

Dietary inclusion of almond hull on growth performance, nutrient digestibility, faecal microbiome, faecal score, and noxious gas emissions in weaning pigs

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ABSTRACT. In a 42-day study, 195 [(Landrace × Yorkshire) × Duroc)] piglets, weighing 6.52 ± 0.59 kg, were randomly assigned to three treatment groups with 13 replicates and 5 pigs (3 barrows and 2 gilts) per pen, and used to evaluate the impact of weaning pig diets containing almond hulls (AHs) on growth performance, nutrient utilization, faecal microbiome, noxious gas emissions, and faecal score. The following dietary treatment groups were applied: basal diet as control, basal diet + 3% AH as TRT1; basal diet + 6% AH as TRT2. At the end of the experiment, the average daily gain and gain-to-feed ratio showed an increasing trend. However, despite these changes, the average daily feed intake remained constant, as did nutrient digestibility, faecal microbiome (Salmonella, Escherichia coli, and Lactobacillus), and faecal score. Among the noxious gases, only NH, tended to decrease, while other gases (CO,, H,S, methyl mercaptans, acetic acid) remained stable. These findings indicated that the inclusion of AHs in the diet could improve growth performance and reduce NH, gas production without negative impact on nutrient absorption, microbial populations, and faecal score in weaned piglets.

Introduction

Weaning is a pivotal and challenging period in the pig industry, where various factors contribute to optimal development of animals. Weaned pigs do not have fully developed immune systems, and do not synthesise sufficient amounts of specialised enzymes required to digest the various nutrient components in their diets (Hedemann et al., 2006). However, Low (1985) has indicated that dietary fibre promotes the release of saliva, bile, gastric, and pancreatic fluids, which improves nutrient digestion and the growth of piglets. Almond hulls (AHs), a byproduct of almond kernel processing, may potentially be the best source of dietary fibre, as they constitute 35–62% of the raw almond weight (Prgomet et al., 2017), and their annual production volume amounts to 6 million tons (Li et al., 2018). AHs exhibit the same physical and chemical characteristics as hulls of other cereals, such as oat, rice, and soybean. While researchers have studied how different fibre levels affect pig performance, nutrient digestibility, and microbial composition (Huang et al., 2018), there is not much research on how AHs specifically affect weaning pig nutrition. The addition of moderate amounts of oat hulls has been shown to minimise the probability of diarrhoea and increase the performance of animals administered low-fibre diets (Mateos et al., 2006). Lizardo et al. (1997) found that adequate fibre sources, such as beet pulp, maintain the structural and functional integrity of the digestive system by providing fermentable substrate for flora in the large intestine. The inclusion of a source of insoluble fibre in the diets of piglets improved their intestinal morphology by increasing the length of the villi, leading to improved nutrient digestion (Hedemann et al., 2006). However, Freire et al. (2000) reported that piglets fed diets containing soybean hulls demonstrated reduced growth performance, lower nutrient digestibility, and higher feed-to-gain ratios. Additionally, Mateos et al. (2006) demonstrated that adding oat hulls into the diet did not influence nutrient digestibility, but resulted in decreased feed intake and compromised growth performance of piglets. On the other hand, diets formulated using different fibre sources and inclusion levels, when fed to weaned piglets, can exert different effects on pig performance, nutrient digestibility, and the abundance of faecal microbiota (Yu et al., 2016).

We hypothesised that incorporating lower AH levels in the diet would improve growth performance by reducing noxious gas emissions without adversely affecting digestibility, and faecal score. The objective of this study was to assess the impact of AHs as a feed ingredient on growth performance, nutrient digestibility, faecal microbiome, faecal score, and noxious gas emissions in weaned pigs.

Table 1. Ingredient composition of experimental diets (as-fed basis), %

Material and methods

The research protocol (Approval No. DK-1-2213) and all animal research procedures have been approved by Dankook University's Institutional Animal Care and Use Committee in Cheonan, South Korea.

