Nutrient Composition and *In Vitro* Fermentation Characteristics of Common Vegetable Peels as Livestock Feed

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Abstract

The study evaluated the nutritional composition and *in vitro* rumen fermentation characteristics of four common vegetable peels: cucumber, carrot, potato, and ginger as potential feed resources for livestock feed. The chemical composition and *in vitro* gas production, dry matter digestibility, and volatile fatty acid profiles of these peels were assessed in a complete randomized design (CRD). The results showed That cucumber peels exhibited higher ($P < 0.0001$) organic matter (OM), crude protein (CP), acid detergent fibre (ADF), and acid detergent lignin (ADL) content; 24.81%, 21.37%, 44.50% and 44.50%, respectively, whereas lowest DM content (5.50%). Carrot peels had the lowest CP (5.61%) and NDF (34.30%) content. Potato peels were characterized by higher DM (14.56%) content and lower OM (7.95%), ADF (25.68%), and ADL (26.67%) content. The ginger peels had the highest ($P < 0.0001$) NDF (86.64%) content. *In vitro* gas production studies revealed carrot peels had the highest cumulative gas production, while the lowest in ginger peels. Similarly, carrot peels exhibited the highest ($P < 0.0001$) *in vitro* dry matter digestibility (IVDMD) and lowest *in vitro* neutral detergent fibre digestibility (IVNDFD). The potato peel volatile fatty acid (VFA) profile produced the highest concentrations of acetic, propionic, butyric acids and total VFA. In conclusion, cucumber, carrot, and potato peels have the potential to be used as potential feed resources for livestock. Further research (*in vivo*) is required to assess their effects on animal performance and health.

Keywords: vegetable peels, nutrient composition, *in vitro* rumen fermentation, digestibility, volatile fatty acids, livestock feed.

Introduction

The increasing demand for livestock products has led to a surge in feed costs and environmental concerns (Rojas-Downing et al., 2017; Sharma, 2014). To mitigate these issues, there is a growing interest in exploring alternative feed sources, particularly those derived from agricultural by-products. Vegetable waste, including peels from fruits and

vegetables, represents a significant proportion of farm waste and often ends up in landfills, contributing to environmental pollution. The global livestock industry is facing significant challenges due to the escalating costs of raw materials used in ruminant feed. This upward trend in prices is driven by several factors, including geopolitical tensions, climate change and extreme weather events, and increased demand for animal protein. To address these challenges, livestock producers and industry stakeholders may need to explore alternative feed sources, improve feed efficiency, and implement innovative feeding strategies. Additionally, policymakers can play a role by promoting sustainable agriculture practices, investing in research and development, and implementing policies that support the livestock industry. However, these peels are rich in nutrients, such as fibre, carbohydrates, and minerals, and have the potential to be utilised as a valuable feed resource for ruminant livestock (Jalal et al., 2023; Malenica et al., 2023; Ray, 2022).

Ruminant livestock, such as cattle and sheep, rely on microbial fermentation in the rumen to digest plant-based feedstuffs (Paswan et al., 2022). The rumen microbiome plays a crucial role in breaking down complex plant cell wall components, including cellulose and hemicellulose, into volatile fatty acids (VFAs), which serve as the primary energy source for the host animal (Cholewińska et al., 2020; Qi et al., 2024; Zhang et al., 2022). By understanding the fermentation characteristics of different feedstuffs, it is possible to optimise ruminant diets and improve animal performance (Dai et al., 2019).

Vegetable peels, often discarded as waste, are rich in nutrients, including fibre, carbohydrates, and minerals (Haque et al., 2023; Rifna et al., 2023). By incorporating these underutilized resources into ruminant diets, it is possible to reduce reliance on conventional feedstuffs and mitigate environmental impacts. However, the nutritional value and fermentation characteristics of different vegetable peels can vary significantly (Ganesh et al., 2022; Sagar et al., 2018).

