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The Potentials of Two Underutilized Acidic Fruits (*Averrhoa bilimbi* L. and *Phyllanthus acidus* L.) as Phytobiotics for Broiler Chickens

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ABSTRACT

Phytobiotics have attracted considerable interest as the substitutes for subtherapeutic antibiotics in broiler production. Among the phytobiotics, *Averrhoa bilimbi* L. and *Phyllanthus acidus* L. fruits seem to have potential to be developed. Both underutilized acidic fruits contain several bioactive components that may serve as antimicrobial agents, antioxidants, immunomodulators and acidifier, which are beneficial for broiler performance and health. The bioactive components responsible for as well as the mechanisms through which the components exert the phytobiotic activities are highlighted in the present review. The current application of *A. bilimbi* and *P. acidus* fruits in broiler production is also presented. Overall, although *A. bilimbi* and *P. acidus* fruits show phytobiotic properties, they are still less utilized by the farmers to improve the growth and health performance of broiler chickens

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Introduction

The application of antibiotics as an antimicrobial agent and growth promoter is no longer permitted in broiler production worldwide due to the food safety reason. To deal with the increased morbidity and mortality as the consequence of the antibiotic withdrawal from broiler feed, intensive search is now promoted to find the effective alternative to conventional antibiotics. Several antibiotic alternatives have actually been studied, one of which is phytobiotics or phytogenic compounds. The latter compounds are bioactive components that can be extracted from plant sources such as herbs, spice and botanicals (Sugiharto, 2016). *Averrhoa bilimbi* L. fruit is one of the botanicals that is potential to be exploited as the antibiotic alternative for broiler production. *A. bilimbi* belongs to the family of Oxalidaceae and is easily and widely cultivated in Asia. In Indonesia *A. bilimbi* is known as belimbing wuluh and has traditionally been used by people to cure several diseases such as itches, whooping cough, fever, hypertension and inflammation (Dewi *et al.*, 2019). Beyond from its pharmacological properties, *A. bilimbi* is not an important commodity in fresh fruit market. Being considered as an underutilized fruit makes *A. bilimbi* not cultivated on a commercial scale (Peris *et al.*, 2013). *Phyllanthus acidus* L. is another underutilized botanical that has pharmacological effects (Afrin *et al.*, 2016). This plant belongs to the family of Euphorbiaceae and is known as ceremai in Indonesia. In Asia and South America, *P. acidus* has traditionally been used to treat inflammation, rheumatism, respiratory diseases such as bronchitis and asthma, hepatic diseases and diabetes (Tan *et al.*, 2020). A number of studies have recently been conducted to justify the use of *A. bilimbi* and *P. acidus* as folkloric medicines. These investigators documented that both acidic fruits contained several bioactive components that may serve as antimicrobial agents, antioxidants, immunomodulators, etc., which are beneficial for the human health (Andrianto *et al.*, 2017; Fidrianny *et al.*, 2018; Seebaluck-Sandoram *et al.*, 2019). Owing to these latter facts and the need for antibiotic alternative for broilers, *A. bilimbi* and *P. acidus* fruits seem therefore potential to be used as the antibiotic substitute for broilers. The present review aimed to highlight the potentials of *A. bilimbi* and *P. acidus* as phytobiotics for broiler chickens.

Antimicrobial activity of *A. bilimbi* and *P. acidus* fruits

The retraction of antibiotics from broiler feed has been

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confirmed to elevate the prevalence of bacterial diseases in broiler chickens (Sugiharto, 2016; Sugiharto and Ranjitkar, 2019). Some natural antimicrobial agents have been investigated to replace the role of antibiotics in feed, among which botanicals such as *A. bilimbi* and *P. acidus* have frequently been tested for their antimicrobial properties. Both fruits showed antibacterial activities towards broad ranges of Gram-positive and Gram-negative bacteria as listed in Table 1.

