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Performance and digestibility of low initial body of weight broilers fed infertile egg powder

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ABSTRACT

Infertile egg powder (IEP) is an alternative feed ingredient that is rich in protein and derived from hatchery waste. This study aimed to evaluate the effects of incorporating IEP into the diets of low initial body weight broilers on growth performance, nutrient digestibility, ammonia emissions, and footpad health. A completely randomized design (CRD) was employed with three treatments: low-weight DOC on a basal diet (T0), normal-weight day-old chick (DOC) on a basal diet (T1), and low-weight DOC on a basal diet supplemented with 4% IEP (T2), each with eight replications. Results showed that T2 broilers had significantly higher body weight gain, final body weight, cumulative feed intake, and performance index compared to T1 and T0 throughout both starter and finisher phases ($P < 0.05$). Crude protein and fat digestibility were also significantly improved in T2. However, no significant differences were observed in feed conversion ratio (FCR) across treatments. Furthermore, T2 exhibited the highest litter ammonia concentration among the experimental groups. The study concluded that IEP can effectively enhance growth and nutrient utilization in underweight broilers, offering a sustainable solution for optimizing poultry production, provided that environmental management practices are concurrently improved.

Introduction

Production of broiler chicken is one of the most vibrant areas in livestock sector, which contributes to the supply of meat in the world. In 2024, the world production of chicken meat was estimated at 103.72 million metric tons which was a slight increase as compared to the previous year because of the increase in the major producers such as Brazil and China which was offsetting the losses in other regions (USDA, 2024). Broiler farming productivity and profitability is based on a number of factors such as genetics, hatchery management, nutrition and environmental conditions. Initial body weight of day-old chicks (DOC) is one of the most important factors in broiler performance. The heavier chicks are more likely to survive, grow quicker and utilize the feed more effectively in comparison with the lighter chicks (Leandro *et al*., 2006). Nevertheless, because of the differences that occur during the hatching process, a certain proportion of DOCs may have poor body weights, which may result in reduced rates of growth and the predisposition to disease (Sozcu and Ipek, 2017)

DOCs with low weight tend to have poor yolk absorption resulting in poor nutrient reserves during the most important early post-hatching period (Vieira and Moran, 1999). This is the case since the yolk sac, which is a source of vital nutrients, is not completely used and thus there is insufficient energy and protein needed to support early growth. In fact, the poor absorption of yolks might negatively impact the maturation of the gastrointestinal tract, which has a poor impact on the digestibility of nutrients and growth (Proszkowiec-Weglarz *et al*., 2020; Wang *et al*., 2020; Nalle *et al*., 2021). Moreover, the absence of an adequate absorption of the yolk may lead to the problems with the development of the gut microbiota and maturation of the intestinal barrier that can precondition the infections and gastrointestinal disorders (Noy *et al*., 2001). Moreover, the lack of nutrient reserves may undermine metabolic processes because the vital organs such as liver and intestines are not nourished as much, thus the early physiological adaptation is impaired. According to Vieira and Moran (1999), these constraints result in lower growth rates, increased mortality, and poor feed efficiency and hence nutritional interventions are essential to help them during their early stage of life.

Infertile eggs may be used as source of protein in broiler diets, after processing to powder (El-Deek *et al*., 2011). Suprayogi *et al*. (2023) mention that IEP has 5454.9 kcal/kg metabolizable energy, 31.47% crude protein, 30.10% crude fat, 0.59% crude fiber, 1.99% ash, 0.55% calcium, and 0.18% phosphorus. Proteins, lipids, antibodies, bioactive constituents, and lysozyme are present in infertile eggs (Al-Harthi *et al*., 2011). Eggs also have different amino acids both essential and non-essential which are significant to broiler chickens. Histidine, lysine, methionine, threonine, leucine, isoleucine, valine, phenylalanine and tryptophan are essential amino acids in eggs (Gruhn and Hennig, 2009). Threonine in IEP is directly linked to improved gut health that in turn promotes optimum growth and performance in broilers. In addition, bioactive components like lysozyme, avidin, and phosvitin have other functions in enhancing the health of the intestine by acting as antimicrobial, antioxidant and nutrient-binding (Anton *et al*., 2006; Xu *et al*., 2012). Thus, IEP supplementation of broiler diets does not only provide high-quality protein but also improves digestion and performance through synergistic effects of its bioactive components and essential amino acids (Suprayogi *et al*., 2023). Broiler diets supplemented with infertile egg powder (IEP) have been reported to promote the growth of important body tissues, including muscle, bones and immune organs (Lei and Kim, 2013). Furthermore, Suprayogi *et al*. (2023) determined that the addition of 5% IEP to the diet positively affected the growth performance of broilers to a significant extent.

