Net Carbohydrate and Protein System; CP, crude protein; CP_{ip}, CP insoluble in pepsin; DM, dry matter; E, ensiled; ESOM, enzyme-soluble organic matter; FL, fructoselysine; GT, grain temperature; HPLC, high performance liquid chromatography; Lys, lysine; ME, metabolizable energy; MES, ME estimated for swine; MG-H1, methylglyoxal hydroimidazolone; N, native; NFC, non-fibre carbohydrates; NFE, nitrogen-free extract; Pyr, pyrraline; RMSE, root mean square error; RUP, rumen-undegraded protein; SAT, supplied air temperature; SP, soluble protein; TIA, trypsin inhibitor activity; TP, true protein; TR, throughput rate..

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Ensiling and toasting distinctly changed CP fractions. Following dry ensiling, soluble protein was reduced from 73 % to 33 % of CP ($P < 0.01$). Toasting led to a further decrease in soluble protein to 11 % of CP ($P < 0.05$). Toasting increased irreversibly insoluble parts of CP, starting at 80 °C grain temperature ($P < 0.05$). Lysine was 11.6 g/kg DM in native and 9.6 g/kg DM in ensiled pea grains ($P < 0.01$). Fuctoselysine, carboxymethyllysine, and pyrraline increased 26- ($P > 0.05$), 28- ($P > 0.05$), and 8-fold ($P < 0.01$) after dry ensiling, respectively. Toasting decreased lysine starting at 80 °C ($P < 0.05$). Lysine adducts specifically changed, with pyrraline massively increased at any stage of temperature increase ($P < 0.05$). Arginine was 7.6 g/kg DM in native peas. The arginine adduct methylglyoxal hydroimidazolone was tripled after dry ensiling ($P < 0.01$) and increased throughout toasting temperature stages ($P < 0.05$). The native pea grains contained 3.8 g inhibited trypsin/kg DM, which only toasting reduced starting at 80 °C ($P < 0.05$). To avoid fixing the pea protein, a maximum temperature of 80 °C in the grain should not be exceeded, which, however, limits meaningful application of heat treatments.

1. Introduction

Among other pulses, field peas (*Pisum sativum*) are valuable protein suppliers for human and animal nutrition. They can be grown locally in many areas of the world, which helps to reduce imports of plant proteins such as from soybeans. This might have both economic and ecological benefits (Khan et al., 2016). Pea production levels are variable (FAOSTAT, 2024). In 2022, the production of dry peas was 14.2 Mt (on 7.2 Mha) worldwide and 6.0 Mt (on 2.6 Mha) in Europe (FAOSTAT, 2024). Field pea grains are typically characterised by crude protein (CP) concentrations of 180 to more than 250 g/kg dry matter (DM) (NRC, 2001; Bachmann et al., 2020a), which is about half of the CP of soybean or rapeseed meals (Konishi et al., 1999; NRC, 2001; Getachew et al., 2004). The storage proteins of pea grains are mainly composed of globulins, namely 7S vicilin and 11S legumin (Duranti, 2006), which is why 74–78 % of pea CP is readily soluble in contact with intestinal fluids (NRC, 2001; Bachmann et al., 2019). Pea grains may have about 478 g starch/kg DM (DLG, 1997). This is a multiple of the starch concentration of soybean or rapeseed meals and makes peas a unique dual-purpose feed, rich in both protein and energy (Schroeder, 2002).

In dairy cows, up to a one-to-one substitution of soybean meal and a partial substitution of cereals did not depress feed intake and performance as long as equal levels of rumen-undegraded protein (RUP) have been considered in the ration (Corbett et al., 1995; Khorasani et al., 2001). Rumen-undegraded protein has a 15 %-share of the CP of pea grains, while it is 35 % in soybean meal and 25 % in rapeseed meal (DLG, 1997). Post-ruminal CP was tabulated with 187 g/kg DM (75 % of CP) in peas, 60 % in soybean meal, and 54 % in rapeseed meal (DLG, 1997). This might show greater production of protein by rumen microorganisms due to the surplus of energy provided by the peas. In beef calves, creep feeding allows inclusion of peas up to more than 60 % without detrimental effects on ruminal fermentation, digestion, or performance (Gelvin et al., 2004). Peas have been successfully used also in growing and finishing beef cattle with 40 %-inclusion level, or in gestating cows (Soto-Navarro et al., 2012). In piglets and growing-finishing pigs, peas may be used with 15 up to 36 % of the diet without negative effects on performance provided the diets are balanced for amino acids, especially methionine and tryptophan (Gatel and Grosjean, 1990; Stein et al., 2004). Also in lactating sows, partial replacement of 30 % of the soybean meal by peas is possible (Landblom, 2002). However, the reports in breeding sows are controversial (Gatel and Grosjean, 1990). Pea grains may contain relevant quantities of antinutritive constituents such as tannins, lectins, or saponins, but mostly protease inhibitors with a specific trypsin and chymotrypsin inhibiting activity (Landblom, 2002; Robinson and Domoney, 2021; Ohm and Südekum, 2024). The latter may decrease protein digestibility and cause pancreatic hypertrophy (Landblom, 2002). For the application of peas in diets for pigs, it is recommended to choose cultivars with low concentrations of antinutritive substances. This may obviate the need for thermic processing (Hugman et al., 2020). In broiler chickens, replacement of soybean meal and maize with up to 48 % peas did not impair animal performance or carcass quality traits (Dotas et al., 2014). Inclusion of field peas into layer diets at 25 % was not negative for production as well (Perez-Maldonado et al., 1999). One experiment has shown that pea grains can partly replace oats in rations for growing horses without any adverse effects on growth or performance and without any noticeable digestive disorders (Landblom et al., 2005). The horses showed a good acceptance of the peas in a pelleted mixed feed (Landblom et al., 2005).