Several possible mechanisms by which *A. bilimbi* fruit extract exert the antibacterial activities have been proposed, one of which is the role of flavonoids in the fruit extract (Hamdanah *et al*., 2015). In their study, Xie *et al*. (2015) suggested that flavonoids may inhibit the synthesis of nucleic acid, interfere the function of cytoplasmic membrane, inhibit the cellular energy metabolism, block the attachment and formation of biofilm, impede the porin on the cell membrane, alter the membrane permeability and attenuate the pathogenicity of pathogenic bacteria. In addition to flavonoids, the presences of saponins and triterpenoids seem also to serve important role in inhibiting the growth of pathogenic bacteria (Wahab *et al*., 2009). According to Khan *et al*. (2018a), saponins could damage the bacterial cell wall and thereby deteriorate the metabolic process resulting in bacterial death. With regard to triterpenoids, the lipophilic characteristic of triterpenoids may help to destroy the cell membranes resulting in bacterial death. Also, triterpenoids can react with the porin on the outer walls leading to the bacterial cell wall lysis (Wu *et al*., 2013). The role of oxalic acid in the antibacterial activity of *A. bilimbi* fruit has also been documented. The strong acid of oxalic acid seems to inhibit the bacterial growth as previously revealed by Mokhtar and Aziz (2016). Different from the above mentioned studies, the *A. bilimbi* fruit extract was not active against Gram-positive Enterococcus faecalis in the study of Dharsono *et al*. (2018). Perhaps, the thicker cell walls in Gram-positive bacteria was more difficult to be lysed by the active components of *A. bilimbi* fruit extract. Yet, the latter inference seems not convincible as the growth of some Gram-positive bacteria were able to be inhibited by the *A. bilimbi* fruit extract in the study of Abraham (2016) and Seebaluck-Sandoram *et al*. (2019) as listed in Table 1. It is important to note that antibacterial properties of *A. bilimbi* fruit is greatly influenced by the maturity stage of the fruit. It was apparent in the study of Mokhtar and Aziz (2016) that the extract derived from the young *A. bilimbi* fruit was more potent against pathogenic bacteria when compared with that from the ripe *A. bilimbi* fruit. The latter authors further suggested that the young fruit

Table 1. Examples of studies documenting the antibacterial activities of *A. bilimbi* and *P. acidus* fruits

References	Findings
A. bilimbi fruit	
Dewi *et al*. (2019)	Ethanol extract of *A. bilimbi* fruit had antibacterial activity against *Streptococcus pyogenes*
Seebaluck-Sandoram *et al*. (2019)	Ethanolic extract of *A. bilimbi* L. fruit had wide spectrum of antibacterial activity towards *Escherichia coli*, *Staphylococcus aureus* and *Vibrio parahaemolyticus*
Lisnawati *et al*. (2018)	Aqueous extract of *A. bilimbi* fruit showed antibacterial effect on *S. aureus* and *E. coli*
Abraham (2016)	Chloroform, methanol and petroleum ether extracts of *A. bilimbi* fruit showed antibacterial activity against Gram-positive (*S. aureus* and *Bacillus subtilis*) and Gram-negative bacteria (*Klebsiella pneumoniae* and *Serratia marcescens*)
Mokhtar and Aziz (2016)	Water extract of *A. bilimbi* fruit showed antibacterial activity against *S. aureus, B. cereus, E. coli, Pseudomonas aeruginosa* and *Salmonella* spp.
Hamdanah *et al*. (2015)	Ethanol extract of *A. bilimbi* fruit demonstrated antibacterial activity against *S. aureus* and *E. coli*
Sukandar *et al*. (2014)	Ethanol extract of *A. bilimbi* fruit exhibited antibacterial activity towards *Staphylococcus epidermidis*, *Propionibacterium acnes*, Methicillin-resistant *Staphylococcus aureus* (MRSA) and Methicillin Resistant Coagulase-Negative Staphylococci (MRCNS)
Wahab *et al*. (2009)	Chloroform and methanol extract of *A. bilimbi* fruit showed antibacterial activity against *Aeromonas hydrophila*, *E. coli*, *K. pneumoniae, Saccharomyces cerevisiae*, *S. aureus*, *Streptococcus agalactiae* and *B. subtilis*
Zakaria *et al*. (2007)	Aqueous extract of *A. bilimbi* fruit showed antibacterial activity against Gram-positive *S. aureus*, *S. epidermidis*, *Bacillus cereus, Corynebacterium diphtheriae*, *Kocuria rhizophila* and also Gram-negative *Salmonella typhi, Citrobacter freundii* and *Aeromonas hydrophila*. Chloroform extract of *A. bilimbi* fruit also exhibited antibacterial activity against Gram-positive *S. aureus*, *S. epidermis*, *B. cereus*, *K. rhizophila* and *C. diphteriae* and Gram-negative *S. typhi, C. fuendii*, *A. hydrophila* and *Proteus vulgaris*
P. acidus fruit	
Seebaluck-Sandoram *et al*. (2019)	Ethanolic *P. acidus* fruit extract exhibited wide spectrum of antibacterial activity against *S. aureus* and *V. parahaemolytius*
Foyzun *et al*. (2016)	Crude extract of *P. acidus* fruit exhibited mild and moderate antimicrobial activity against *Shigella dysenteriae*, *S. typhi, Vibro cholera, Pseudomonous aeruginosa*, *S. aureus*, *B. cereus* and *E. coli*
Rahman *et al*. (2011)	Methanol extract of *P. acidus* fruit showed antibacterial activities against *B. megaterium*, *B. subtilis*, *S. typhi* and *S. dysenteriae*
Habib *et al*. (2011a)	Chloroform extract of *P. acidus* fruit showed narrow spectrum of antibacterial activity against *S. dysenteriae*, *E. coli*, *S. aureus* and *Sarcina lutea*
Habib *et al*. (2011b)	Ether extract of *P. acidus* fruit exhibited antimicrobial effect towards *S. typhi*, *S. aureus*, *B. cereus*, *E. coli* and *Bacillus megaterium*
Meléndez and Caprilesa (2006)	Methanolic extract of *P. acidus* fruit could inhibit the growth of *E. coli* and *S. aureus*