Although the overall positive effects of IEP on broiler performance have been previously investigated, little research has been done on the specific potential of IEP as a compensatory nutritional approach to low-weight DOC an emerging problem in commercial hatchery practice. Thus, this research presents a new angle since it is based on the idea of how

IEP supplementation can reduce the early developmental disadvantage in underweight broiler chicks. This research aimed at determining the impact of adding IEP to the diet of broiler chicks of low initial body weight. Specifically, the researchers made an emphasis on the impact on growth performance, nutrient digestibility, ammonia emissions and footpad dermatitis, which are all major indicators of broiler welfare and production efficiency.

Materials and methods

Ethical approval

Research procedures were approved by the Research Ethics Committee of the Faculty of Animal and Agricultural Sciences, Universitas Diponegoro, (nr. 60-10/A-19/KEP-FPP).

Preparation of infertile egg powder

Infertile eggs were collected from a local hatchery ensuring they had intact shells, no embryonic development, and were not spoiled. The eggs were manually separated from shells, homogenized, mixed with 10% rice bran, and freeze-dried. The resulting powder contained 34.3% crude protein, 26.9% crude fat, and 4687 kcal/kg metabolizable energy (Table 1).

Table 1. Nutrient contents of IEP.

Nutrient	Content
Metabolizable energy (kcal/kg)[1]	4687
Crude protein (%)	34.3
Crude fat (%)	26.9
Crude fiber (%)	2.39
Ash (%)	2.1

[1]Calculated according to Bolton (1967): 40.81 {0.87 [crude protein + 2.25 crude fat + nitrogen-free extract] + 2.5.

Birds, diets, and experimented design

The current study used 264 day-old-broiler chicks (DOC) of Cobb JP 313 strain. The chicks were vaccinated against Newcastle disease and infectious bronchitis at the hatchery by a spraying technique. In the case of starter (0-21 days; Table 2) and finisher (22-31 days; Table 3) diets, diets were formulated based on the Cobb 500 nutrient requirements. The experimental layout was composed of three treatment groups with 8 replications each to make 24 experimental units. These were T0 (normal-weight DOC fed basal diet), T1 (low-weight DOC fed basal diet) and T2 (low-weight DOC fed diet with 4 percent IEP). Each replicate was consisting of 10 chicks.

Management and data collection

During brooding (first 7 days), chicks were placed under infrared bulbs for heat. From day 1, birds were distributed to experimental pens according to treatments. Feed and water were provided *ad libitum*.

Growth performance measurement

The parameters that were measured in broiler chicken were feed intake, body weight gain, feed conversion ratio (FCR), performance index (PI) and income over feed cost (IOFC). The amount of feed was quantified weekly by weighing the amount of feed provided and deducting the leftover feed (Razak *et al*., 2016). The difference between the final body weight and the initial body weight was used to calculate body weight gain (Rusli *et al*., 2019). The formula to calculate the FCR was the total feed intake/body weight gain (Siregar, 2017). The PI was employed to measure overall production efficiency and it was determined by using the formula outlined by Febrianto *et al*. (2023):

PI =((100-mortality (%)) x live body weight (kg))/"FCR x age at harvest (days)" x 100

Table 2. Ingredients and nutritional compositions of the experimental diets of broilers (day 1-21).

