Effect of Steam Explosion Parameters on Total Glucosinolate Detoxification in Rapeseed Meal (Postprint)

Authors: He Yonghui, Wang Qinghua, Liu Changzhong, Liu Xingyou, Hongbing Xie, Wang Yanrong, He Yun

Date: 2017-10-11T00:00:00+00:00

Abstract

This experiment aimed to investigate the effects of steam explosion parameters (water-to-material ratio, steam pressure, holding time) on the detoxification efficacy of total glucosinolates in rapeseed meal, and to evaluate the nutrient digestibility of detoxified rapeseed meal using an in vitro biomimetic method. A single-factor experimental design was adopted to screen for the optimal water-to-material ratio, holding time, and steam pressure, with each treatment consisting of 4 levels and 3 replicates per level. The results demonstrated that steam explosion treatment exhibited a significant detoxification effect on total glucosinolates in rapeseed meal, with detoxification rates ranging from 73.71% to 86.98%. Using total glucosinolate detoxification rate as the screening criterion and following the single-factor experimental design, the optimal parameters were determined to be a water-to-material ratio of 25%, a holding time of 60 s, and a steam pressure of 2.0 MPa. When using nutrient digestibility of rapeseed meal as the screening criterion, it was found that compared with the untreated group, steam pressures of 1.0 and 1.5 MPa (with fixed water-to-material ratio of 25% and holding time of 60 s) had no significant effect on nutrient digestibility ($P>0.05$). However, at a steam pressure of 2.0 MPa, the digestibility of crude protein, lysine, and arginine was significantly or highly significantly reduced ($P<0.05$ or $P<0.01$). Increasing steam pressure could highly significantly decrease the contents of total glucosinolates and isothiocyanates ($P<0.01$). Considering both total glucosinolate detoxification rate and nutrient digestibility indices comprehensively, when the water-to-material ratio is fixed at 25% and the holding time at 60 s, the appropriate steam pressure should not exceed 2.0 MPa.

Full Text

Abstract

This experiment was conducted to investigate the effects of steam explosion parameters (water-to-material ratio, steam pressure, and retention time) on the detoxification efficiency of total glucosinolates in rapeseed meal, and to evaluate the nutrient digestibility of detoxified rapeseed meal using an in vitro biomimetic method. Single-factor experimental designs were employed to screen for optimal water-to-material ratio, retention time, and steam pressure. Each experimental factor comprised four levels with three replicates per level. The results demonstrated that steam explosion treatment effectively detoxified total glucosinolates in rapeseed meal, achieving detoxification rates of 73.71% to 86.98%. Using total glucosinolate detoxification rate as the screening criterion in single-factor experiments, the optimal parameters were determined to be a water-to-material ratio of 25%, retention time of 60 s, and steam pressure of 2.0 MPa.

When nutrient digestibility was used as the screening criterion, steam explosion at 1.0 and 1.5 MPa (with water-to-material ratio fixed at 25% and retention time at 60 s) showed no significant effect on nutrient digestibility compared with the untreated group ($P > 0.05$). However, at 2.0 MPa, steam explosion significantly or extremely significantly reduced the digestibility of crude protein, lysine, and arginine ($P < 0.05$ or $P < 0.01$). Increasing steam pressure extremely significantly decreased the contents of total glucosinolates and isothiocyanates ($P < 0.01$). Considering both total glucosinolate detoxification rate and nutrient digestibility, when the water-to-material ratio is fixed at 25% and retention time at 60 s, the appropriate steam pressure should not exceed 2.0 MPa.

Keywords: steam explosion; rapeseed meal; glucosinolates; detoxification; biomimetic method; digestibility

Introduction

Rapeseed meal (RSM) contains 34%–38% crude protein. The primary toxic and harmful component in RSM is total glucosinolates (TG). While TG itself is non-toxic, it can be degraded by endogenous enzymes or intestinal microorganisms to produce oxazolidinethione, isothiocyanates, and other compounds that cause thyroid and liver enlargement and dysfunction, thereby affecting animal growth, performance, and even causing death. This severely restricts the application of RSM in monogastric animal feed, resulting in utilization rates of less than 30% for RSM feed resources in China. This situation contrasts sharply with China' s severe shortage of protein feed resources and heavy reliance on imports, necessitating the development of an efficient detoxification technology for rapid removal of TG from RSM.