Ensiling is a common method of the preservation of feeds (Bolsen et al., 1996). Dry silage is when the material is ensiled with 550–860 g DM/kg. In classical wet silages, microbial fermentation alters nutrient concentrations and predominantly affects the pea protein. In whole-crop peas, wet ensiling hydrolyses the protein, which increases the proportion of soluble protein (SP) and predominantly of non-protein nitrogen (Mustafa et al., 2002; Cavallarin et al., 2007). By contrast, dry ensiling of pea grains was found to rather increase the insoluble protein fractions (Bachmann et al., 2020b). Ensiling marginally affects nutrient digestibility (Harris and Raymond, 1963; Aksu et al., 2004), but, especially in legume grains, it may reduce bioactive compounds with antinutritional properties (Gefrom et al., 2013; Goodarzi Boroojeni et al., 2017). The trypsin inhibitor activity (TIA) can be reduced by ensiling (Goodarzi Boroojeni et al., 2017). In the case of having cultivars with high native TIA, ensiling insufficiently reduces TIA, and additional thermic treatment can be required (Roy et al., 2010; Ma et al., 2011). However, in modern pea cultivars, the TIA is often very low and such cultivars may not require any heat treatment for TIA reduction. Thermic and hydro-thermic treatment of peas without previous ensiling or subsequent to ensiling may also increase resistant starch and RUP (Goelema et al., 1998; Mustafa et al., 1998; Yu et al., 2002; Ljøkjel et al., 2003; Masoero et al., 2005; Vaga et al., 2017; Bachmann et al., 2019). Heat treatment can also lead to protein damage. It is important to carefully consider when and to what extent heat treatment is necessary and useful. In this study, toasting

temperatures up to 110 °C were applied at the surface of the grains, which is lower than those that other working groups considered optimal to protect the protein from ruminal degradation (Ljøkjel et al., 2003). The temperature range was chosen to identify from which temperature on effects turn to be negative in the sense of protein damage.

The following hypotheses arose from the literature review: (i) dry ensiling of pea grains and the combination of dry ensiling and toasting (thermic treatment) do not distinctly change crude nutrient concentrations; (ii) dry ensiling and toasting decrease the solubility of CP; (iii) the formation of Maillard polymers provides one possible explanation for protein solubility reduction; and (iv) dry ensiling and toasting efficiently inactivate TIA starting from moderate to high levels in the native grains.

2. Materials and methods

2.1. Treatments

For the current study, a total of 27.3 t of grains of the field pea cultivar Alvesta (KWS SAAT SE, Einbeck, Germany) was harvested in 2017 in Köllitsch (Saxony) with 786 ± 7.08 g DM/kg. Before ensiling, 120 g of homo-fermentative lactic acid bacteria, i.e., *Lactobacillus plantarum* and *Pediococcus acidilactici* strains at together 6.8×10^6 colony forming units per g fresh matter (Josilac® classic; Josera GmbH & Co. KG, Kleinheubach, Germany) were diluted in 200 L water and added to the grains. The material was crushed using a portable Murska 2000 S2x2 roller mill (Murska, Ylivieska, Finland) and ensiled in a silage plastic bag (BAG Budissa Agroservice GmbH, Malschwitz, Germany) for a period of 9 months. Characteristics of the dry silage were as follows: pH 6.1, 3.7 g lactic acid, 1.8 g acetic acid, 0.18 g propionic acid, 0.12 g n-butyric acid, 0.33 g i-butyric acid, 0.13 g n-valeric acid, 0.07 g i-valeric acid, 0.20 g ethanol, 0.31 g 1,2-propandiol, and 0.47 g 1-propanol/kg DM. Aerobic stability of the dry silage lasted for more than 7 days.

The ensiled grains were subsequently toasted using a mobile toaster (EcoToast 100, Agrel GmbH, Arnstorf, Germany) at atmospheric pressure. Doing this, the supplied air temperature and the throughput rate of the material were graduated as shown in Table 1. The grain temperature is the temperature at the grain surface, measured with a sensor at the exit immediately after toasting. The pea grain dry silage was toasted in summer with 20 °C initial temperature of the material and 25 °C ambient temperature.

2.2. Chemical analyses

Samples of the native and treated peas were freeze-dried and ground to pass a 1-mm sieve of a standard laboratory sample mill. Dry matter, crude ash (CA), CP, acid ether extract (AEE), sugar, crude fibre (CF), and detergent fibres were analysed using methods no. 3.1, 4.1.1, 5.1.1 B, 6.1.1, 6.5.1, 6.5.2, 6.5.3, 7.1.1, and 8.1 of the Association of German Agricultural Inspection and Research Institutes (VDLUFA, 2023). Neutral detergent fibre (aNDFom) was assayed with heat-stable amylase added to the neutral detergent solution, and aNDFom and acid detergent fibre (ADFom) were expressed exclusive of residual ash. The proportion of nitrogen-free extract (NFE) was calculated as NFE = 1000 – CA – CP – AEE – CF. The proportion of non-fibre carbohydrates (NFC) was calculated as NFC = 1000 – CA – CP – AEE – aNDFom. Starch was determined using the amyloglucosidase method (method no. 7.2.5; VDLUFA, 2023). The enzyme-soluble organic matter (ESOM) was determined using method no. 6.6.1 (VDLUFA, 2023). The protein fractions defined by the Cornell Net Carbohydrate and Protein System (CNCPS) A – non-protein nitrogen, B1 – true protein (TP) soluble in borate-phosphate buffer at pH 6.7–6.8, but precipitable, B2 – TP insoluble in borate-phosphate buffer minus TP insoluble in neutral detergent, B3 – TP insoluble in neutral detergent but soluble in acid detergent, and C – TP insoluble in acid detergent, were determined according to Licitra et al. (1996). For each fraction, residual nitrogen was determined using the Kjeldahl method. The protein fractions were used to calculate TP, where TP = B1 + B2 + B3 + C and SP, where SP = A + B1. The protein that is insoluble in pepsin (CP_{ip}) was analysed using the Kjeldahl method after 48 h of incubation in a pepsin-hydrochloric acid solution according to Weissbach et al. (1985). In the pea silages, organic acids and alcohols were determined by high performance liquid chromatography (HPLC) and refractive index detection (method no. LKS FMUAA 1662018–05) using a Shimadzu LC-20A Prominence (Shimadzu Corp., Kyoto, Japan) and a Hi-Plex H 8-µm column (300 mm × 7.7 mm; Agilent Technologies Inc., Santa Clara, CA, USA). The method was accredited according to DIN EN ISO/IEC 17025:2005. The aerobic stability of the silages was tested according to the procedure proposed by Honig (1990). Maillard reaction products were determined by HPLC-mass spectroscopy as described in detail by Krause et al. (2003), Schwarzenbolz et al. (2016), and Hofmann et al. (2020). An Agilent 1200 series system connected to an Agilent 6410 mass spectrometer with electrospray

Table 1
Combinations of supplied air temperature (SAT) and throughput rate (TR) used in the mobile toaster, and the resulting graduation of grain temperature (GT).

SAT (°C)	TR (kg/h)	GT (°C)
100	100	60
140	100	70
160	100	75
160	100	77
170	100	80
180	100	85
190	100	90
190	70	100
200	50	110

ion source (Agilent Technologies Inc., Santa Clara, CA, USA) was used. Furosine was quantified after hydrolysis with 6 N hydrochloric acid by separation on a cation exchange resin column and post-column derivatisation with ninhydrin. From the analysed furosine, the Amadori product fructoselysine was calculated (Krause et al., 2003). Lysine and arginine were analysed by HPLC and ninhydrin post-column derivatisation following the European Union Commission Directive 98/64/EC. Trypsin inhibitor activity was determined according to Kakade et al. (1969).