Ingredients (%)	T0, T1	T2
Yellow corn	53.5	55.09
Palm oil	2.32	-
Soybean meal	40.13	36.86
DL-methionine	0.19	0.19
Bentonite	0.75	0.75
Limestone	1	1
Monocalcium phosphate	1.3	1.3
Premix[1]	0.34	0.34
Choline chloride	0.07	0.07
Salt	0.4	0.4
Infertile egg powder	-	4
Calculated compositions (%, unless otherwise noted)		
ME[2] (kcal/kg)	2900	2900
Crude protein	22	22
Crude fat	2.4	3.4
Crude fiber	5.5	5.5
Ca	1.1	1.2
P (available)	0.6	0.6
Analyzed compositions (%, unless otherwise noted)		
ME[2] (kcal/kg)	2947	2949
Crude protein	22.3	22.2
Crude fiber	3.88	2.79
Crude fat	2.69	2.2
Ash	0.6	0.6

[1]The following nutrients are provided per kilogram of feed: 1,100 mg Zn, 1,000 mg Mn, 75 mg Cu, 850 mg Fe, 4 mg Se, 19 mg I, 6 mg Co, 1,225 mg K, 1,225 mg Mg, 1,250,000 IU vitamin A, 250,000 IU vitamin D3, 1,350 g pantothenic acid, 1,875 g vitamin E, 250 g vitamin K3, 250 g vitamin B1, 750 g vitamin B2, 500 g vitamin B6, 2,500 mg vitamin B12, 5,000 g niacin, 125 g folic acid and 2,500 mg biotin.
[2]ME (metabolizable energy) was calculated according to formula: 40.81 {0.87 (crude protein + 2.25 crude fat + nitrogen - free extract) + 2.5}.
T0: normal-weight DOC receiving control diet; T1: low-weight DOC receiving control diet and T2: low-weight DOC receiving diet containing 4% infertile egg powder.

Nutrient digestibility and digesta passage rate

Digestibility of protein, fiber, fat, and rate of passage of digesta was determined by the total collection method. The collection of the total excreta was done by the Fe_2O_3 marker method. Only one bird was taken in every unit of the experiment. The feces excreted were sprayed with 0.1 N HCl after every two hours to avoid loss of nitrogen. Weighed before and after sun-drying, the excreta were used to obtain wet and air-dry weights. The dried excreta were crushed, and then proximate analysis was done. The formula proposed by Santia *et al*. (2019) was used to calculate crude protein digestibility (%), fat digestibility (%) was determined based on the formula by Krismiyanto *et al*. (2014). However, apparent Metabolizable Energy (AME) was calculated following the method of Sibbald (1975).

Meanwhile, the digesta passage rate was measured using the total collection method with 0.5% Fe_2O_3 as an external marker by observing the first appearance of red-colored feces. The digesta passage rate value was then determined by calculating the average time difference between the administration of the marked feed and the appearance of the first red-colored excreta.

Table 3. Ingredients and nutritional compositions of the experimental diets of broilers (day 22-32).

Ingredients (%)	T0, T1	T2
Yellow corn	59	60.55
Palm oil	4.7	2.4
Soybean meal	32.25	29
DL-methionine	0.19	0.19
Bentonite	0.75	0.75
Limestone	1	1
Monocalcium phosphate	1.3	1.3
Premix[1]	0.34	0.34
Choline chloride	0.07	0.07
Salt	0.4	0.4
Infertile egg powder	-	4
Calculated compositions (%, unless otherwise noted)		
ME[2] (kcal/kg)	3100	3101
Crude protein	19	19
Crude fat	2.5	3.5
Crude fiber	5.4	5.5
Ca	1.1	1.1
P (available)	0.6	0.6
Analyzed compositions (%, unless otherwise noted)		
ME[2] (kcal/kg)	3101	3091
Crude protein	20.9	20.6
Crude fiber	3.19	3.7
Crude fat	3.08	2.9
Ash	4.78	5.27

[1]The following nutrients are provided per kilogram of feed: 1,100 mg Zn, 1,000 mg Mn, 75 mg Cu, 850 mg Fe, 4 mg Se, 19 mg I, 6 mg Co, 1,225 mg K, 1,225 mg Mg, 1,250,000 IU vitamin A, 250,000 IU vitamin D3, 1,350 g pantothenic acid, 1,875 g vitamin E, 250 g vitamin K3, 250 g vitamin B1, 750 g vitamin B2, 500 g vitamin B6, 2,500 mg vitamin B12, 5,000 g niacin, 125 g folic acid and 2,500 mg biotin.
[2]ME (metabolizable energy) was calculated according to formula: 40.81 {0.87 (crude protein + 2.25 crude fat + nitrogen - free extract) + 2.5}.
T0: normal-weight DOC receiving control diet; T1: low-weight DOC receiving control diet and T2: low-weight DOC receiving diet containing 4% infertile egg powder.