Experimental design, animals, and diets

In a 42-day trial, 195 crossbred weanling piglets ([Yorkshire × Landrace] × Duroc), weighing 6.52 ± 0.59 kg, were randomly assigned to 3 nutritional treatments. Each treatment comprised 13 replicate pens, with 2 gilts and 3 barrows per pen, based on body weight and sex. The dietary treatment included a basal diet with 0% AH (control), a basal diet with 3% AH (TRT1), and a basal diet with 6% AH (TRT2). The trial period was divided into 3 phases: 0–1 week of age (phase 1), 2–3 weeks of age (phase 2), and 4–6 weeks of age (phase 3). The formulation of the diets in the present experiment (Table 1) was carried out according to the guide-

		Weaning								
Items		Phase 1			Phase 2			Phase 3		
	CON	TRT1	TRT2	CON	TRT1	TRT2	CON	TRT1	TRT2	
Maize	37.62	34.01	30.36	52.03	48.40	44.78	60.85	57.26	53.62	
Soybean meal, 48% CP	18.25	18.50	18.76	16.68	16.94	17.18	19.05	19.29	19.54	
Fermented soybean meal	5.00	5.00	5.00	4.00	4.00	4.00	3.00	3.00	3.00	
Blood plasma protein	5.00	5.00	5.00	3.00	3.00	3.00	2.00	2.00	2.00	
Tallow	2.90	3.29	3.69	2.69	3.08	3.47	2.25	2.64	3.04	
Lactose	13.46	13.46	13.46	7.78	7.78	7.78	3.18	3.18	3.18	
Sugar	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	
Whey protein	11.00	11.00	11.00	7.00	7.00	7.00	3.00	3.00	3.00	
Mono-di-calcium phosphate	1.10	1.12	1.14	1.30	1.32	1.34	1.35	1.35	1.38	
Limestone	0.93	0.88	0.85	0.94	0.90	0.86	1.00	0.96	0.92	
Salt	0.20	0.20	0.20	0.10	0.10	0.10	0.10	0.10	0.10	
Methionine (99%)	0.22	0.22	0.22	0.15	0.15	0.16	0.09	0.09	0.09	
L-lysine (78%)	0.51	0.51	0.51	0.65	0.65	0.65	0.57	0.57	0.57	
Vitamin / Mineral premix*	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	
ZnO	0.38	0.38	0.38	0.25	0.25	0.25	0.13	0.13	0.13	
Choline (25%)	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	0.03	
Almond hull	-	3.00	6.00	-	3.00	6.00	-	3.00	6.00	
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	
Calculated values,%										
crude protein	20.00	20.00	20.00	18.00	18.00	18.00	18.00	18.00	18.00	
metabolizable energy, kcal/kg	3 450	3 450	3 450	3 400	3 400	3 400	3 350	3 350	3 350	
fat	4.56	4.90	5.26	4.79	5.13	5.48	4.66	5.01	5.36	
calcium	0.80	0.80	0.80	0.80	0.80	0.80	0.80	0.80	0.80	
phosphorus	0.60	0.60	0.60	0.60	0.60	0.60	0.60	0.60	0.60	
lysine	1.60	1.60	1.60	1.50	1.50	1.50	1.40	1.40	1.40	
methionine	0.48	0.48	0.48	0.40	0.40	0.40	0.35	0.35	0.35	
lactose	20.00	20.00	20.00	12.00	12.00	12.00	5.00	5.00	5.00	

CON – basal diet; TRT1 – basal diet + 3% almond hull; TRT2 – basal diet + 6% almond hull; CP – crude protein; * provided per kg of complete diet: IU: vit. A 16 800, vit. D₃ 2 400; mg: vit. E 108, vit. K 7.2, riboflavin 18, niacin 80.4, thiamine 2.64, D-pantothenic 45.6, cobalamin 0.06, Cu (as CuSO₄) 12, Zn (as ZnSO₄) 60, Mn (as MnSO₄) 24, I (as Ca(IO₃)₂) 0.6, Se (as Na₂SeO₃) 0.36

lines of NRC (2012). Each pig was provided with a 0.26 m \times 0.53 m area in a controlled environment room equipped with a mechanical aeration system. Throughout the experiment, piglets had unlimited access to water and feed from pens equipped with a nipple drinker and feeder. Artificial light was provided for 12 h daily. During the first week, the ambient temperature of the room was maintained at approx. 30 °C, and then decreased by 1 °C per week thereafter.