Steam explosion technology involves treating raw materials with high-temperature and high-pressure steam (200–260 °C, 2.0–5.0 MPa) for a period before explosive discharge from the chamber within milliseconds. Due to its

short duration and high energy density, this process can alter the chemical structure of raw materials and has been widely applied in papermaking, biomass energy utilization, and bioactive compound extraction. In the feed industry, steam explosion has primarily been used for developing coarse feed resources such as wheat straw and corn stover. However, no studies have investigated the detoxification of TG in RSM using steam explosion technology. Our laboratory has previously demonstrated that steam explosion can detoxify cottonseed meal, leading us to hypothesize that it could also detoxify RSM. Therefore, this study aimed to investigate the effects of three steam explosion parameters—water-to-material ratio, steam pressure, and retention time—on TG detoxification efficiency in RSM, and to evaluate the detoxification effect using an in vitro biomimetic method to identify optimal steam explosion parameters.

Materials and Methods

1.1 Experimental Materials

Commercial RSM was used in the experiments, containing 27.52 mmol/kg TG, 4.4% erucic acid, 0.33 mg/g oxazolidinethione, 1.7 mg/g isothiocyanates, and 38.69% crude protein.

1.2 Experimental Equipment

A steam explosion test bench (QBS-80B, Hebi) was used for the experiments.

1.3 Experimental Design

Commercial RSM was passed through a 10-mesh sieve. Distilled water was added to achieve specific water-to-material ratios (mass percentage), mixed thoroughly, and sealed in self-sealing bags for 8–10 hours to ensure complete moistening. The steam explosion test bench was adjusted to the target steam pressure, RSM was loaded into the chamber, and after maintaining pressure for a specified retention time, the pressure was released. Single-factor experiments were designed to examine three parameters: water-to-material ratio (0, 20%, 25%, 30%), retention time (30, 60, 90, 120 s), and steam pressure (1.0, 1.5, 2.0, 2.5 MPa). The experimental design is shown in Table 1 . Each treatment was repeated three times (n = 3). After collection, samples were dried to constant weight in an oven at 65 °C. Samples ground through a 40-mesh sieve were used for TG, oxazolidinethione, and isothiocyanate content determination, while samples ground through a 60-mesh sieve were used for in vitro nutrient digestibility analysis.

Steam explosion intensity ($\log_{10}(R_0)$) is an index for comprehensively comparing the effects of steam temperature and retention time, calculated as: $R_0 = t \cdot \exp[(T-100)/14.75]$, where t is retention time (min) and T is steam temperature (°C) [17].

1.4 Analytical Methods

TG content was determined using the NY/T 1582-2007 method with an Agilent 1200 HPLC system (USA). Oxazolidinethione and isothiocyanate contents were measured using NY/T 1799-2009 and NY/T 1596-2008 methods, respectively, with an Eppendorf Biospectrometer basic spectrophotometer (Germany). Dry matter, energy, crude protein, and amino acid digestibility were analyzed using the porcine in vitro biomimetic "SDS-II Monogastric Animal Simulated Digestion System" [18]. Energy content was determined using the ISO 9831:1998 method with a PARR 1281 automatic oxygen bomb calorimeter (USA). Crude protein content was measured using the Dumas combustion method with a Rapid N III nitrogen analyzer (Germany). Amino acid content was analyzed after hydrolysis with 6 mol/L HCl at 110 °C for 24 h using a Hitachi L-8800 automatic amino acid analyzer (Japan).

1.6 Statistical Analysis

Data were analyzed using SAS 9.2 statistical software. Differences were considered extremely significant at $P < 0.01$, significant at $P < 0.05$, and not significant at $P > 0.05$.

Results

2.1 Effects of Steam Explosion Treatment on TG Detoxification Rate in RSM

As shown in Table 2 , steam explosion treatment achieved TG detoxification rates of 73.71%–86.98% in RSM. The highest detoxification rate of 86.98% was obtained at a water-to-material ratio of 25%, steam pressure of 2.0 MPa, and retention time of 60 s.

With steam pressure fixed at 2.0 MPa and retention time at 60 s, TG detoxification rate initially increased then decreased as water-to-material ratio increased from 0 to 30%, ranging from 73.71% to 86.92%. The 25% water-to-material ratio yielded the highest detoxification rate of 86.92%, which was not significantly different from the 30% ratio ($P > 0.05$). Therefore, the optimal water-to-material ratio was determined to be 25%.

With water-to-material ratio fixed at 25% and steam pressure at 2.0 MPa, TG detoxification rate initially increased then decreased as retention time extended from 30 to 120 s, ranging from 76.31% to 86.98%. The 60 s retention time produced the highest detoxification rate of 86.98%, which was significantly or extremely significantly higher than other retention times ($P < 0.05$ or $P < 0.01$). Further extension of retention time did not improve detoxification efficiency, establishing 60 s as the optimal retention time.