contains more bioactive compounds contributing to the antibacterial features of the fruits. Owing to this, the difference in the extends of antibacterial activity of *A. bilimbi* fruit among the studies could be due to the difference in the ripening stage of *A. bilimbi* fruits used in the studies.

In *P. acidus* fruit extract, the presences of glycoside, tannin and resin seem to account for the antibacterial effect of the fruit (Rahman *et al*., 2011). In accordance, Habib *et al*. (2011b) reported that glycoside and steroid are responsible for the antibacterial activities of *P. acidus* fruits. Likewise, terpenoids is another bioactive chemical that is attributed to the antibacterial properties in Phyllanthus fruits (Nisar *et al*., 2018). Tagousop *et al*. (2018) reported that glycoside was able to lyse and damage the cytoplasmic cell membrane resulting in bacterial death. The antibacterial activity of tannins has been detailed reviewed by Akiyama *et al*. (2001). They pointed out that through its astringent property tannins could trigger the complexation with enzymes and substrates resulting in the inhibition of microbial enzyme activity. The toxicity of tannins and the complexation with metal ions may also lead to the bacterial cell death. With regard to resin, Ganewatta *et al*. (2014) suggested that the principal mechanism by which resin act as antibacterial agent is that resin can damage and lyse the membrane cells of bacteria leading to bacterial death. The presences of peroxide and vinyl bonds in the structures of steroids have been attributed to the toxicity and disturbed cell proliferation resulting in reduced bacterial population and death (Doğan *et al*., 2017). From the earlier studies, it was shown that the spectrum of antibacterial activities of *P. acidus* could vary (Habib *et al*., 2011a; Foyzun *et al*., 2016; Seebaluck-Sandoram *et al*., 2019). It was very likely that the differences in the preparation of *P. acidus* extract (such as the ripening stage of fruit and type of solvents used for extraction) as well as the species of bacteria used may affect the antibacterial spectrum of the fruit. Tarafdar *et al*. (2016) revealed that the antibacterial activities of *P. acidus* is solvent dependent with ethanolic extract is the most, and the hexane extract is the least potent antibacterial agents. Taking all the findings together, *A. bilimbi* and *P. acidus* could potentially be developed as the natural antibiotic drugs, which is safe for broilers as well as humans as consumers (Aziz, 2016).

Antioxidative activity of *A. bilimbi* and *P. acidus* fruits

Study has revealed that the excessive use of synthetic antioxidants may induce the carcinogenic effect on the humans as consumers of broiler products (Sugiharto *et al*., 2017). In support to the effort to reduce the use of synthetic antioxidants in broiler production, the search for the alternative natural safe antioxidants are therefore strongly encouraged. Among the natural antioxidant alternatives, botanicals, with regards particularly to *A. bilimbi* and *P. acidus* fruits, seem to be potential as natural antioxidant sources for broiler chickens. Table 2 shows some in vitro studies reporting the antioxidative properties of *A. bilimbi* and *P. acidus* fruits.