Chemical analysis, sampling and measurements

Growth performance

Body weights of piglets were recorded on days 1, 7, 21, and 42 to determine average daily gain (ADG). Average daily feed intake (ADFI) was calculated by measuring the amount of feed consumed and remaining feed in each pen. The gain-to-feed ratio (G:F) was calculated based on the ADG and ADFI values.

Nutrient digestibility

Chromium oxide (Cr₂O₃, 0.20%) was added to the crumbled diets and administered to pigs for 7 days before faecal sample collection. Two pigs (one barrow and one gilt) were selected from each pen after the 42-day experiment to provide faecal samples via rectal palpation. Firstly, samples were pooled on a pen basis, and then a randomly chosen sample was stored at -20 °C until analysis. Faecal samples were dried at 60 °C for 72 h to conduct chemical analysis, followed by grinding to pass through a 1mm screen. The coefficient of apparent total tract nutrient digestibility (CATTD) of dry matter (DM) and nitrogen (N) levels in both the feed and faecal samples was determined according to the AOAC (2000) method. To determine the coefficient of CATTD of energy (E) and excreta samples, samples were transferred to a calorimeter (Parr Instruments, Moline, IL, USA) to assess thermal combustion and chromium content, which was analysed using a Shimadzu UV-1201 atomic absorption spectrophotometer (Shimadzu, Kyoto, Japan). The formula used for apparent total tract digestibility (ATTD) was as follows:

digestibility (%) = $[1 - (Nf \times Cd)/(Nd \times Cf)] \times 100$,

where: Nf – number of nutrients in faeces (% DM), Nd number of nutrients in the diet (% DM), Cd – chromium content in the diet (% DM), and Cf – chromium content in faeces (% DM).

Faecal microbiome

Two pigs from each pen were selected to collect faecal samples through rectal palpation. These samples were promptly transported in an icebox to the laboratory for further analysis of the gut microbiota. Each faecal sample (1 g) was diluted with 9 ml of 1% peptone water and vortexed for thorough mixing. Sequential dilutions from 10^{-1} to 10^{-6} were prepared and 50 µl of each dilution was plated onto three selective agar media: De Man-Rogosa-Sharpe (MRS) agar (Difco Laboratories, Detroit, MI, USA) for Lactobacillus, MacConkey agar (Difco Laboratories, Detroit, MI, USA) for Escherichia coli, and SS agar (Difco Laboratories, Detroit, MI, USA) for Salmonella. Following inoculation, plates were incubated at specific temperatures: Lactobacillus at 39 °C for 48 h, E. coli at 37 °C for 24, and Salmonella at 37 °C for 24 h. Subsequently, colony counts were recorded and the results were presented as \log_{10} converted data.

Noxious gas emission

At the end of the study, fresh faecal samples (300 g) were collected from two pigs in each pen. The samples were placed in a 2.6 l plastic box with small holes on one side and sealed with tape. The boxes were kept at room temperature for 7 days for the fermentation process to occur. After fermentation, gas detection was performed using a MultiRAE Lite PGM-6208 (RAE, USA) to measure the levels of ammonia (NH₂), hydrogen sulphide (H₂S), methyl mercaptans, carbon dioxide (CO_2) , and acetic acid. For these measurements, a hole was made in the adhesive tape covering the box, and 100 ml of air from approximately two centimetres above the faecal surface was sampled. Each box was then resealed with adhesive tape after air sampling. After 48 h, the measurements were carried out again, and the gas contents were determined by averaging two readings from the same box.

Faecal score

Faecal samples were collected and recorded from each pen at 8:00 and 20:00 on days 7, 21, 28, 35, and 42. The faecal score was determined as the average of five pigs in each pen using the following faecal scoring system: 1 - firm, formed stool; 2 - hard, dry pellets; 3 - soft, moist stool that retain its shape; 4 - unformed, soft stool that conform to the shape of the vessel; and 5 - watery, pourable liquids (Ahammad et al., 2023).

Statistical analyses

The data were analysed using the GLM technique implemented in the SAS software (SAS, 2014). Data representing growth performance, nutrient digestibility, faecal score, faecal microbiome, and noxious gas emissions were analysed on a pen basis. Linear, and quadratic polynomial contrasts were applied to examine the effect of dietary treatment. The standard error of the mean (SEM) was used to account for variability of the data, with the level of significance determined at P < 0.05 and tendencies identified at P < 0.10.