With water-to-material ratio fixed at 25% and retention time at 60 s, TG detoxification rate initially increased then decreased as steam pressure increased from

1.0 to 2.5 MPa, ranging from 76.02% to 85.39%. The 2.0 MPa steam pressure yielded the highest detoxification rate of 85.39%, which was significantly or extremely significantly higher than other pressures ($P < 0.05$ or $P < 0.01$). Further increase in steam pressure did not enhance detoxification efficiency, establishing 2.0 MPa as the optimal steam pressure.

2.2 Effects of Steam Pressure on TG and Its Degradation Products in RSM

Table 3 presents the contents of TG and its degradation products at different steam pressures with water-to-material ratio fixed at 25% and retention time at 60 s. Compared with the untreated group, increasing steam pressure extremely significantly reduced TG and isothiocyanate contents ($P < 0.01$), while oxazolidinethione was not detected. At 2.0 MPa steam pressure, TG detoxification rate reached its maximum of 85.39%, which was significantly or extremely significantly higher than other steam pressures ($P < 0.05$ or $P < 0.01$).

2.3 Effects of Steam Pressure on Nutrient Digestibility in RSM

Table 4 shows the effects of different steam pressures on nutrient digestibility in RSM with water-to-material ratio fixed at 25% and retention time at 60 s. Steam explosion at different pressures had no significant effect on the digestibility of threonine, alanine, isoleucine, leucine, tyrosine, phenylalanine, histidine, or proline ($P > 0.05$). However, the digestibility of dry matter, gross energy, crude protein, aspartic acid, serine, glutamic acid, glycine, valine, lysine, and arginine tended to decrease with increasing steam pressure. Compared with the untreated group, steam explosion at 1.0 and 1.5 MPa had no significant effect on nutrient digestibility ($P > 0.05$), while 2.0 MPa steam explosion significantly or extremely significantly reduced the digestibility of crude protein, lysine, and arginine ($P < 0.05$ or $P < 0.01$).

Discussion

3.1 Effects of Steam Explosion Treatment on TG Detoxification Rate in RSM

Steam explosion treatment effectively improved TG detoxification in RSM. With fixed steam pressure and retention time, increasing water-to-material ratio from 0 to 25% significantly enhanced TG detoxification rate, indicating that an appropriate water-to-material ratio facilitates uniform penetration of high-temperature steam into RSM, weakening intermolecular bonding and promoting steam-induced structural modification and degradation, thereby improving treatment efficacy while mitigating the negative effects of high temperature on protein and amino acid digestibility. However, excessive water-to-material ratio hinders steam penetration and weakens the steam explosion effect, which explains why the 30% water-to-material ratio showed lower detoxification efficiency than the 25% ratio.

With fixed water-to-material ratio and steam pressure, TG detoxification rate initially increased then decreased with extended retention time, suggesting that appropriate extension enhances steam explosion intensity and detoxification efficacy. However, further extension causes excessive steam condensation on RSM, which impedes steam penetration and reduces treatment effectiveness, explaining the decreased efficiency observed at retention times exceeding 60 s.

With fixed water-to-material ratio and retention time, TG detoxification rate initially increased then decreased with increasing steam pressure, indicating that moderate pressure enhancement improves detoxification, but higher pressure does not necessarily yield better results. Excessive high-temperature, high-pressure steam condensation on RSM weakens the structural modification and degradation effects, which accounts for the inferior detoxification performance at 2.5 MPa compared with 2.0 MPa.

This study confirms that appropriate water-to-material ratio, steam pressure, and retention time significantly improve TG detoxification rates in RSM. Although all water-to-material ratio treatments had the same steam explosion intensity of 3.4 (Table 1), detoxification efficacy varied significantly with water-to-material ratio, demonstrating that this parameter substantially influences treatment outcomes. However, the steam explosion intensity formula only reflects steam pressure (temperature) and retention time, not the water-to-material ratio, indicating limitations of this index. Since steam explosion detoxification efficacy is affected by water-to-material ratio (moisture), steam pressure, and retention time, research on parameter optimization should address all three factors comprehensively.