In general, the presence of phenolic substances has been attributed to the antioxidant activity of plants and plants-derived products. In their investigation, Suluvoy and Grace (2017) noticed that the content of total phenolic compounds was in parallel with the nitric oxide (NO·) scavenging activity and thus antioxidant activity of *A. bilimbi* L. fruit extract. In accordance with the latter investigators, Asna and Noriham (2014) reported that flavonoids, which is the largest group of phenolic compounds, are also present and responsible for the antioxidant activity of *A. bilimbi* fruit. Besides, the fruit also contains saponins, tannins, terpenoids and steroids that can be accounted as antioxidant properties of *A. bilimbi* fruit (Asna and Noriham, 2014). It is apparent from Table 2 that there are variations in the antioxidant properties and activities of *A. bilimbi* fruit. One possible factor that may be responsible for such divergences is the maturity or ripening stages of the fruits. Based on their study, De Lima *et al*. (2001) documented that the maturity stage affected the physicochemical characteristics and thereby antioxidative properties of *A. bilimbi* fruits. In the form of extract, the solvents used for the extraction or maceration may also influence the antioxidative properties of *A. bilimbi* fruit. Fidrianny *et al*. (2018) showed the variations in the total phenolic content (TPC) among the *A. bilimbi* fruit extracted using different solvents. In their study, the highest TPC content was found in *A. bilimbi* fruit extracted using ethanol as compared with n-hexane and ethyl acetate. The different capability of solvents to dissolve the chemical compounds particularly phenols seem to affect the TPC content of *A. bilimbi* fruit. Other factor that may influence the antioxidant capacity of *A. bilimbi* fruit is the drying process, at which Chauhan and Kapfo (2013) reported that sun-drying reduced the TPC and antioxidant activity of *A. bilimbi* fruit. The *P. acidus* fruit has been reported to contain several antioxidative properties, including flavonoids, tannins, terpenoids and saponins (Ruma 2016; Pradeep *et al*., 2018). In addition to the phenolic compounds, ascorbic acid has also been revealed to contribute to the antioxidant activity of *P. acidus* fruit (Loan *et al*., 2017; Pradeep *et al*., 2018). In agreement to *A. bilimbi* fruit, the antioxidant activity of *P. acidus* fruit extract is also influenced by the solvents used to prepare the fruit extract. Recently, Nisar *et al*. (2018) noticed that water extract of *P. acidus* fruit showed greater antioxidant activity as compared to ethanolic extract of *P. acidus* fruit. They further suggested that water extract may result in more dissolved antioxidant compounds when compared with other solvents.

In concomitant with the in vitro studies, result from the in vivo studies also exhibited the antioxidant activity of *A. bilimbi* fruit. For instance, Thamizhselvam *et al*. (2015) documented that *A. bilimbi* fruit juice enhanced the antioxidant activity both in blood and tissues of wistar albino rats intoxicated or challenged with paracetamol. Moreover, Suluvoy *et al*. (2017) reported that *A. bilimbi* fruit extract reduced the levels of NO and increased the activity of superoxide dismutase (SOD) enzyme and glutathione (GSH) level in the colon tissue of winstar rat suffered from ulcerative colitis. In term of *P. acidus*, Jain *et al*. (2011) documented that the ethanolic extract of *P. acidus* fruit increased the level of GSH, and activites of SOD, catalase (CAT) and glutathione peroxidase (GPx) enzymes in rats and mice. The fruit extract was also able to alleviate the oxidative damage in rats and mice induced by carbon tetrachloride. In agreement with this, Chakraborty *et al*. (2012) revealed that the *P. acidus* fruit extract downregulated NO release and thereby the excessive release of NO and the oxidative damage could be ameliorated. Considering its antioxidative activity, *P. acidus* fruit extract may be employed as therapeutic manner for oxidative stress-mediated diseases such as inflammation and cardiovascular diseases (Nisar *et al*., 2018).