Results

In week 3, there was a tendency for ADG to improve, with a significant increase observed at week 6. Throughout the entire period, the gain-tofeed (G:F) ratio exhibited an upward trend, while ADFI remained unaffected in all phases (Table 2). There were no significant effects on digestibility of DM, nitrogen level, and energy across all nutrients (Table 3). The inclusion of almond hulls exhibited a trend towards reducing ammonia gas discharge, with no discernible effects on H_2S , methyl mercaptans, acetic acid, and CO_2 (Table 4).

 Table 2. Effect of dietary almond hull supplementation on growth performance in weaning pigs

Items	CON	TRT1	TRT2	SEM	P-value		
nems	CON	IKII	IRIZ	SEIVI	linear	quadratic	
Week 0-1							
ADG, g	261	263	265	5	0.6536	0.9402	
ADFI, g	289	291	292	6	0.6975	0.9880	
G:F	0.904	0.905	0.907	0.005	0.7177	0.8454	
Week 2–3							
ADG, g	369	372	385	6	0.0968	0.5355	
ADFI, g	456	459	474	8	0.1248	0.5330	
G:F	0.809	0.811	0.811	0.006	0.7989	0.9393	
Week 4-6							
ADG, g	516	529	541	8	0.0373	0.9939	
ADFI, g	719	729	740	11	0.1940	0.9565	
G:F	0.718	0.726	0.731	0.009	0.3318	0.8953	
Overall							
ADG, g	425	432	443	7	0.0678	0.8626	
ADFI, g	560	566	577	8	0.3519	0.9374	
G:F	0.783	0.788	0.800	0.006	0.0569	0.6374	

CON – basal diet; TRT1 – basal diet + 3% almond hull; TRT2 – basal diet + 6% almond hull; ADG – average daily gain; ADFI – average daily feed intake; G:F – gain-to-feed ratio; SEM – standard error of the mean; P < 0.05 indicates that data are significantly different

Table 3. Effect of dietary almond hull supplementation on coefficients of total tract digestibility in weaning pigs, %

Items	CON		тото	CEM	P-value	
Items	CON	ON TRT1 TRT2 SEM		SEIVI	linear	quadratic
Dry matter	83.68	83.87	84.19	0.76	0.6410	0.9510
Nitrogen	80.27	80.84	81.19	1.05	0.5444	0.9367
Digestible energy	82.42	83.49	83.56	1.25	0.5279	0.7490

CON – basal diet; TRT1 – basal diet + 3% almond hull; TRT2 – basal diet + 6% almond hull; SEM – standard error of the mean; P > 0.05 indicates that data are not significantly different

 Table 4. Effect of dietary almond hull supplementation on gas emission in weaning pigs, ppm

Items	CON	TRT1	тото	OEM	P-value	
	CON	INII	IRIZ	SEIVI	linear	quadratic
NH ₃	2.88	2.75	2.13	0.18	0.0509	0.4316
H₂S	2.20	2.13	2.28	0.43	0.3405	0.8014
Methyl mercaptans	3.88	3.75	3.50	0.55	0.3757	0.7253
Acetic acid	7.38	7.00	7.13	0.26	0.2283	1.0000
CO ₂	0.783	0.788	0.800	190	0.4366	0.8779

CON – basal diet; TRT1 – basal diet + 3% almond hull; TRT2 – basal diet + 6% almond hull; SEM – standard error of the mean; P < 0.05 indicates that data are significantly different

Dietary AH supplementation demonstrated no significant influence on the abundance of *Lactobacillus*, *Salmonella*, and *E. coli* populations (Table 5). Furthermore, the addition of up to 6% AH to the diet of weaning pigs did not cause any changes in the faecal score during this feeding trial (Table 6).