In recent years, there has been increasing interest in using agricultural by-products as feed additives to enhance ruminant nutrition and reduce greenhouse gas emissions (Jalal et al., 2023; Malenica et al., 2023; Salami et al., 2019). Vegetable peels, in particular, have gained attention due to their potential to modulate rumen fermentation and improve feed efficiency (Jalal et al., 2023; Wanapat et al., 2024). However, the specific effects of different vegetable peels on rumen microbial activity and fermentation parameters remain largely unexplored.

By evaluating their chemical composition, *in vitro* gas production, and rumen microbial fermentation parameters, we aim to assess their potential as a sustainable and cost-effective feed resource for ruminant livestock. Understanding the effects of these vegetable peels on rumen microbial activity and fermentation can

provide valuable insights into their potential to improve ruminant feed efficiency and reduce environmental impacts. This study evaluates the chemical composition, *in vitro* gas production, and rumen microbial fermentation parameters of four common vegetable peels: cucumber (*Cucumis sativus*), carrot (*Daucus carota*), potato (*Solanum tuberosum*), and ginger (*Zingiber officinale*).

Materials and methods

Study area

The study was carried out in UPM, Serdang, Selangor. The samples were collected from Serumpun Café in UPM, while the rumen fluid was collected from Shah Alam abattoir. Proximate analysis of the samples was conducted in the Nutrition Laboratory and the *in vitro* rumen fermentation analysis was conducted in the pasture Laboratory. Both laboratories were in the Department of Animal Science, Faculty of Agriculture, Universiti Putra Malaysia.

Experimental design

The rumen fluid was collected from cattle at Shah Alam abattoir in the morning before feeding. The experimental design used was a completely randomized design (CRD) since the samples were randomly placed in the water bath for incubation.

Treatment and sample preparation

Cucumber, Carrot, Potato and Ginger peels were collected, dried in 60 ^{0}C ovens and then ground into smaller particles. The samples were then stored in an airtight container for proximate analysis and *in vitro* rumen fermentation analysis.

Chemical analysis

The chemical analysis was conducted for moisture, ash, crude fibre, protein, fat, and carbohydrate contents of the vegetable peels was determined as described by AOAC (2016). Neutral detergent fibre (NDF), acid detergent fibre (ADF), and acid detergent lignin (ADL) were determined based on the (Van Soest & Mason, 1991). All the analyses were repeated three times.

Rumen collection

Rumen fluid was collected from rumen-fistulated animals before morning feeding. The fluid was transferred into a pre-warmed thermos flask, continuously flushed with CO_2 gas to maintain anaerobic conditions, and immediately transported to the laboratory. The pooled rumen fluid was then filtered through six layers of cheesecloth to remove large particulate matter.

Buffered media preparations

Buffer rumen medium was prepared by combining five stock solutions which are: microminerals (Solution A), buffer (Solution B), macrominerals (Solution C), resazurin, and a reducing agent. Strained rumen fluid was added to the medium at a 1:2 (v/v) ratio. Rumen fluid was buffered using a prepared Buffer pH 6.8 at a 1:3 ratio (Marten and Barnes, 1979; Ankom, 2018). One gram of substrate was incubated over 24 h with 25 ml rumen fluid, and 75 ml buffer in a 39 ^{0}C water bath as described by (Menke

& Steingass, 1988) and (Tunkala et al., 2022).

In vitro fermentation

In vitro rumen fermentation was carried out according to the method of Menke & Steingass (1988).

pH determination

The pH of the samples was measured with a calibrated pH meter. The probe was cleaned and rinsed with distilled water before being used on other samples, and it was then dried with tissue paper. The reading was measured at 48 hours.

In Vitro Dry Matter (IVDMD) and Organic Matter Digestibility (IVOMD)

At 48 hours, all glass syringes were taken out from the rack. After the reading at 48 hours was recorded, all the content was poured into a pre-weighed sintered glass and placed into a centrifuge tube which will be kept inside the freezer at -20 °C. While the substrate in sintered glass was rinsed with distilled water using a vacuum pump. After filtration, the sintered glasses were placed in the oven at 105 °C for IVDMD and then inside the furnace at 550 °C for IVOMD. Each sample's weight was recorded.