Ammonia emission

Ammonia levels were determined on day 30 in the morning, afternoon and evening by using KXL-803 ammonia detector (manufactured by Xinxiang Chaosheng Electric Equipment, China). The sampling of litter was done by taking 100 grams of the experimental unit and then putting them along with an ammonia detector in a clip plastic bag. The ammonia sensor is going to be triggered when the NH_3 concentration surpasses 25 ppm.

Footpad dermatitis

The evaluation of footpad dermatitis was performed by sampling one broiler per experimental unit and then scoring the footpad health standards of the Aviagen company (Jong and Harn, 2012). Score 0 No lesion, or small superficial lesion only, slight discoloration on a small part of the footpad, mild hyperkeratosis (thickening of the outer layer of skin), or healing lesion; Score 1 Mild lesion, discoloration of the footpad, superficial lesion, presence of dark papillae, and hyperkeratosis; Score 2 Severe lesion, affected foot skin with ulcers or scabs, signs of hemorrhaging, or swollen footpads.

Statistical analysis

Data were analyzed with SPSS version 25.0. Data were presented as means and subjected to orthogonal contrast analysis. Differences were considered statistically significant at 5%.

Results and Discussion

Growth performance

At the starter period (1 21 days) low-weight DOC on basal diets (T1) had lower final body weight and feed intake than normal-weight DOC (T0), but there was no significant difference in body weight gain ($P > 0.05$). The results confirm previous assertions in the introduction that

Table 4. Production traits of broiler chicks fed treatment diets.

Items	Treatment groups			SEM	*P value*		
	T0	T1	T2		T0 *vs.* T1	T0 *vs.* T2	T1 *vs.* T2
Starter (1-21)							
BWG (g)	779	745	815	9.50	0.092	0.069	0.001
FBW (g)	821.75	779	849.75	9.53	0.039	0.155	0.001
FI (g)	1027	969	1078	12.98	0.023	0.041	<0.001
FCR	1.32	1.30	1.32	0.01	0.294	0.806	0.200
PI	296.54	285.40	306.28	3.82	0.213	0.275	0.026
Finisher (22-32)							
BWG (g)	735	697	748	10.61	0.138	0.596	0.050
FI (g)	1323	1311	1369	8.63	0.498	0.017	0.004
FCR (g)	1.81	1.88	1.84	0.026	0.269	0.589	0.583
PI	302.54	281.15	326.45	6.91	0.161	0.119	0.002
Cumulative (1-32)							
BWG (g)	1597	1536	1667	15.4	0.025	0.004	<0.001
FBW (g)	1742	1698	1914	25.2	0.263	<0.001	<0.001
FI (g)	2350	2281	2447	20.6	0.083	0.020	<0.001
FCR	1.47	1.48	1.46	0.01	0.554	0.594	0.266
PI	369.50	356.05	409.50	6.48	0.251	0.002	<0.001

BWG: body weight gain, calculated as final body weight minus initial body weight; FBW: final body weight; FI: feed intake, calculated as the total amount of feed consumed); FCR: feed conversion ratio, calculated as the feed intake per unit gain; PI: performance index
T0: normal-weight DOC receiving control diet; T1: low-weight DOC receiving control diet and T2: low-weight DOC receiving diet containing 4% infertile egg powder.
SEM: standard error of the means.