2.3. Scanning electron microscopic imaging

Scanning electron micrographs of pea starch granules and their embedding, covering, or surrounding matrix structures have been recorded under vacuum using a JEOL 640 unit (JEOL Ltd., Tokyo, Japan) to visualize morphological changes that may follow dry ensiling and toasting. Priorly, pea grains were oven-dried at 40 °C, ground to approximately 1 mm sieve size, spread out on a microscope slide, and sputter coated with gold. The electron beam is generated by stimulating a wolfram cathode, usually with an excitation voltage of 20 keV. The starch granules, however, ruptured during visualizing, which is why the excitation voltage was reduced to 15 keV.

2.4. Calculations of the energy value

The energy value of native or treated peas was calculated exemplarily for application in dairy cows, fattening pigs, poultry, and horses using the following equations. The concentration of metabolizable energy (ME) in dairy cows was calculated as ME (MJ/kg DM) = 9.67 – 0.01698 CA + 0.0034 CP + 0.01126 AEE + 0.00123 starch – 0.00097 aNDFom + 0.0036 ESOM according to the Society of Nutrition Physiology (GfE, 2009). The concentration of ME for swine (MES) was calculated as MES (MJ/kg DM) = 0.021503 CP + 0.032497 AEE – 0.021071 CF + 0.016309 starch + 0.014701 (1000 – CA – (CP + AEE + CF + starch)) (GfE, 2008). The concentration of nitrogen-corrected apparent ME (AMEN) was calculated as AMEN (MJ/kg DM) = 0.01551 CP + 0.03431 AEE + 0.01669 starch + 0.01301 sugar according to the World's Poultry Science Association (WPSA, 1984). The concentration of ME for horses was calculated as ME (MJ/kg DM) = -3.54 + 0.0129 CP + 0.042 AEE – 0.0019 CF + 0.0185 NFE with a constant of 8 kJ/g CP renal energy loss according to Kienzle and Zeyner (2010) and as ME (MJ/kg DM) = -3.865 + 0.0166 CP + 0.042 AEE – 0.0019 CF + 0.0185 NFE with consideration of dynamic estimates of the renal energy loss according to Kuchler et al. (2020). All nutrient concentrations are considered as g/kg DM.

2.5. Statistical analysis

Statistical analysis was performed using SAS 9.4 (SAS Institute Inc., Cary, NC, USA). Effects of dry ensiling were analysed using the pooled *t*-test (with homogeneous sample variances) or the Satterthwaite *t*-test (with inhomogeneous sample variances). The homogeneity of sample variances was proven using the folded F-test. The effects of toasting temperature (i.e., the resulting grain temperature) were analysed using linear, quadratic, or linear plus quadratic regression with the MIXED and REG procedures. For each analyte, the best fitting model was identified using the Akaike Information Criterion (Akaike, 1974). Least squares means were estimated from the regression models for 5 °C-grain temperature stages within the measured temperature range (60–110 °C). Heterogeneous residual variances were considered according to temperature stages for CP_{ip}, according to grouped temperature stages (60–80 °C, 85–90 °C, 100–110 °C) for AEE, ADFom, NFC, ME, AMEN, and the CNCPS fraction C, or according to squared temperature stages for CA. For the remaining analytes, homogenous residual variances were considered. The studentized residuals were confirmed to have Gaussian distribution by Shapiro-Wilk and/or Kolmogorov-Smirnov test. The Pearson correlation between CNCPS fraction C and CP_{ip} was calculated using the CORR procedure. For all statistical tests, the level of significance was $P < 0.05$.

3. Results and discussion

3.1. Treatment effects on nutrient concentrations and the energy value

3.1.1. Ensiling effects on nutrient concentrations

The effect of dry ensiling on crude nutrient and fibre concentrations of the peas is shown in Table 2. The silage had a final DM concentration of 768 ± 7.82 g/kg. The lower DM concentration compared to the native material was due to the addition of water for the application of inoculants. No DM reduction was detected during ensiling. A significant reduction of the concentration has been detected in ADFom ($P < 0.05$). Total sugar concentration increased a 1.7-fold ($P < 0.05$).

3.1.2. Toasting effects on nutrient concentrations

The CF and detergent fibres increased during toasting, starting at grain temperatures of 80 °C (aNDFom), 95 °C (ADFom, ADL), and 110 °C (CF) ($P < 0.05$; Table 3). In contrast, total sugar and NFC concentrations decreased starting at 85 and 80 °C grain temperature, respectively ($P < 0.05$). Toasting did not alter the concentration of CA, CP, AEE, starch, and NFE (Table 3).

3.1.3. Ensiling effects on the energy value

The estimated ME concentrations in the pea grains were not affected by dry ensiling (Table 4).

Table 2
Mean concentration and standard deviation of crude nutrients, detergent fibres, starch, and sugar in native and ensiled field pea grains.

Pea treatment	Ash	CP	AEE	CF	aNDFom	ADFom	ADL	Starch	Sugar	NFE	NFC
Native	32 ± 0.48	190 ± 1.60	27 ± 4.7	60 ± 3.4	118 ± 14.7	83 ± 4.6[a]	6 ± 0.8	521 ± 8.93	39 ± 11[b]	691 ± 4.06	630 ± 11.5
Ensiled	33 ± 0.63	187 ± 1.63	23 ± 5.8	56 ± 2.5	119 ± 15.7	72 ± 0.87[b]	5 ± 2	530 ± 8.45	65 ± 6.1[a]	701 ± 6.78	639 ± 13.2

ADFom, acid detergent fibre expressed exclusive of residual ash; ADL, acid detergent lignin; AEE, acid ether extract; aNDFom, neutral detergent fibre assayed with heat stable amylase and expressed exclusive of residual ash; CF, crude fibre; CP, crude protein; DM, dry matter; NFC, non-fibre carbohydrates; NFE, nitrogen-free extract.
The analytes are given as g/kg DM.
[ab]Different superscripts indicate the difference between native and ensiled pea grains ($P < 0.05$).

Table 3
Effect of toasting ensiled field pea grains with a grain temperature graduated between 60 and 110 °C on concentrations of crude nutrients, detergent fibres, starch, and sugar.