IVDMD or IVOMD (%):

{[Initial DM or OM (g) – Undigested DM or OM (g) – Blank] / Initial DM or OM (g)} X 100%

Ammonia concentration determination

The sample ammonia concentration was estimated by the colourimetric technique as described by Weatherburn (1967) using a multiscan colourimetric plate reader (Thermo Multiskan Spectrum, Thermo Fisher Scientific).

Volatile fatty acids (VFA)determination

The VFA including acetic acid, propionic acid, isobutyric acid, butyric acid, isovaleric acid (including 2-methylbutyric acid), and valeric acid concentrations were determined by Gas Chromatography fitted with a flame ionization detector using methyl valerate as the internal standard (Tunkala et al., 2022). Briefly, rumen fluid was kept at −20 °C. Each sample received a fresh addition of 1 mL of metaphosphoric acid diluted in 1.5 M 98% H2SO4 (Emsure® Merck, Darmstadt, Germany) and left overnight. The mixture was then centrifuged at 10,000g for 20 min at 4 °C. The internal standard, 4-methyl-valeric acid, was subsequently added to an equivalent volume of the supernatant that had been extracted, filtered through a 0.45-µm syringe filter, and placed in a 1.5 mL-screw-capped glass vial (Supelco, Inc., Sigma-Aldrich, St. Louis, MO, USA) (Sigma-Aldrich Chemical Co., St. Louis, MO, USA). The VFA separation was carried out using a gas chromatograph with a 6890N Network GC System gas chro- matograph (Hewlett-Packard, Avondale, PA, USA), using a bonded phase fused silica capillary column (15 m, 0.25 mm ID, 0.25 m film thickness) from the Quadrex 007 Series (Quadrex Corp., New Haven, CT, USA). The column tem- perature was 200 °C, the injector and detector temperatures were 230 °C, and the flow rate of the carrier gas, purified nitrogen, was 60 mL/min. An external standard (Sigma- Aldrich Chemical Co., St. Louis, MO, USA) and 10-

mM concentrations of propionic, butyric, isobutyric, valeric, isovaleric, and 4-methyl-valeric acids were employed to identify the sample's peak locations. Per individual points of the internal and external standards, the molar concentration was determined.

Statistical analysis

The data obtained were analysed using one-way analysis of variance (ANOVA) by a general linear model (GLM) procedure in SAS software 9.4 Version (SAS Institute Inc., Cary, NC, USA). Duncan's multiple range test was used to separate means at $P < 0.05$ significance level.

Table 1. Types of treatment groups for proximate and pf control for *in vitro* rumen fermentation analysis

Treatments	Sample types[#]
A	Cucumber
B	Carrot
C	Potato
D	Ginger
Control 1	Blank
Control 2	Standard Hay
Control 3	Standard Concentrate

[#]There were three and six replicates for each control and treatment group, respectively (i.e. twenty-four samples for proximate analysis and thirty-three samples for the *in vitro* rumen fermentation gas production).

Results and discussion

Proximate analysis

The chemical composition of the vegetable peels varied significantly (Table 2). Potato peels exhibited the highest DM content, while cucumber peels had the lowest. Conversely, cucumber peels had the highest ($P < 0.05$) OM, CP, ADF, and ADL content compared to the other peels. Carrot peels, on the other hand, had the lowest CP content when compared with other vegetable peels. Ginger and cucumber peels were shown to have higher fibre fractions of neutral detergent fibre (NDF) content, significantly differing from the other peels.