Immunomodulatory activity of *A. bilimbi* and *P. acidus* fruits

Immune system is a crucial apparatus of the body to fight against infectious disease agents. In addition to vaccination program, the outbreak or infectious diseases in broiler farms were conventionally controlled by the use of antibiotics. In the post-antibiotic era, the use of natural immunomodulators is however promoted to protect the chicks from the infectious diseases. For human application, several natural immunomodulatory agents have been investigated, among which are the fruits of *A. bilimbi* and *P. acidus*. Using Wistar rats as human

Table 2. Examples of studies documenting the antioxidative properties of *A. bilimbi* and *P. acidus* fruits

References	Findings
A. bilimbi fruit	
Utami *et al*. (2019)	Freeze-dried *A. bilimbi* fruit contained total phenolic content (TPC) of 6.35 µg gallic acid equivalents (GAE)/mg, total flavonoid content (TFC) of 0.29 µg quercetin equivalents (QE)/mg and IC_{50} 2,2,1-diphenyl-1-picrylhydrazyl (DPPH) scavenging activity and 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid ($ABTS^+$)-reducing activity of 279.99 and 631.78 µg/mL, respectively
Chauhan *et al*. (2018)	Methanol *A. bilimbi* fruit extract showed $ABTS^+$ radical scavenging activity of 11.5 µM and IC_{50} value of 69.42 µg/mL
Fidrianny *et al*. (2018)	Ethanol extract of *A. bilimbi* fruit had IC_{50} DPPH of 29.26 µg/mL
Suluvoy and Grace (2017)	*A. bilimbi* L. fruit extract had total phenolic compounds at a concentration of 209.25 GAE mg/g and nitric oxide (NO) scavenging activity with IC_{50} value of 108.10
Asna and Noriham (2014)	Water extract of *A. bilimbi* fruit contained TPC of 41.00 ± 2.75 mg GAE/g, TFC of 23.32 ± 3.50 mg QE/g
Rahman *et al*. (2014)	Ethanol extract of *A. bilimbi* fruit had TPC of 38.79 ± 0.95 mg/g GAE, TFC of 1.67 ± 0.87 mg QE/g and DPPH radical scavenging activity (IC_{50}) of 635.07 ± 8.41µg/mL
Yan *et al*. (2013)	Ethanol extract of *A. bilimbi* fruit exhibited TPC of 629.17 ± 14.38 mg GAE/100 g of dry weight, TFC of 153.38 ± 8.02 mg rutin/100 g of dry weight, antioxidant activity of 28.41 ± 5.31% and DPPH radical scavenging activity (IC_{50}) of 6.93 ± 0.25 mg/mL
Chowdhury *et al*. (2012)	Hydromethanolic extract of *A. bilimbi* fruit showed strong DPPH radical scavenging activity with IC_{50} value of 20.35 µg/mL, TPC of 417.09 ± 6.58 mg/g in ascorbic acid equivalent (AAE). The extract also contained TPC of 106.16 ± 2.82 mg/g in GAE and TFC of 276.73 ± 25.25 mg/g in QE
P. acidus fruit	
Mallawaarachchi *et al*. (2019)	Water extract of *P. acidus* fruit contained TPC of 112.89 mg GAE/100 g fruit in fresh weight, total monomeric anthocyanin content (TMAC) of 10.41 mg C3G/100 g fruit in fresh weight and vitamin C of 17.12 mg ascorbic acid (AA)/100 g fruit in fresh weight. The extract also had DPPH IC_{50} of 5.60 mg of lyophilized sample/mL or 124.14 mg of fruit in fresh weight/mL
Pradeep *et al*. (2018)	At the concentration of 500 µg/mL methanol extract of *P. acidus* fruit showed the inhibition activity of 92.20 ± 0.72% toward DPPH and 96.98 ± 0.40% toward ABTS
Andrianto *et al*. (2017)	Water extract of *P. acidus* fruit showed high antioxidant activity with IC_{50} 26.06 µg/mL
Foyzun *et al*. (2016)	Crude extract of *P. acidus* fruit showed IC_{50} value of 5.96 µg/mL
Moniruzzaman *et al*. (2015)	Methanolic extract of *P. acidus* fruit contained TPC and TFC of 116.98 mg of GAE and 168.24 mg of QE per gm of dried extract. The fruit extract had DPPH IC_{50} of 15.62 µg/mL
Chakraborty *et al*. (2012)	Methanol extract of *P. acidus* fruit had TPC and TFC of 73.08 ± 0.68 mg GAE/g and 61.28 ± 0.06 mg QE/g, respectively
Jain *et al*. (2011)	Ethanol extract of *P. acidus* fruit showed DPPH radical scavenging activity of 87% at the concentration of 155 µg/mL and IC_{50} of 68.5 µg/mL
Habib *et al*. (2011a)	Chloroform extract of *P. acidus* fruit showed moderate DPPH scavenging activity (no specific value was mentioned)
Habib *et al*. (2011b)	Ether extract of *P. acidus* fruit had mild DPPH scavenging activity (no specific value was mentioned)
Rahman *et al*. (2011)	Methanol extract of *P. acidus* fruit contained moderate antioxidant potential (no specific value was mentioned)