Pea treatment	Ash	CP	AEE	CF	aNDFom	ADFom	ADL	Starch	Sugar	NFE	NFC
Ensiled + toasted (60 °C)	34	196	22	64	153	82	8	516	54	684	597
	(34 34)	(193 198)	(20 24)	(60 67)	(129 177)	(80 84)	(1 15)	(504 527)	(48 60)	(678 689)	(574 619)
Ensiled + toasted (65 °C)	34	195	21	62	165	78	7	514	52	687	585
	(34 34)	(193 197)	(20 23)	(60 64)	(145 186)	(77 80)	(2 11)	(504 524)	(47 57)	(683 691)	(566 604)
Ensiled + toasted (70 °C)	34	194	21	61	177	76	6	512	50	689	573
	(34 34)	(193 195)	(20 23)	(59 63)	(160 195)	(75 77)	(3 10)	(504 521)	(46 54)	(687 692)	(558 589)
Ensiled + toasted (75 °C)	34	193	21	61	190	76	7	511	48	691	562
	(34 34)	(192 195)	(20 22)	(59 62)	(175 204)	(75 77)	(4 10)	(503 518)	(44 51)	(688 693)	(549 575)
Ensiled + toasted (80 °C)	34	193	21	61	202	78	9	509	45	692	550
	(34 34)	(192 194)	(20 22)	(59 62)	(189 215)	(77 79)	(6 13)	(502 515)	(42 48)	(689 694)	(538 562)
Ensiled + toasted (85 °C)	34	193	21	61	214	81	13	507	42	692	539
	(33 34)	(191 194)	(20 22)	(60 63)	(201 227)	(79 83)	(9 16)	(500 513)	(39 46)	(689 695)	(526 551)
Ensiled + toasted (90 °C)	34	192	20	63	226	87	17	504	40	691	527
	(33 34)	(191 194)	(19 21)	(61 64)	(212 240)	(83 90)	(13 21)	(497 511)	(36 43)	(688 694)	(512 542)
Ensiled + toasted (95 °C)	33	192	20	64	238	94	23	502	37	690	515
	(33 34)	(191 194)	(19 21)	(63 66)	(222 255)	(88 99)	(19 26)	(494 510)	(33 41)	(687 693)	(497 533)
Ensiled + toasted (100 °C)	33	192	20	67	251	102	29	500	34	688	504
	(33 34)	(191 194)	(19 21)	(65 69)	(231 270)	(94 110)	(26 33)	(490 510)	(29 39)	(685 691)	(482 525)
Ensiled + toasted (105 °C)	33	192	20	70	263	113	37	497	30	685	492
	(33 34)	(191 194)	(18 21)	(67 72)	(240 286)	(102 124)	(32 42)	(485 509)	(24 36)	(681 689)	(466 518)
Ensiled + toasted (110 °C)	33	193	19	73	275	125	47	495	27	681	480
	(33 34)	(190 195)	(18 21)	(70 76)	(248 302)	(111 140)	(40 53)	(480 509)	(20 34)	(676 687)	(451 510)
RMSE	1.9	2.34	2.7	3.1	32.3	9.63	6.4	12.9	8.1	5.17	31.6
R^2	0.012	0.191	0.120	0.655	0.563	0.770	0.818	0.201	0.508	0.350	0.544

ADFom, acid detergent fibre expressed exclusive of residual ash; ADL, acid detergent lignin; AEE, acid ether extract; aNDFom, neutral detergent fibre assayed with heat stable amylase and expressed exclusive of residual ash; CF, crude fibre; CP, crude protein; DM, dry matter; NFC, non-fibre carbohydrates; NFE, nitrogen-free extract; RMSE, root mean square error.
Values are least squares means estimated from linear (AEE, aNDFom, NFC), quadratic (ash, starch, sugar), or linear plus quadratic regression (CP, CF, ADFom, ADL, NFE) at 5 °C-grain temperature stages within the measured temperature range; the 95 %-confidence intervals are given in brackets; the analytes are given as g/kg DM.

Table 4
Energy concentration of native and ensiled field pea grains exemplarily calculated for the feeding of dairy cows, fattening pigs, poultry, and horses.

	Dairy cows	Pigs	Poultry	Horses	
Pea treatment	ME	MES	AMEN	ME[a]	ME[b]
Native	14.1 ± 0.0739	14.8 ± 0.0168	13.1 ± 0.241	12.8 ± 0.0209	13.2 ± 0.0113
Ensiled	14.0 ± 0.0627	14.7 ± 0.0268	13.4 ± 0.232	12.7 ± 0.0928	13.1 ± 0.0974

AMEN, nitrogen-corrected apparent ME; ME, metabolizable energy; MES, ME in swine.
Values are given as means and standard deviation. All energy values are given as MJ/kg dry matter.
Differences between native and ensiled pea grains have not been found.
[a] Calculated with a constant of 8 kJ/g crude protein renal energy loss according to Kienzle and Zeyner (2010).
[b] Calculated with consideration of dynamic estimates of renal energy loss according to Kuchler et al. (2020).

3.1.4. *Toasting effects on the energy value*

Toasting the peas slightly decreased energy concentrations starting approximately at 80 °C grain temperature ($P < 0.05$; Table 5). The AMEN concentration was affected most by toasting. It was reduced by 1 MJ/kg DM between 60 and 110 °C grain temperature (Table 5).

The nutrient concentrations and the energy value of native, ensiled, and ensiled plus toasted field pea grains were within the ranges measured in previous and in the current project (Castell et al., 1996; Gefrom et al., 2013; Bachmann et al., 2019, 2020b, 2022; David et al., 2024). In the light of cultivar differences, the AMEN content of the peas we calculated was somewhat higher than that reported by other authors (Nalle et al., 2011). Ensiling with more than 700 g DM/kg preserved the pea grains in an anaerobic environment (Bachmann et al., 2022). In the current dry silages, lactic acid fermentation was largely suppressed and no consumption of sugar appeared. The pH was 6.1, despite the use of lactic acid inoculants. This meant that proteolytic and desmolytic processes were largely absent, but valuable nutrients were retained. Although ensiling in the classical sense was not achieved, the pea grains were sufficiently stable against aerobic storage. Effects of toasting on crude nutrient concentrations and energy content depend on the specific processing conditions. Since there are no standardized procedures, a direct comparison with literature data is not possible. In this study, a relative increase in fibre fractions was detected, which resulted from a reduction in total sugar concentration. The latter is probably associated with the excessive formation of Maillard reaction products at high temperatures. We did not find any effects of thermic treatment on the energy content, which confirms our previous observations (Bachmann et al., 2019).

3.2. *Effects on starch morphology*

Scanning electron micrographs of native, ensiled, and ensiled plus toasted field peas are shown in Fig. 1. Starch granules of native field pea grains had an oval, reniform, or irregular shape. The granules had smooth surfaces. Clear demarcations existed to the surrounding or covering matrix structures, which were visible as sharp-edged fragments. Fragmentation of matrices was a result of milling of the substrates before imaging. The pea starch granules have been found in a length range of 6.4–31.9 µm.