reduced initial body weight reduces early growth performance because of physiological immaturity, including the failure to absorb yolk, and slow development of gastrointestinal tract (Chamblee *et al*., 1992; Iji *et al*., 2001; Van de Ven *et al*., 2012). Adding infertile egg powder (IEP) to the diets of low-weight DOC (T2) resulted in a marked increase in their growth performance in comparison with T1. At starter stage, T2 birds had a significant increase in body weight gain, final body weight, feed intake and performance index (P < 0.05). It is assumed that these improvements are predetermined by the high digestibility and nutritional value of IEP, which includes highly digestible proteins and functional bioactive compounds (lysozyme, immunoglobulins, phosvitin) (Kovacs-Nolan *et al*., 2005; Nasution *et al*., 2018; Ratriyanto *et al*., 2020). These are active compounds that promote the growth of the guts, enhance the immune system and enhance nutrient absorption. Lysozyme is an antimicrobial enzyme that cleaves the 14 glycosidic bonds of the peptidoglycan, especially Gram-positive bacteria in the intestines. It is able to decrease the microbial load of the gastrointestinal tract and support the integrity of mucosa in poultry (El-Ratel *et al*., 2024). Specifically, immunoglobulins (e.g., IgY derived from eggs) target antigens and toxins in the gut, preventing the colonization of pathogens and neutralizing dangerous substances, which, in turn, positively affect the state of the gut and decrease inflammatory reactions (Schade *et al*., 2005). A highly phosphorylated egg yolk protein, phosvitin, has antioxidant properties because it chelates free iron ions and thereby prevents lipid peroxidation in the gastrointestinal tract. Studies in vitro and poultry have shown that peptides of phosvitin have the potential to increase the activity of antioxidant enzymes in intestinal cells and decrease markers of oxidative stress. This enhances integrity of epithelial cells and helps in regeneration of mucosa in broiler chicken (Katayama *et al*., 2006). Collectively, these elements result in a more stable intestinal environment through the modulation of gut microbiota, lower inflammation, and epithelial repair. This results in better villus morphology and increased nutrient absorption- which are important in the promotion of compensatory growth in low-weight DOC. Compensatory growth is the accelerated response to growth that occurs following a period of early-life growth retardation that allows broilers to match their normal-weight counterparts. Such biological reaction is facilitated by the increased feed efficiency, digested nutrients, and metabolic changes which focus on tissue deposition and muscle development when favourable conditions are regained, e.g., nutrient-rich diets (Hornick *et al*., 2000; Tolkamp *et al*., 2011). Such catch-up mechanisms can therefore be facilitated by the bioactive compounds in IEP which create a favourable environment to take place.

When the finisher phase (22 32 days) was reached, T2 still had a significantly better body weight gain and feed intake compared to T1 (P < 0.05) but no differences in FCR across the groups were recorded. The index of performance of T2 also surpassed T1 (P < 0.05). In cumulative (1-32 days), T2 broilers showed greatest BWG and feed intake which significantly exceeded T0 and T1 (P < 0.05). Their final body weight and performance index were also much improved, pointing out the capacity of IEP to offer compensation to initial developmental limitations. The strategic value of IEP as a functional feed ingredient capable of supporting the adverse effects of poor chick quality is underscored by the fact that performance in T2 was better at all stages of growth.

Nutrient digestibility and digesta passage rate

This increase in nutrient digestibility was especially pronounced in low-weight DOC broilers fed IEP (T2) as they had a much greater crude protein digestibility when compared to broilers in the basal diet group (T1) (P < 0.05). This implies that IEP supplementation is a good way of increasing protein utilization in underweight chicks, probably due to better gut performance and enzyme activity. Also, T2 showed greater crude fat digestibility than T0 and T1 (P < 0.05), which means that the lipid composition and emulsifying capacity of IEP may be applied to the more efficient digestion of fat. On the other hand, the treatment groups did not differ significantly with regards to apparent metabolizable energy (AME) and passage rate of digesta, which means that even though IEP increased the utilization of macronutrients, it did not alter the availability of energy or the rate of passage of digesta during the period of observation.

These results support the notion that IEP increases the intake of nu-

Table 5. Nutrient Digestibility and Digesta Passage Rate.

Items	Treatment groups			SEM	*P value*		
	T0	T1	T2		T0 *vs*. T1	T0 *vs*. T2	T1 *vs*. T2
Crude protein (%)	79.36	78.42	80.99	0.51	0..436	0.18	0.04
Crude fat (%)	64.01	61.55	69.82	1.16	0.30	0.02	0.00
AME (kcal/kg)	2296	2238	2255	36.03	0.54	0.66	0.85
Digesta passage rate (minute)	208.62	192.5	207.87	7.21	0.38	0.97	0.40

AME: apparent metabolizable energy
T0: normal-weight DOC receiving control diet; T1: low-weight DOC receiving control diet and T2: low-weight DOC receiving diet containing 4% infertile egg powder.
SEM: standard error of the means.

Table 6. Ammonia Level.