3.2 Effects of Steam Pressure on TG and Its Degradation Products in RSM

With fixed water-to-material ratio and retention time, moderate steam pressure increase significantly reduced TG, oxazolidinethione, and isothiocyanate contents. At 2.0 MPa, optimal detoxification of TG, isothiocyanates, and oxazolidinethione was achieved. This may be attributed to steam explosion' s ability to disrupt or degrade the chemical structures of these compounds and inactivate myrosinase in RSM, preventing the formation of more toxic compounds such as oxazolidinethione and isothiocyanates during processing. However, excessive steam pressure reduces treatment efficacy due to moisture interference with structural modification and degradation, explaining the decreased detoxification at 2.5 MPa.

Compared with existing detoxification technologies, the steam explosion technique in this study achieved lower TG detoxification rates (73.71%–86.98%) than microbial solid-state fermentation with mixed strains (92.01%–94.85%) and mixed solvent methods (92.01%–94.85%), but superior to enzymatic hydrolysis and most microbial fermentation treatments (48.34%–60.62%). Chemical solvent methods suffer from poor palatability, significant nutrient loss, detoxifi-

cant residues, and wastewater pollution. Microbial detoxification requires long fermentation times (24–48 h). In contrast, steam explosion treatment simply requires adjusting RSM moisture with tap water, hydrating at room temperature, and maintaining pressure for several tens of seconds to rapidly and efficiently remove TG, isothiocyanates, and oxazolidinethione from RSM. The treated RSM meets Chinese national standards for feed-grade rapeseed meal (isothiocyanate content below 0.75 g/kg), offering advantages of simple pretreatment, short detoxification time, high efficiency, environmental friendliness, and non-corrosive equipment.

3.3 Effects of Steam Pressure on Nutrient Digestibility in RSM

This study demonstrates that at appropriate steam pressure ($ $1.5 MPa), steam explosion effectively detoxifies RSM without significantly affecting nutrient digestibility. However, higher steam pressure (>2.0 MPa) significantly reduces nutrient digestibility, indicating that excessive pressure (temperature) can induce Maillard reactions that compromise nutritional quality and reduce protein and amino acid content and digestibility. The degree of damage depends on steam pressure (temperature) and retention time. Therefore, when investigating steam explosion conditions for protein feedstuffs like RSM, attention must be paid not only to detoxification rate but also to effects on protein quality and amino acid digestibility. Balancing both detoxification efficacy and nutrient digestibility, the optimal steam explosion conditions are a water-to-material ratio of 25%, retention time of 60 s, and steam pressure not exceeding 2.0 MPa.

For monogastric animals, feed protein quality depends on both amino acid quantity and digestibility. Improper high-temperature treatment can trigger Maillard reactions that reduce protein and amino acid (especially lysine) content and digestibility. While in vivo methods are the most direct and reliable for evaluating digestibility, time and cost constraints often necessitate in vitro methods. The "Monogastric Animal Simulated Digestion System" can mimic animal digestive processes using computer control to accurately evaluate energy and crude protein digestibility of pig feed in vitro, providing valuable data for feed processing evaluation. However, in vitro biomimetic methods cannot replace animal digestion and growth trials, necessitating further animal studies to evaluate nutrient utilization.

Using TG detoxification rate as the evaluation criterion, single-factor experiments identified optimal steam explosion parameters of 25% water-to-material ratio, 60 s retention time, and 2.0 MPa steam pressure. However, considering both TG detoxification rate and nutrient digestibility, when water-to-material ratio is fixed at 25% and retention time at 60 s, the appropriate steam pressure should not exceed 2.0 MPa.

References

[1] TOUSSAINT B, EXCOFFIER G, VIGNON M R. Effect of steam explosion treatment on the physico-chemical characteristics and enzymic hydrolysis of poplar cell wall components[J]. Animal Feed Science and Technology, 1991, 32(1/2/3): 235-242.

[2] YU Z D, ZHANG B L, YU F Q, et al. A real explosion: the requirement of steam explosion pretreatment[J]. Bioresource Technology, 2012, 121: 335-341.

[3] ZHU J J, YONG Q, CHEN S X, et al. Analysis of degradation products from steam explosion of corn straw[J]. Chemistry and Industry of Forest Products, 2009, 29(2): 22-26.

[4] ZHANG X, LI H W, MA X J. Changes in apparent enzyme activity and physicochemical properties of steam-exploded corn straw during enzymatic hydrolysis[J]. Transactions of the Chinese Society of Agricultural Engineering, 2013, 29(11): 203-209.

[5] SU T F, ZHAO G Z, REN T B, et al. Characterizations of physico-chemical changes of corn biomass by steam explosion[J]. Transactions of the Chinese Society of Agricultural Engineering, 2015, 31(6): 253-256.