Table 5
Energy concentration of ensiled plus toasted field pea grains exemplarily calculated for the feeding of dairy cows, fattening pigs, poultry, and horses.

	Dairy cows	Pigs	Poultry	Horses	
Pea treatment	ME	MES	AMEN	ME[a]	ME[b]
Ensiled + toasted (60 °C)	13.9 (13.8 14.0)	14.6 (14.5 14.7)	13.1 (12.9 13.3)	12.5 (12.4 12.6)	12.9 (12.9 13.0)
Ensiled + toasted (65 °C)	13.9 (13.8 13.9)	14.6 (14.5 14.7)	13.0 (12.9 13.2)	12.5 (12.4 12.5)	12.9 (12.9 13.0)
Ensiled + toasted (70 °C)	13.8 (13.8 13.9)	14.5 (14.5 14.6)	12.9 (12.8 13.1)	12.5 (12.4 12.5)	12.9 (12.8 13.0)
Ensiled + toasted (75 °C)	13.8 (13.8 13.9)	14.5 (14.4 14.6)	12.9 (12.7 13.0)	12.4 (12.4 12.5)	12.9 (12.8 12.9)
Ensiled + toasted (80 °C)	13.8 (13.8 13.8)	14.5 (14.4 14.5)	12.8 (12.6 12.9)	12.4 (12.4 12.5)	12.8 (12.8 12.9)
Ensiled + toasted (85 °C)	13.8 (13.7 13.8)	14.4 (14.3 14.5)	12.7 (12.5 12.8)	12.4 (12.3 12.4)	12.8 (12.8 12.9)
Ensiled + toasted (90 °C)	13.7 (13.7 13.8)	14.4 (14.3 14.4)	12.6 (12.4 12.7)	12.4 (12.3 12.4)	12.8 (12.7 12.8)
Ensiled + toasted (95 °C)	13.7 (13.6 13.7)	14.3 (14.2 14.4)	12.5 (12.3 12.6)	12.3 (12.3 12.4)	12.8 (12.7 12.8)
Ensiled + toasted (100 °C)	13.7 (13.6 13.7)	14.2 (14.1 14.3)	12.3 (12.2 12.5)	12.3 (12.2 12.4)	12.7 (12.6 12.8)
Ensiled + toasted (105 °C)	13.6 (13.6 13.7)	14.2 (14.1 14.3)	12.2 (12.0 12.5)	12.3 (12.2 12.3)	12.7 (12.6 12.8)
Ensiled + toasted (110 °C)	13.6 (13.5 13.7)	14.1 (14.0 14.3)	12.1 (11.8 12.4)	12.2 (12.1 12.3)	12.6 (12.5 12.7)
RMSE	0.066	0.128	0.268	0.091	0.092
R^2	0.689	0.583	0.548	0.481	0.521

AMEN, nitrogen-corrected apparent ME; ME, metabolizable energy; MES, ME in swine; RMSE, root mean square error.
Values are least squares means estimated from linear (ME cows) or quadratic (MES, AMEN, ME horse) regression at 5 °C-grain temperature stages within the measured temperature range; the 95 %-confidence intervals are given in brackets. All energy values are given as MJ/kg dry matter.
[a] Calculated with a constant of 8 kJ/g crude protein renal energy loss according to Kienzle and Zeyner (2010).
[b] Calculated with consideration of dynamic estimates of renal energy loss according to Kuchler et al. (2020).

3.2.1. Ensiling effects on starch morphology

Dry ensiling did not visibly affect starch granule morphology.

3.2.2. Toasting effects on starch morphology

A slight disbanding of the sharp demarcations between starch granules and matrix structures became visible after toasting at 110 °C grain temperature, but overall, treatment effects were on a marginal level.

As long as moisture and pressure are absent during processing, distinct changes in starch structure will not appear (Xie et al., 2018). This is confirmed by the current results. Under the influence of heat and pressure, the starch granules gelatinise and may retrograde and become less digestible (Liu et al., 2018). Gelatinisation temperatures and enthalpy decrease with the increase of pressure (Liu et al., 2018). The processing of peas used in this study does not compel us to expect distinct changes of starch digestibility due to gelatinisation or retrogradation.

3.3. Effects on protein fractions and solubility

3.3.1. Ensiling effects on protein fractions and solubility

Ensiling distinctly affected protein fractions in pea grains (Table 6). True protein decreased ($P < 0.01$). Non-protein nitrogen (fraction A) increased by 3.3 %-points ($P < 0.01$). The soluble fraction B1 was markedly reduced (44 %-points; $P < 0.01$). The lesser soluble protein fractions B2 and B3 increased by 35 and 5.3 %-points, respectively ($P < 0.01$). The insoluble C fraction and CP_{ip} were on a very low level in the native grains (0.7 and 4.8 % of CP, respectively) and were not affected by dry ensiling (0.6 and 4.5 % of CP, respectively). These changes in the fractions of the protein resulted in a reduction of SP by at least 40 %-points ($P < 0.01$).

3.3.2. Toasting effects on protein fractions and solubility

The effect of toasting subsequent to dry ensiling is shown in Table 7. True protein and the fraction A were virtually not affected. Soluble protein decreased from 28 % to 11 % of CP within the 60–110 °C grain temperature range ($P < 0.05$), which was the result of an up to 16 %-points decrease of the B1 fraction ($P < 0.05$). Fraction B2 decreased by 33 %-points, while B3 increased by 29 %-points ($P < 0.05$). Toasting increased insoluble parts of the protein, starting at a grain temperature of 80 °C (fraction C) or 90 °C (CP_{ip}) ($P < 0.05$).