model, Suluvoy *et al.* (2017) reported that *A. bilimbi* fruit extract could attenuate the excessive increase in inflammation-related cytokines such as interleukin (IL)-1β, IL-6, tumour necrosis factor (TNF)-α as well as inflammatory markers such inducible nitric oxide synthase (iNOS) and cyclooxygenase (COX)-2 enzymes. In term of *P. acidus*, Hossen *et al.* (2015) also reported that methanol extract of *P. acidus* fruit attenuated the expressions of iNOS and COX-2 and inhibited the nuclear factor-kappaB (NF-κB) expression. The fruit extract also down-regulated the upstream signalling events of NF-κB translocation, phosphorylation of Src and Syk, and formation of Src/Syk signalling complexes. Previously, Chakraborty *et al.* (2012) also performed the in vitro and ex vivo experiment and found that *P. acidus* was able to regulate the immune system through inhibition of superoxide anion scavenging activity.

There are several bioactive compounds that are responsible for the immunomodulatory actions of *A. bilimbi* and *P. acidus* fruits. Saponins in the *A. bilimbi* fruit is known capable of reinforcing the functions of immune system through the enhancement of lysozyme activity (Khan *et al.*, 2018a). Also, terpenoids in *A. bilimbi* fruit has been associated with the immune modulation as Khan *et al.* (2018b) suggested that terpenoids could modulate the immune responses and impede the delayed type of hypersensitivity reaction. A very recent study by Jantan *et al.* (2019) suggested that adenosine (purine nucleoside) in *P. acidus* fruit is also able to modulate the functions of innate immune response particularly mononuclear phagocyte through 4 G-protein-coupled cell membrane receptors. Other compound in *P. acidus* fruit called kaemferol has also been known to have immunomodulatory effect. The latter component could inhibit the iNOS mRNA expression and thus essential for controlling the excessive inflammation (Jantan *et al.*, 2019). For both fruits, ascorbic acids content also contributes to the immune modulating properties of the fruits. Detailed explanation in the role of ascorbic acids on the immune functions is reviewed elsewhere by Carr and Maggini (2017).