[6] WANG F Q, TONG Y X, LI C B, et al. Detoxification of steam-exploded corn cob hydrolysate and its fermentation for biofuel butanol production[J]. Transactions of the Chinese Society of Agricultural Engineering, 2016, 32(5): 257-262.

[7] ZHANG B B, ZENG G M, FU Y, et al. Study on extraction technology of flavonoids from ginkgo leaves by steam explosion[J]. Journal of Cellulose Science and Technology, 2012, 20(1): 39-44, 71.

[8] SHI M, WANG H Y, CHENG Y L, et al. Preliminary study on safety evaluation of Ganoderma lucidum extract prepared by steam explosion pretreatment[J]. Science and Technology of Food Industry, 2014, 35(6): 338-342.

[9] HE Y H, WANG Q H, HUANG H L, et al. Steam explosion for increasing water-soluble pentosan content of wheat bran and its thermogravimetric analysis[J]. Transactions of the Chinese Society of Agricultural Engineering, 2015, 31(13): 286-291.

[10] LIU C, ZHANG R T, LIU B G, et al. Effect of steam explosion treatment on phenolic acid composition of wheat antioxidant capacity[J]. Transactions of the Chinese Society of Agricultural Engineering, 2016, 32(6): 308-314.

[11] VIOLA E, ZIMBARDI F, CARDINALE M, et al. Processing cereal straws by steam explosion in a pilot plant to enhance digestibility in ruminants[J]. Bioresource Technology, 2008, 99(4): 681-689.

[12] CHANG J, YIN Q Q, REN T B, et al. Effect of steam explosion pretreatment and microbial fermentation on degradation rate of corn stover[J]. Trans-

actions of the Chinese Society of Agricultural Engineering, 2011, 27(4): 277-280.

[13] CHANG J, YIN Q Q, JIANG Y B, et al. Study on effect of biological straw on apparent metabolizable energy for broilers and its optimal proportion for replacing corn[J]. Chinese Journal of Animal Nutrition, 2012, 24(8): 1557-1563.

[14] HE Y H, WANG Q H, MIAO Z G, et al. A rapid feed processing technology for removing gossypol content from cottonseed meal: China, 201410036507.2[P]. 2014-05-07.

[15] WANG Q H, HE Y H, LU H W, et al. Study on detoxification effect of steam explosion technology on free gossypol in cottonseed meal[J]. Chinese Journal of Animal Nutrition, 2016, 28(2): 524-530.

[16] HE Y H, WANG Q H, LIU X Y, et al. A rapid feed processing technology for removing glucosinolate content from rapeseed meal: China, 201410069566.X[P]. 2014-05-28.

[17] VALIENTE C, ARRIGONI E, ESTEBAN R M, et al. Chemical composition of olive by-product and modifications through enzymatic treatments[J]. Journal Science Agriculture, 1995, 69(1): 27-32.

[18] ZHAO F, ZHANG L, MI B M, et al. Using a computer-controlled simulated digestion system to predict the energetic value of corn for ducks[J]. Poultry Science, 2014, 93(6): 1410-1420.

[19] IROBA K L, TABIL L G, SOKHANSANJ S, et al. Pretreatment and fractionation of barley straw using steam explosion at low severity factor[J]. Biomass and Bioenergy, 2014, 66: 286-300.

[20] REN T B, MA X Q, XU G Z, et al. Optimization of steam explosion pretreatment conditions for corn stover using response surface methodology[J]. Transactions of the Chinese Society of Agricultural Engineering, 2011, 27(9): 282-286.

[21] LAN S L, MAO X W, XIAO T Y, et al. Optimization of mixed microbial solid-state fermentation conditions for detoxification of rapeseed meal using response surface methodology[J]. Chinese Journal of Animal Nutrition, 2013, 25(3): 617-627.

[22] HU Y N, LI A K, WANG Z S, et al. Study on nutritional characteristics of rapeseed meal by microbial solid-state fermentation[J]. Journal of the Chinese Cereals and Oils Association, 2012, 27(3): 76-80.

[23] LAN W J, PENG M J, LYU Q, et al. Study on detoxification technology of rapeseed meal by mixed solvent method[J]. China Oils and Fats, 2012, 37(6): 28-32.

[24] CHEN L, GAO L X, HUANG Q H, et al. Prediction of digestible energy of feed ingredients for growing pigs using computer-controlled simulated digestion system[J]. Journal Animal Science, 2014, 92(9): 3887-3894.