Table 5. Effect of dietary almond hull supplementation on faecal microflora in weaning pigs, $log_{1n}cfu/g$

Items	CON		TRT2	OEM	P-value		
Items	CON	IKII	IRIZ	SEIVI	linear	quadratic	
Lactobacillus	9.17	9.21	9.22	0.04	0.3904	0.8063	
E. coli	6.27	6.23	6.20	0.05	0.3686	0.9604	
Salmonella	3.90	3.88	3.86	0.14	0.8384	0.9687	

CON– basal diet; TRT1 – basal diet + 3% almond hull; TRT2 – basal diet + 6% almond hull; SEM – standard error of the mean; P > 0.05 indicates that data are not significantly different

 Table 6. Effect of dietary almond hull supplementation on faecal score in growing pigs

Itomo	Faeca	I score ¹			P-value	
Items	CON	TRT1	TRT2	SEM	linear	quadratic
Initial	3.30	3.29	3.30	0.03	0.5094	0.5673
Week 1	3.26	3.26	3.25	0.03	0.6987	0.5039
Week 3	3.25	3.25	3.24	0.02	1.0000	0.8027
Week 6	3.23	3.22	3.20	0.04	0.4449	0.7902

CON – Basal diet; TRT1 – CON + 3% almond hull; TRT2 – CON + 6% almond hull; SEM – standard error of the mean; P > 0.05 indicates that data are not significantly different; ¹ Faecal score = 1 – hard, dry pellet; 2 – firm, formed stool; 3 – soft, moist stool that retains shape; 4 – soft, unformed stool that assumes shape of container; 5 – watery liquid that can be poured

Discussion

High levels of insoluble fibre in AHs, including pectin, hemicellulose, cellulose, and lignin, have been associated with increased body weight in animals (Holtman et al., 2015). However, research on AH supplementation in the diets of weaned pigs is scarce, making comparisons between studies challenging. Therefore, we are discussing the effects of other hulls with similar physical and chemical structure to AH in weaning pigs or other animals with similar digestive systems. The current findings showed that supplementing AH in the diet improved the ADG of weaned pigs, which was consistent with findings by Ani et al. (2013), who observed increased ADG and BWG in weaning pigs with the inclusion of 10% soybean hull in the pig diets. Similarly, moderate additions (1.5-8%) of insoluble dietary fibre sources were shown to increase ADFI and ADG in weaned piglets (Flis et al., 2017). Clouard et al. (2018) observed a significant influence on growth performance of weaned piglets with three dietary fibre treatments: 2% oat, 1.5-2% lignocellulose, and 4-8% coarse wheat bran. However, dietary inclusion of 5% cellulose, an insoluble fibre source, had no impact on body weight and feed intake of weaned piglets. In addition, Freire et al. (2000) reported that piglets receiving diets containing 200 g/kg of soybean hulls exhibited reduced growth performance, decreased nutrient digestibility, and a higher G:F ratio. Excessive dietary fibre intake has been associated with growth impairment in monogastric animals (Mateos et al., 2012). Therefore, it is plausible to suggest that incorporating AH fibre into the diet may be beneficial for improving pig growth during the weaning period. Discrepancies between our findings and those of other studies could be attributed to factors such as differences in environmental conditions, pig breeds, diverse developmental stages, fibre sources, and levels of hull inclusion analysed in individual works.

In the present study, the inclusion of AH did not significantly impact the digestibility of DM, N, and E in the diet of weaning pigs. Similarly, Jeaurond et al. (2008) found that the incorporation of 5% sugar beet pulp to pig diets did not affect the digestibility of DM and N levels. Mateos et al. (2006) reported that digestibility of DM, N, and E were not affected when 4% cooked and expanded oat hulls were supplemented in the diet of weaned pig. Conversely, Chen et al. (2020) found that adding 1% insoluble fibre in the diet may enhance nutritional digestion in the first two weeks after weaning. In addition, Pieper et al. (2012) noted that the inclusion of 5% sugar beet pulp and 8% wheat bran decreased DM digestibility without affecting N and E digestibility. Different results obtained in the present study compared to other studies might be due to changes in pig physiology during weaning, as well as differences in environmental conditions, pig breeds, dietary fibre sources, and levels of hull inclusion.