Acidification activity of *A. bilimbi* and *P. acidus* fruits

A healthy intestine (particularly small intestine) has been associated with the maximal production performance and wellbeing of broiler chickens. A number of factors may contribute to the healthy small intestine, among which the pH seems to affect the microbial ecology and morphology of the small intestine (Sugiharto and Ranjitkar, 2019). In the normal condition, the pH of small intestine of broiler chickens is about 6.43 (Mabelebele *et al*., 2014). Any changes in the pH values may adversely affect the intestinal functions and thereby health and productivity of chickens. Modern broiler chickens are usually raised under the stress condition (e.g., high stocking density, heat stress, etc.), which implies in the altered gut environment (Sugiharto *et al*., 2017). Organic acid-based acidifier has long been used to maintain the normal environment and functions of the intestine of broiler chickens (Açıkgöz *et al*., 2011; Sugiharto *et al*., 2019; Widiastuti *et al*., 2019). Due to food safety reason, recently there has been a growing interest to use natural-based product (additives and supplements) in broiler production, and that natural organic acids-based acidifier is now gaining more attention from poultry nutritionists. Underused acidic fruits such as *A. bilimbi* and *P. acidus* seem to be potential as the natural acidifier for broilers due to their acidic characteristics or low pH values. De Lima *et al*. (2001) reported that *A. bilimbi* had pH values of 0.9 to 1.5, and therefore could be exploited as natural alternative for acidifier for broilers. Moreover, *A. bilimbi* contains natural organic acids including acetic acid, citric acid and oxalic acid, with citric acid being the major organic acid (Renatami *et al*., 2018). Indeed, citric acid has been reported to capable of improving the growth performance, nutrient retention and health of broiler chickens (Chowdhury *et al*., 2009; Fazayeli-Rad *et al*., 2014). However, the high content of oxalic acid in the fruit (De Lima *et al*., 2001) should be treated with caution since at a high level this anti-nutritive factor could compromise the nutrient utilization by the chickens. Fermenting of *A. bilimbi* fruit could reduce the level of oxalic acid in the stuffs (Sugiharto and Ranjitkar, 2019), and thereby improve the efficacy of *A. bilimbi* fruit as natural acidifier for broilers. In line with *A. bilimbi*, *P. acidus* fruit also has low pH value (about 3.4) and contains several natural organic acids including tartaric, malic, ascorbic and citric acids (Nambiar *et al*., 2016). In term of poultry production, the application of citric (Chowdhury *et al*., 2009; Fazayeli-Rad *et al*., 2014) and ascorbic acids (Khan *et al*., 2012) have broadly known to improve the growth and health performance of broiler chickens. In agreement with *A. bilimbi*, *P. acidus* fruit however contains some antinutritional factors that may compromise the digestibility of nutrients. In their study, Rout and Basak (2015) reported that *P. acidus* fruit contained oxalate of 0.95 mg/g dry weight, phytate 9.48 mg/g dry weight, tannin 0.053 TAE g/g dry weight and saponins 0.05 g/g dry weight. With regard to phytate, the high level of this component in the fruit should be looked at with attention as it can compromise the nutrient utilization by the chickens. Again, fermentation seems to aid in alleviating the phytate content of *P. acidus* fruit as suggested by Sugiharto and Ranjitkar (2019).

Current application of *A. bilimbi* and *P. acidus* fruits in broiler production

The use of phytobiotics as the alternative to subtherapeutic antibiotics in broiler production is now gaining much more interest. Although still limited, research on *A. bilimbi* has been carried out to confirm the efficacy of this acidic fruit in improving the growth performance and health of broilers. For example, Escala *et al*. (2015) used the extract of *A. bilimbi* fruit as supplement through drinking water, and found that the fruit extract was able to improve the weight gain and feed intake of broilers. In laying hens, Wijayanti *et al*. (2019) reported that combination of *A. bilimbi* powder and probiotics (*Lactobacillus* spp. and *Bacillus* spp.) reduced pH values, decreased total pathogenic bacteria (*E. coli and Salmonella* spp*.) and in*creased Lactobacillus in the small intestine. Likewise, Wiradimadja *et al*. (2015) noticed that the administration of water extract of *A. bilimbi* fruit (at the levels of 0.5, 1.0 and 1.5% from diets) increased cumulative feed consumption and final body weight of the Indonesian native chickens. Although the detailed parameters were not measured by the above investigators, it seems that *A. bilimbi* fruit could improve the intestinal structure and ecology and eventually increase nutrient utilization by the chickens. Considering its highly acidic characteristic (De Lima *et al*., 2001), the application of *A. bilimbi* fruit particularly in the form of juice or filtrate should be with caution as the excessive acid treatment may cause over-acidification of the intestine of broiler. The latter condition may adversely affect the intestinal morphology and ecology and thus compromise the nutrient digestibility and utilization (Nourmohammadi and Khosravinia, 2015). With regard to *P. acidus* fruit, the application of this fruit in broiler production or poultry production in general is scarce. No published study is found in the literature so far. Overall, considering the phytobiotic properties *A. bilimbi P. acidus* fruits, research is therefore needed to confirm the potentials of both acidic fruits in replacing the role of antibiotics in broiler production.

Conclusion

Literature study shows that *A. bilimbi P. acidus* fruits exhibit phytobiotic properties including antibacterial, antioxidative, immunomodulatory and acidification activities. However, both acidic fruits are less utilized by the farmers to improve the growth and health performance of broiler chickens.

Conflict of interest

Author declares no conflict of interests exist.

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