Items	Treatment groups			SEM	*P value*		
	T0	T1	T2		T0 *vs*. T1	T0 *vs*. T2	T1 *vs*. T2
NH_3 litter (ppm)	22.75	23.62	29.75	0.62	< 0.001	0.03	< 0.001

T0: normal-weight DOC receiving control diet; T1: low-weight DOC receiving control diet and T2: low-weight DOC receiving diet containing 4% infertile egg powder.
SEM: standard error of the means.

Table 7. Footpad Dermatitis Score.

Items	Treatment groups			SEM	*P value*		
	T0	T1	T2		T0 *vs*. T1	T0 *vs*. T2	T1 *vs*. T2
Footpad lesion	1.5	1.13	1.38	0.12	0.13	0.61	0.31

T0: normal-weight DOC receiving control diet; T1: low-weight DOC receiving control diet and T2: low-weight DOC receiving diet containing 4% infertile egg powder.
SEM: standard error of the means.

trients. Better digestibility was observed in the group of low-weight DOC receiving IEP, which was probably related to the lack of anti-nutritional factors and high bioavailability of the nutrients of the egg origin. All these make the enzymatic hydrolysis and nutrient absorption in the small intestine maximized (Suprayogi *et al*., 2023). The digestive enzyme production and mucosal immunity strengthening by bioactive components of IEP also contribute to the compensatory growth of T2 birds (Nasution *et al*., 2018; Ratriyanto *et al*., 2020).

The positive influence of the digestibility of fat in T2 can be explained by the high lipid content of IEP that stimulates the excretion of bile and lipase and contributes to emulsification and fat digestion (Ratriyanto *et al*., 2020). In addition, the phospholipids in the egg yolk possess emulsifying qualities that increase the production of the micelles, which boosts the solubility and transfer of the lipids throughout the enterocytes. The absence of the difference between AME and passage rate can be associated with the same amount of energy availability in treatments because of the homogenous fiber and feed composition (Fouad and El-Senousey, 2014).

Environmental and welfare implication

Ammonia concentration in the litter was significantly higher in broilers of IEP-supplemented group (T2) than in the low-weight basal-fed group (T1) and in the normal-weight control (T0) ($P < 0.05$). This increment can be attributed to the rise in the ingestion of proteins and excretion of nitrogen in T2 which is as a result of the improvement in digestibility and the growth performance. Moreover, T1 also showed a very high level of ammonia than T0 ($P < 0.05$), which may have been caused by poor nitrogen utilization through gut delayed development in low-weight chicks. Although these differences existed in the concentration of ammonia, no significant differences were found in the footpad dermatitis (FPD) scores between the treatments, indicating that the litter condition did not reach beyond acceptable limits to cause welfare decline, or that the short rearing period did not allow the expression of the FPD symptoms.

The ammonia level in T2 is also high, which means excretion of more nitrogen, most likely due to the consumption of more protein. The unused protein in poultry is released as uric acid that is broken down by microbes to form ammonia (Naseem and King, 2018). Although protein digestibility was lower in T2, there is a possibility that nitrogen retention can be limited due to metabolic requirements, and increased amounts of ammonia can be produced. Interestingly, T1 had even greater ammonia than T0, though they had less intake- perhaps because of poor protein use and retention.

Despite the increased levels of ammonia, there was no significant impact on the footpad dermatitis scores, probably because of good litter management, the short trial period, or good environmental control. Clinical measures of broiler welfare include footpad dermatitis scores, which denote the extent of inflammation and necrotic lesions on the sole of the feet caused by wet and soiled litter, which is frequently associated with long-term contact with the litter. The absence of significant differences means that the threshold conditions of FPD development were not achieved during the trial. But what is interesting is the growing tendency of lesions in T2. Elevated ammonia has been reported to elevate litter moisture and pH, which are important in the pathogenesis of footpad dermatitis (Shepherd and Fairchild, 2010; Hinz *et al*., 2019). This finding indicates that moderate protein supplementation and aggressive litter management is essential in the use of high-protein feedstuffs such as IEP.

Conclusion

The supplementation of IEP in low-weight DOC diets improve growth performance and enhance nutrient digestibility. Although environmental concerns such as increased litter ammonia were noted, these can be managed with appropriate strategies. Therefore, infertile egg powder represents a promising and sustainable alternative protein source for optimizing broiler productivity and promoting circular resource utilization, but it must be processed using low-cost technologies to ensure economic feasibility.

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Conflict of interest

The authors have no conflict of interest to declare.

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