There were no changes in the CP concentration due to dry ensiling and toasting, but a clear shift from soluble to insoluble fractions

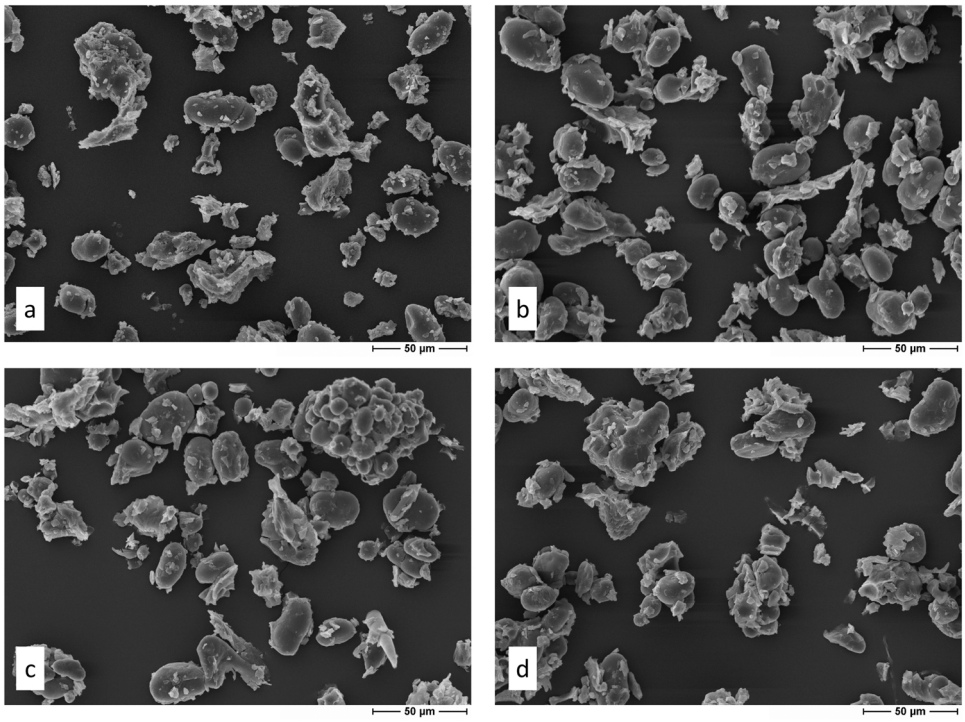

Fig. 1. Scanning electron micrographs of starch granules and their embedding, covering, or surrounding matrices in native field peas (a), ensiled field peas (b), ensiled field peas toasted with 90 °C grain temperature (c), and ensiled field peas toasted with 110 °C grain temperature (d), made by × 500 magnification and 15 keV. In native field pea grains, the starch granules had smooth surfaces and clear demarcations to the matrix structures visible as sharp-edged fragments. Dry ensiling did not affect starch granule morphology and even after toasting at 110 °C grain temperature, only a slight disbanding of the demarcations was observed.

Table 6
Mean concentration and standard deviation of TP, SP, CNCPS protein fractions, CP_{ip}, amino acids, Maillard reaction products, and inhibited trypsin in native and ensiled field pea grains.

Pea treatment	TP	SP	A	B1	B2	B3	C	CP_{ip}	Lys	Arg	FL	CML	Pyr	MG-H1	TIA
Native	178 ± 0.481[a]	73 ± 1.7[a]	6.2 ± 0.60[b]	67 ± 1.7[a]	25 ± 0.52[b]	1.5 ± 1.2[b]	0.65 ± 0.34	4.8 ± 0.83	11.6 ± 0.189[a]	7.6 ± 0.053	0.14 ± 0.014	0.79 ± 0.62	0.35 ± 0.17[b]	1.2 ± 0.72[b]	3.8 ± 1.2
Ensiled	169 ± 0.640[b]	33 ± 2.1[b]	9.5 ± 0.26[a]	23 ± 2.5[b]	60 ± 1.1[a]	6.8 ± 1.1[a]	0.57 ± 0.060	4.5 ± 0.77	9.6 ± 0.18[b]	7.8 ± 0.34	3.7 ± 0.54	21.8 ± 18.2	2.8 ± 0.88[a]	3.9 ± 0.65[a]	2.5 ± 0.35

A, non-protein nitrogen; Arg, arginine; B1, buffer-soluble TP; B2, buffer-insoluble TP minus TP insoluble in neutral detergent; B3, TP insoluble in neutral detergent but soluble in acid detergent; C, TP insoluble in acid detergent; CML, carboxymethyllysine; CNCPS, Cornell Net Carbohydrate and Protein System; CP, crude protein; CP_{ip}, CP insoluble in pepsin; DM, dry matter; FL, fructoselysine; Lys, lysine; MG-H1, methylglyoxal hydroimidazolone; Pyr, pyrraline; SP, soluble protein; TIA, trypsin inhibitor activity; TP, true protein.
Dry matter is given as g/kg, SP, CNCPS fractions, and CP_{ip} are given as % of CP, Maillard polymers are given as mg/kg DM, TIA is given as g inhibited trypsin/kg DM, FL and all other analytes are given as g/kg DM. TP was calculated as CP minus A, SP was calculated as A plus B1.
[ab]Different superscripts indicate the difference between native and ensiled pea grains ($P < 0.01$).

Table 7
Effect of toasting ensiled field pea grains with a grain temperature graduated between 60 and 110 °C on concentrations of TP, SP, CNCPS protein fractions, CP_{ip}, amino acids, Maillard reaction products, and inhibited trypsin.