The cultivar and potato tissue determine the proximate and mineral content. The dry matter and total soluble solids are highest in the flesh of potatoes, but the protein, fibre, ash, and minerals (except magnesium) are much higher in the peels (Vaitkevičienė, 2019). The current study is supported by Arapoglou et al. (2010), who reported that raw potato peels had a high moisture content (83.3–85.1%) and a high carbohydrate content (8.7–12.4%), but a low total protein content (1.2–2.3%) and fat content (0.1–0.4%). Potato peels are primarily composed of

carbohydrates, dietary fibre, and protein, with the specific composition varying among different genotypes. Carbohydrates constitute the most abundant macronutrient ranging from 69 to 88 g/100 g dry weight (Sampaio et al., 2020). The DM concentration of potato peels is higher, although the DM content of ginger peels is similar to the levels reported by Mbaeyi Nwaoha et al. (2013). The cell wall of potato peels contains suberin, which functions as a physical barrier and prevents water loss (Graça, 2015; Liang & McDonald, 2014; Singh et al., 2021). The actual amount of fibre, fat, protein, and minerals available to the animals is reflected in the dry matter composition of the diet (Bach Knudsen, 2001; Van Soest et al., 1991). The concentration of minerals or inorganic compounds in the samples that serve as inorganic co-factors for metabolic processes and metabolic impairment is indicated by the ash content (Niyi et al., 2019). Ash content in feed is typically associated with mineral content (Thiex et al., 2012). In this study, cucumber peels exhibited the highest ash content, indicating a higher mineral concentration compared to the other vegetable peels.

The CP content varied significantly among the vegetable peels, with cucumber peels exhibiting the highest value (21.37%), followed by potato (16.69%), ginger (7.82%), and carrot (5.61%) peels. While the CP content in cucumber and potato peels was comparable to or slightly lower than those reported by Uchele et al. (2021), the CP content in ginger peel was similar to that reported by Elizabeth et al. (2013). However, the CP content in carrot peels was significantly higher than that reported for whole carrots by Caro-Cusba et al. (2021). The high CP content in cucumber peels, in particular, suggests their potential as a valuable protein source for ruminant livestock. Adequate protein intake is crucial for livestock health and productivity (Baris, 2023; Desta, 2024; Shelly, 2024). As reported by Desta (2024); and Klasing (2013), protein deficiency can lead to decreased appetite, reduced feed intake, and impaired growth and development in livestock. Additionally, protein is essential for milk production in dairy cattle (Kim & Lee, 2021), and poultry production (Pesti & Choct, 2023). Sufficient protein intake can also improve rumen ammonia (NH3) levels, and blood urea nitrogen (BUN), and reduce urinary nitrogen excretion (Melendez et al., 2003). Therefore, the high CP content in cucumber peels, in particular, suggests their potential as a valuable protein source for ruminant livestock.

The NDF content of the vegetable peels varied significantly, ranging from 34.30% in carrot peels to 86.64% in ginger peels. Notably, the NDF content of carrot peels was lower than that reported for whole carrots (55.9%) by (Caro Cusba et al., 2021). NDF is a critical factor influencing feed intake and digestibility in ruminants (Goulart et al., 2020). It is a measure of the total cell wall content, including cellulose, hemicellulose, and lignin (Jung, 1997; Jung & Lamb, 2004). While rumen microorganisms can degrade cellulose and hemicellulose to provide energy,

high NDF levels can limit feed intake and reduce digestibility (Firkins, 2021; Weimer, 2022). However, NDF also plays a crucial role in rumen health by stimulating saliva production, which helps to buffer the rumen pH and promote microbial activity (Diao et al., 2019; Kumar et al., 2024). Therefore, a balance between NDF content and other nutrients is essential for optimal ruminant performance.

The ADF content varied significantly among the vegetable peels, with ginger peels exhibiting the highest value (43.43%) and potato peels the lowest (25.68%). ADF primarily measures the less digestible components of plant cell walls, including cellulose and lignin (Raffrenato et al., 2017, 2018) As the ADF content of forage increases, its digestibility typically decreases, leading to lower energy value (Marten et al., 2015). Therefore, the high ADF content in ginger peels suggests they may have a lower digestibility than the other peels. However, it's important to note that the specific composition of the ADF fraction (cellulose, hemicellulose, and lignin) can influence its digestibility (Raffrenato et al., 2017; Zhong et al., 2021).