Pigs are one of the primary groups within the livestock sector, and offensive gas emissions from pig farms, such as NH₃, H₂S, and total mercaptans cause significant airborne pollution (Lesschen et al., 2011). Our current research is in agreement with prior studies investigating various fibre sources, including insoluble fibre (Schedle et al., 2008), soluble dietary fibre (Jeaurond et al., 2008), and mixed fibre (Pieper et al., 2012), all of which demonstrated a reduction in ammonia gas emissions as a result of these supplements. It has been previously shown that lignin and/or lignin-cellulose complexes were able to alter the fermentation pattern and/or provide molecules that bind ammonia, thereby mitigating ammonia emissions (Schedle et al., 2008). In addition, dietary sources rich in fermentable fibre notably lower faecal and slurry pH by increasing volatile fatty acid (VFA) production in the large intestine, consequently reducing NH₂ emissions (Canh et al., 1998). Another reason for incorporating fibre sources in pig diets is that there is evidence that it results in a reduction in NH₂ emissions and simultaneous improvement in growth performance. This due to the fact that the intricate relationship between pig growth and decreased ammonia gases involves mechanisms such as enhanced nutrient utilisation, a healthier gut microbiota, modified faecal composition, reduced microbial fermentation, optimised digestive processes, and a well-balanced overall diet formulation (Ngoc et al., 2021). Thus, the current study demonstrated that including AHs had a beneficial effect on ADG and G:F ratio, which could be responsible for the lower ammonia emissions.

The content and composition of dietary fibre influence the microbial composition and activity in the gut (Verschuren et al., 2018). Consequently, diets supplemented with insoluble and soluble dietary fibre substrates have been shown to increase the abundance of beneficial gut microbiota (Flis et al., 2017). In addition, dietary fibre was also shown to promote the growth of favourable bacteria such as *Bifidobacteria* and *Lactobacilli*, while suppressing the development of potentially harmful bacteria like *E. coli* and *Clostridia* (Gibson et al., 1995). Our current study demonstrated that dietary inclusion of AHs exerted no significant effect on *Lactobacillus, E. coli*, and *Salmonella* populations. These findings contrasts with the results reported by Chen et al. (2020), who found that supplementing 1% insoluble dietary fibre and 0.5% soluble dietary fibre in the diet increased the count of *Lactobacilli*, but there was no significant difference in the size of *Bifidobacterium, Bacillus, E. coli* or total bacterial populations in the ileum and caecum digesta. Thus, the effect of AHs on faecal microorganisms requires further study due to these inconsistent results. However, there is currently no available literature on the effect of AHs specifically in weaning pigs to be able to clarify this discrepancy.

The faecal score is one of key indicator for assessing the health of piglets' digestive systems, with a higher score indicating a greater incidence of diarrhoea. In the present study, no instances of diarrhoea were observed based on the faecal score evaluation. Our findings were consistent with those of Wellock et al. (2008), who found that the inclusion of soluble NSP (inulin), and insoluble NSP (pure cellulose) at varying levels did not affect the mean faecal score from day 3 to day 14 after weaning. Moreover, the addition of 7.5% or 3% soybean hulls (Pascoal et al., 2012), and 9% citrus pulp (Pascoal et al., 2012) was shown to have no effect on the incidence of postweaning diarrhoea (PWD). On the other hand, the addition of insoluble dietary fibre-rich sources, such as oat hulls (Mateos et al., 2006) or pure cellulose (Pascoal et al., 2012) has been shown to improve the faecal score and reduce the prevalence of PWD. Furthermore, the inclusion of a small number of insoluble fibre sources in the diets for young pigs, which optimised their health and hygiene status, has been associated with a reduced prevalence of PWD during the first two weeks (Molist et al., 2014). Although the faecal score in our current investigation was not significantly improved, its value indicated that the weaning piglets were not affected by diarrhoea. Insoluble fibres are known to decrease diarrhoea incidence due to their water-binding capacity (Chen et al., 2020). Interestingly, the absence of diarrhoea was observed not only in the treatment groups (TRTs) but also in the control group (CON), with no significant difference in diarrhoea incidence between them. This suggests that factors not related to water-binding capacity may contribute to diarrhoea prevention in this study. Further investigation is necessary to elucidate the specific mechanisms involved in preventing diarrhoea observed in both groups.

Conclusions

Increasing the proportion of almond hulls in the diets, particularly at a level of 6%, demonstrated a more favourable impact on average daily gain, and gain-to-feed ratio, as well as prevented environmental pollution by reducing ammonia gas emissions in weaned pigs. Substituting maize with almond hulls in the pig industry could potentially offer economic advantages.

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Conflict of interest

The Authors declare that there is no conflict of interest.

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