Pea treatment	TP	SP	A	B1	B2	B3	C	CP_{ip}	Lys	Arg	FL	CML	Pyr	MG-H1	TIA
Ensiled + toasted (60 °C)	176	28	9.9	18	64	8.2	0.57	6.1	7.4	7.3	7.8	22.9	36.3	4.7	2.1
	(174 179)	(26 30)	(8.4 11)	(16 20)	(59 70)	(5.1 11)	(0.24 0.90)	(4.7 7.4)	(6.8 7.9)	(6.9 7.8)	(6.6 8.9)	(12.5 33.3)	(−13.9 86.5)	(−0.99 10.4)	(2.0 2.3)
Ensiled + toasted (65 °C)	174	26	11	15	64	11	0.36	5.2	7.1	6.8	7.7	33.1	96.6	11.3	2.0
	(172 176)	(24 27)	(9.8 12)	(13 16)	(60 68)	(8.5 14)	(0.13 0.58)	(4.4 6.0)	(6.7 7.6)	(6.5 7.1)	(6.9 8.5)	(26.0 40.2)	(53.9 139)	(6.2 16.3)	(1.9 2.2)
Ensiled + toasted (70 °C)	172	23	12	12	63	14	0.40	4.9	6.9	6.3	7.5	41.2	157	18.4	2.0
	(170 173)	(22 25)	(11 12)	(11 13)	(60 65)	(12 16)	(0.19 0.62)	(4.4 5.4)	(6.5 7.3)	(6.1 6.5)	(6.9 8.1)	(36.2 46.2)	(121 193)	(14.0 22.8)	(1.9 2.1)
Ensiled + toasted (75 °C)	170	21	12	9.4	61	17	0.71	5.3	6.6	5.8	7.2	47.3	217	26.0	1.9
	(169 171)	(20 22)	(11 13)	(8.5 10)	(59 64)	(15 19)	(0.53 0.89)	(4.9 5.8)	(6.3 7.0)	(5.7 6.1)	(6.6 7.7)	(43.0 51.6)	(187 247)	(22.2 30.0)	(1.8 2.0)
Ensiled + toasted (80 °C)	169	20	12	7.3	59	20	1.3	6.3	6.4	5.5	6.7	51.3	277	34.2	1.8
	(168 170)	(19 21)	(12 13)	(6.3 8.2)	(56 61)	(18 22)	(1.1 1.5)	(5.7 6.9)	(6.1 6.7)	(5.3 5.7)	(6.2 7.3)	(46.9 55.7)	(251 304)	(30.8 37.6)	(1.7 1.9)
Ensiled + toasted (85 °C)	169	18	12	5.5	56	23	2.1	7.9	6.1	5.1	6.1	53.3	338	42.9	1.7
	(167 170)	(17 19)	(12 13)	(4.5 6.5)	(53 58)	(21 24)	(1.6 2.6)	(7.2 8.6)	(5.8 6.4)	(4.9 5.4)	(5.6 6.7)	(48.5 58.0)	(312 363)	(39.7 46.0)	(1.6 1.7)
Ensiled + toasted (90 °C)	169	16	12	4.1	52	26	3.2	10	5.8	4.9	5.4	53.2	398	52.1	1.5
	(167 170)	(15 18)	(12 13)	(3.1 5.1)	(49 55)	(24 28)	(2.3 4.1)	(9.4 11)	(5.5 6.2)	(4.7 5.1)	(4.9 6.0)	(48.3 58.0)	(371 426)	(48.7 55.4)	(1.4 1.6)
Ensiled + toasted (95 °C)	169	15	12	3.0	48	29	4.5	13	5.6	4.7	4.6	51.0	458	61.8	1.4
	(168 171)	(14 16)	(11 13)	(2.0 4.0)	(45 50)	(26 31)	(3.1 5.9)	(12 14)	(5.2 6.0)	(4.4 4.9)	(4.0 5.2)	(46.2 55.9)	(426 490)	(57.9 65.7)	(1.3 1.5)
Ensiled + toasted (100 °C)	171	14	11	2.3	43	32	6.2	17	5.3	4.5	3.7	46.9	519	72.1	1.3
	(169 172)	(12 15)	(10 12)	(1.2 3.4)	(40 46)	(29 34)	(4.1 8.2)	(15 18)	(4.9 5.8)	(4.3 4.8)	(3.0 4.4)	(41.6 52.1)	(481 557)	(67.3 76.8)	(1.1 1.4)
Ensiled + toasted (105 °C)	172	12	11	2.0	37	34	8.0	21	5.1	4.4	2.6	40.6	579	82.9	1.1
	(170 174)	(11 14)	(9.4 12)	(0.54 3.3)	(33 41)	(31 37)	(5.2 11)	(19 22)	(4.5 5.6)	(4.1 4.7)	(1.8 3.5)	(34.1 47.1)	(534 624)	(77.1 88.6)	(0.97 1.3)
Ensiled + toasted (110 °C)	174	11	9.4	2.0	31	37	10	26	4.8	4.4	1.5	32.3	639	94.2	0.99
	(172 177)	(9.1 14)	(7.9 11)	(0.02 3.8)	(25 36)	(34 41)	(6.4 14)	(24 28)	(4.2 5.4)	(4.0 4.8)	(0.31 2.6)	(23.3 41.3)	(586 692)	(87.2 101)	(0.80 1.2)
RMSE	2.38	2.1	1.4	1.8	5.1	4.2	2.8	3.3	0.59	0.33	0.85	8.36	61.4	7.67	0.21
R^2	0.567	0.855	0.388	0.889	0.826	0.812	0.873	0.853	0.646	0.897	0.864	0.576	0.894	0.926	0.727

A, non-protein nitrogen; Arg, arginine; B1, buffer-soluble TP; B2, buffer-insoluble TP minus TP insoluble in neutral detergent; B3, TP insoluble in neutral detergent but soluble in acid detergent; C, TP insoluble in acid detergent; CML, carboxymethyllysine; CNCPS, Cornell Net Carbohydrate and Protein System; CP, crude protein; CP_{ip}, CP insoluble in pepsin; DM, dry matter; FL, fructoselysine; Lys, lysine; MG-H1, methylglyoxal hydroimidazolone; Pyr, pyrraline; RMSE, root mean square error; SP, soluble protein; TIA, trypsin inhibitor activity; TP, true protein.
Values are least squares means estimated from linear (B3, Lys, Pyr), quadratic (MG-H1, TIA), or linear plus quadratic regression (TP, SP, A, B1, B2, C, CP_{ip}, Arg, FL, CML) at 5 °C-grain temperature stages within the measured temperature range; the 95 %-confidence intervals are given in brackets.
Dry matter is given as g/kg, SP, CNCPS fractions, and CP_{ip} are given as % of CP, Maillard polymers are given as mg/kg DM, TIA is given as g inhibited trypsin/kg DM, FL and all other analytes are given as g/kg DM. TP was calculated as CP minus A, SP was calculated as A plus B1.

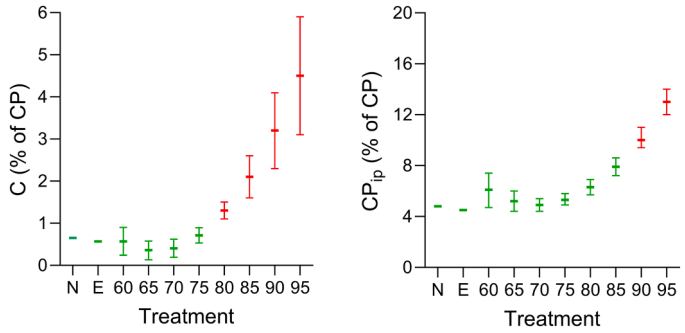

Fig. 2. Effect of toasting ensiled field pea grains with a grain temperature graduated to 5 °C-stages starting at 60 °C on concentrations of the CNCPS fraction C and CP_{ip}. Values are least squares means estimated from linear plus quadratic regression and the 95 %-confidence intervals are given as bars. The figures illustrate the significant increase in these fractions from 80 or 90 °C grain temperature onwards (marked red), and indicate protein damage when exceeding these thresholds. C, true protein insoluble in acid detergent; CNCPS, Cornell Net Carbohydrate and Protein System; CP, crude protein; CP_{ip}, CP insoluble in pepsin; E, ensiled; N, native (untreated grains).

within the protein. This confirms observations by Masoero et al. (2005) and others and is closely associated with the formation of Maillard polymers. The increase in the C fraction from around 80 °C grain temperature onwards is a clear indication of protein damage, since protein fixed in this way can no longer be hydrolysed (Fig. 2). Another indicator for protein damage is CP_{ip}, which significantly increased from 90 °C onwards (Fig. 2). A high correlation of $r = 0.99$ ($P < 0.001$) was found between fraction C and CP_{ip}, which makes these parameters interchangeable.