The ADL content varied significantly among the vegetable peels, ranging from 26.67% in potato peels to 44.50% in cucumber peels. ADL is a component of plant cell walls resistant to microbial degradation (Raffrenato et al., 2017). High ADL content is associated with low feed digestibility. The high ADL content in cucumber peels suggests they may have a lower digestibility than the other peels. However, the specific composition of the lignin and its interactions with other cell wall components can influence its impact on feed digestibility (Jung & Allen, 1995). Generally, a high fibre content in the peels could contribute to rumen microbial activity and digestion. However, the high fibre content, especially in ginger peels, may also limit digestibility and energy intake for ruminants.

Table 2. The proximate composition of vegetable peels (DM basis, %).

Analysis	Treatments				*P-values*
	Cucumber	Carrot	Potato	Ginger	
DM	5.50 ± 00^{d}	9.52 ± 00^{b}	14.56 ± 00^{a}	7.89 ± 00^{c}	<.0001
OM	24.81 ± 0.98^{a}	12.96 ± 0.05^{c}	7.95 ± 0.06^{d}	20.00 ± 0.27^{b}	<.0001
CP	21.37 ± 0.23^{a}	5.61 ± 0.05^{d}	16.69 ± 0.19^{b}	7.82 ± 0.18^{c}	<.0001
NDF	70.91 ± 1.16^{b}	34.30 ± 1.54^{c}	65.75 ± 2.42^{b}	86.64 ± 1.13^{a}	<.0001
ADF	43.43 ± 1.41^{a}	31.86 ± 0.92^{b}	25.68 ± 0.90^{c}	42.75 ± 1.05^{a}	<.0001
ADL	44.50 ± 1.36^{a}	32.14 ± 0.96^{b}	26.67 ± 0.44^{c}	40.70 ± 1.06^{a}	<.0001

Means in the same row with different superscripts ($^{a, b, c, d}$) are significantly different ($P < 0.0001$). *DM= Dry Matter, OM= Organic Matter, CP= Crude Protein, NDF= Neutral Detergent Fibre, ADF= Acid Detergent Fibre, ADL= Acid Detergent Lignin

Table 3. *In vitro* gas production of various types of vegetable peels

Incubation time (H)	Treatments				SEM	*P-values*
	Cucumber	Carrot	Potato	Ginger		
2	0.00^{b}	7.50^{a}	1.67^{b}	0.67^{b}	0.972	<.0001
4	0.33^{b}	10.00^{a}	1.00^{b}	1.00^{b}	0.787	<.0001
6	1.33^{b}	13.00^{a}	2.50^{b}	0.50^{b}	1.029	<.0001
19	9.00^{b}	37.67^{a}	9.50^{b}	0.33^{c}	1.108	<.0001
20	11.17^{b}	39.83^{a}	10.17^{b}	0.17^{c}	0.775	<.0001
22	13.17^{b}	42.17^{a}	11.67^{b}	0.50^{c}	0.907	<.0001
24	17.50^{b}	44.67^{a}	15.50^{b}	0.33^{c}	1.072	<.0001
26	20.17^{b}	43.67^{a}	19.33^{b}	0.33^{c}	2.305	<.0001
28	22.50^{b}	45.17^{a}	22.00^{b}	0.33^{c}	1.931	<.0001
48	32.83^{b}	55.83^{a}	42.83^{ab}	0.17^{c}	3.508	<.0001

Means in the same row with different superscripts ($^{a, b, c, d}$) are significantly different (P < 0.0001). SEM- standard error of the mean.