3.4. Amino acids and the formation of Maillard polymers

3.4.1. Ensiling effects on amino acids and Maillard polymers

The concentration of lysine was 11.6 g/kg DM in native and 9.6 g/kg DM in ensiled pea grains ($P < 0.01$). The lysine adducts fructoselysine, CML, and pyrraline increased a 26- ($P > 0.05$), 28- ($P > 0.05$), and 8-fold ($P < 0.01$), respectively, following dry ensiling (Table 6). Native pea grains had 7.6 g arginine/kg DM. Ensiling did not alter the concentration of arginine (Table 6). The arginine adduct methylglyoxal hydroimidazolone (MG-H1) was tripled after dry ensiling ($P < 0.01$; Table 6).

3.4.2. Toasting effects on amino acids and Maillard polymers

Toasting decreased the concentration of lysine starting at 80 °C grain temperature ($P < 0.05$; Table 7). The concentration of fructoselysine was more than doubled after toasting the ensiled pea grains with 60 °C grain temperature. A further increase of toasting temperature led to a decrease of the fructoselysine concentration, starting at 90 °C grain temperature ($P < 0.05$; Table 7). Carboxymethyllysine rapidly increased, peaked with 53.3 mg/kg DM at 85 °C, and decreased afterwards ($P < 0.05$). Pyrraline massively increased during toasting at any stage of temperature ($P < 0.05$). The concentration of arginine was reduced during toasting starting at 70 °C grain temperature by a 1.8-fold finally ($P < 0.05$). The concentration of MG-H1 increased diametrically opposed ($P < 0.05$; Table 7).

Maillard reaction products result from glycation, i.e., binding the amino group of an amino acid to the carbonyl group of a reduced sugar (ALjahdali and Carbonero, 2017). The present results clearly show an increase in the concentration of specific Maillard reaction products, already as a result of dry ensiling and, depending on the temperature, during toasting. This is accompanied by the binding of lysine or arginine. In silages, extended heating from microbial respiration or plant enzymatic activity may lead to raising temperature and increasing Maillard reaction rates (van Soest and Mason, 1991; Aloba et al., 2022). However, heating is not expected in the dry silages produced in the current study. On the contrary, it was shown that even in hay, usually having a DM content around 800 g/kg and low microbial activity, heating and Maillard reaction can occur during storage (Miao et al., 1994). The extent of Maillard reaction is obviously determined by temperature and the length of exposure (Muck et al., 2003; Aloba et al., 2022). Maillard reaction products can also be found in feed that has been processed or stored at lower temperatures (Hofmann et al., 2020). It remains unclear why measurable concentrations of Maillard polymers occur in such feed. Maillard reaction products seem to widely withstand fermentation or digestion (Kostyukovsky and Marounek, 1995; Nishino and Uchida, 2001). Maillard reaction products impair nutrient availability and advanced glycation end products may increase the prevalence of chronic inflammation in the digestive system (Teodorowicz et al., 2018).

3.5. Effects on antinutritional compounds

3.5.1. Ensiling effects on TIA

Native pea grains contained 3.8 g inhibited trypsin/kg DM. In the ensiled pea grains, TIA was slightly lower. There was no significant effect of ensiling on the TIA (Table 6).

3.5.2. Toasting effects on TIA

Toasting the ensiled peas reduced TIA, starting at 80 °C grain temperature, to a 2.5-fold finally ($P < 0.05$; Table 7).

The TIA in legumes plays a decisive role in nutrition of simple-stomach animals as it irreversibly binds digestive enzymes, principally those secreted by the pancreas (Erdaw and Beyene, 2018). In pigs, Leterme et al. (1990) and Chen et al. (2020) reported depressed ileal CP and amino acid digestibility as a result. A high TIA concentration in the diet correlates with a rapid feed passage, greater nitrogen excretion and poor litter quality (Erdaw et al., 2016; Erdaw and Beyene, 2018). In ruminants, rumen fermentation is able to inactivate TIA (Hoffmann et al., 2003). However, some of it may withstand fermentation and lower intestinal protein digestibility (Holmes et al., 1993; Aldrich et al., 1997; Van der Poel et al., 2005). The reduction of TIA by heat treatment is generally accepted, but the reports are not consistent (Kadam and Smithard, 1987; Masoero et al., 2005; Erdaw and Beyene, 2018). Treatments involving heat and pressure such as extrusion or expansion seem to be most effective (Kadam and Smithard, 1987; Masoero et al., 2005). The TIA in pea cultivars is reported in a range of about 2.0–10 g inhibited trypsin/kg sample (Guillamón et al., 2008; Živanov et al., 2018; Liu, 2021). In the current study, we found 3.8 g inhibited trypsin/kg DM. Goodarzi Boroojeni et al. (2017) measured 0.67 g inhibited trypsin/kg DM, which is comparatively low. Modern cultivars with a low TIA thus may not require any further treatment for reduction.

4. Conclusions

In the present experiment, pea grains were harvested at 786 g DM/kg and ensiled. That way, proteolytic and desmolytic processes were largely absent and the nutritive and energetic value was maintained. However, this process led to a significant increase in Maillard reaction products, which bind lysine or arginine and reduce the solubility of the protein. Toasting this dry silage caused sugar, lysine, and arginine concentrations to fall and those of the Maillard products to rise, depending on the grain temperature reached during the process. A positive effect of dry ensiling and toasting is a significant reduction of the TIA. To avoid fixing the pea protein, a maximum temperature of 80 °C at the grain should not be exceeded. This applies when using the described technology and process parameters. The expected positive effects are then very limited, which is why the effort and benefit of toasting must be critically weighed against each other.

CRediT authorship contribution statement

Martin Bachmann: Writing – original draft, Visualization, Investigation, Formal analysis, Data curation, Conceptualization. **Christian Kuhnitzsch:** Writing – review & editing, Project administration, Investigation, Formal analysis, Data curation, Conceptualization. **Monika Wensch-Dorendorf:** Writing – review & editing, Formal analysis. **Thomas Hofmann:** Writing – review & editing, Methodology, Formal analysis. **Thomas Henle:** Writing – review & editing, Supervision, Resources. **Jörg M.Greef:** Writing – review & editing, Resources. **Siriwan D. Martens:** Writing – review & editing, Methodology, Investigation. **Annette Zeyner:** Writing – review & editing, Supervision, Resources, Project administration, Funding acquisition, Conceptualization. **Olaf Steinhöfel:** Writing – review & editing, Supervision, Resources, Project administration, Funding acquisition, Conceptualization.

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Declaration of Competing Interest

All Authors declare that they have no conflicts of interest.

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