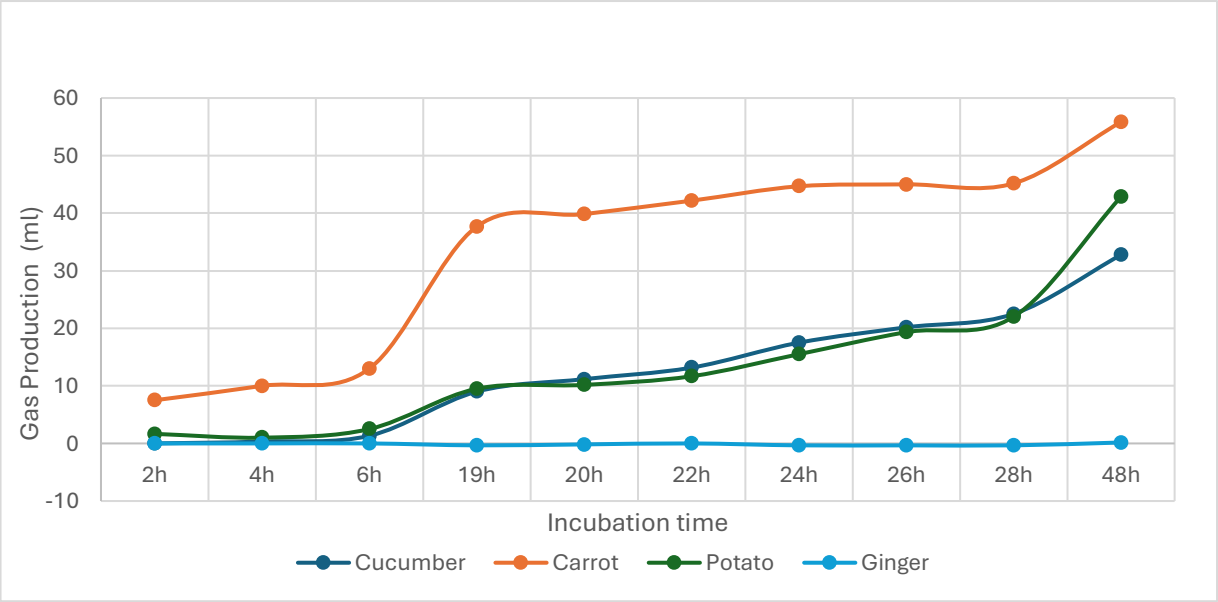

Figure 1. *In vitro* cumulative gas production

Table 4. *In vitro* dry matter digestibility (IVDMD) and *in vitro* neutral detergent fibre digestibility (IVNDFD) of vegetables peels after 48 hours of the incubation period

Analysis	Treatments				*P-values*
	Cucumber	Carrot	Potato	Ginger	
IVDMD (%)	80.37 ± 0.85[b]	91.52 ± 1.06 [a]	87.17 ± 1.11 [a]	40.16 ± 2.45 [c]	<.0001
IVNDFD (%)	77.13 [c]	63.89 [d]	91.78 [b]	99.10 [a]	<.0001

Table 5. The *in vitro* rumen fermentation characteristics of the vegetable peels.

Parameter	Treatments				*P-values*
	Cucumber	Carrot	Potato	Ginger	
pH_{48}	6.67±0.2[a]	6.28 ± 0.03[ab]	6.45 ± 0.05[ab]	6.20 ± 0.02[b]	0.0263
NH_3-N, mg/ml	0.0311±0.0009[ab]	0.0387 ± 0.0048[a]	0.0287 ± 0.0009[ab]	0.0278 ± 0.0012[b]	0.0402
Acetic	116.13±12.24[c]	182.71 ± 9.39[b]	396.89 ± 13.38[a]	43.55 ± 3.73[d]	<.0001
Propionate	418.86±51.15[a]	65.08 ± 0.004[b]	427.51 ± 18.80[a]	79.18 ± 7.72[b]	<.0001
Iso-butyrate	29.75± 4.0[a]	22.74 ± 1.51[ab]	21.02 ± 0.96[ab]	18.07 ± 0.65[b]	0.0096
Butyric	126.71±18.11[a]	114.88 ± 3.96[a]	145.31 ± 4.56[a]	41.24 ± 5.50[b]	<.0001
Iso-valerate	34.78± 5.071[a]	25.82 ± 2.06[ab]	19.80 ± 1.15[b]	23.55 ± 0.54[b]	0.0085
Valerate	19.60±3.77[b]	28.86 ± 1.93[a]	20.15 ± 1.04[b]	4.28 ± 0.47[c]	<.0001
Acetic/Propionic	0.31±0.07[c]	2.81 ± 0.15[a]	0.94 ± 0.022[b]	0.57 ± 0.50[c]	<.0001
Total VFA	746.15±76.49[b]	442.91 ± 14.20[c]	1031.61 ± 37.30[a]	210.44 ± 16.34[d]	<.0001

Means in the same row with different superscripts ([a, b, c, d]) are significantly different ($p<0.0001$). *VFA= Volatile fatty acids

In vitro gas production

In vitro gas production and the rate of gas production per hour did differ ($P < 0.05$) significantly (Table 3). There was a significant ($P < 0.05$) difference among the vegetable peels for in vitro gas production. Overall, the carrot peel produced the most amount of gas, followed by the cucumber and potato peel, while the ginger peel produced the least amount of gas (Table 3). It was obvious that as the incubation time increased from 0 to 48 hours, a linear increase in cumulative gas production was observed for all the vegetable peels (Figure 1). However, carrot peels consistently produced the highest amount of gas, while ginger peels produced the least. From 0 to 6 hours of incubation, the gas production by all of the vegetable peels showed a somewhat flat trend. At 19 hours of incubation, the peels of carrots, potatoes, and cucumbers showed a significant increase in gas production. Nevertheless, throughout the 48-hour incubation period, the ginger peel remains flat. With the exception of the ginger peels, which remained straight all vegetable peels

produced a maximum amount of gas around 48 hours of incubation.

Initial gas production was relatively low for all vegetable peels during the first 6 hours of incubation. Subsequently, a significant increase in gas production was observed, particularly for carrot, potato, and cucumber peels at 19 hours. However, ginger peels exhibited minimal gas production throughout the 48-hour incubation period. The highest gas production was recorded at 48 hours for all treatments except ginger peels.

To ensure the viability of vegetable peels as a ruminant feed, it is crucial to evaluate their impact on rumen fermentation, which directly influences animal performance. Feed digestibility is closely linked to gas production, as high digestibility often correlates with increased microbial activity (Dijkstra et al., 2005; Getachew et al., 2005). After a 48-hour incubation period, carrot peels exhibited the highest gas production. This finding aligns with the results of Shamseldein et al. (2017), who reported that lower fibre content, as observed in carrot peels with low NDF and IVNDFD, is associated with higher gas production. Similar results were obtained for whole carrots. In contrast, ginger peels showed the lowest gas production. This reduced fermentation activity may be attributed to condensed tannins (CT) in ginger peels (Mendel et al., 2017). CTs can exert antimicrobial effects, potentially inhibiting rumen microbial activity and reducing feed digestibility (Chen et al., 2021).

In vitro dry matter digestibility (IVDMD) and in vitro neutral detergent fibre digestibility (IVNDFD)

The IVDMD and IVNDFD of the vegetable peels after 48 hours of incubation showed significant differences ($P < 0.05$) (Table 4). Carrot peels exhibited the highest IVDMD, indicating their higher digestibility, while ginger peels had the lowest IVDMD. Conversely, ginger peels had the highest IVNDFD, suggesting a higher digestibility of fibre components, while carrot peels had the lowest IVNDFD.

Conclusion

The present study evaluated the nutritive value and in vitro rumen fermentation characteristics of four common vegetable peels: cucumber, carrot, potato, and ginger. Significant differences were observed in the chemical composition and fermentation parameters among the vegetable peels. Carrot and potato peels, with their relatively high nutrient content and digestibility, showed promise as potential feed additives for ruminants. However, ginger peels, characterized by high fibre content and the presence of tannins, exhibited lower digestibility and may negatively impact rumen microbial activity. While cucumber peels also showed potential as a feed additive, further research is needed to optimize their inclusion in ruminant diets and to determine their long-term effects on animal performance and health. Additionally, exploring the potential of ginger peels as a feed additive or medicinal feed, in controlled amounts,

may be a promising avenue for future research

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