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Application of fermentation technology in poultry feed for the sustainable poultry industry: A review

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Abstract
The poultry industry plays a pivotal role in global food security, providing a significant source of protein for human consumption. However, the industry faces substantial challenges related to sustainability, including the efficient utilization of resources, environmental concerns, and animal welfare issues. In recent years, fermentation technology has emerged as a promising approach to address these challenges and promote sustainable practices within the poultry sector. Fermentation is a versatile process involving the enzymatic transformation of complex organic substances into simpler compounds through the action of microorganisms like bacteria, yeast, or molds. This review explores the application of fermentation technology in poultry feed production and its implications for the sustainability of the poultry industry. The application of fermented feeds in poultry production has gained significant attention due to its potential to replace antibiotics and reduce feed costs. Fermentation enhances the nutritional value and nutrient availability of feed ingredients, leading to improved poultry performance. It also positively influences gut microbiota, intestinal morphology, lipid profiles, antioxidant status, and immunity in poultry. In conclusion, fermentation technology holds great promise for the sustainable poultry industry by improving feed quality, reducing environmental impacts, and promoting animal health. The integration of fermented feeds into poultry nutrition practices represents a critical step toward achieving a more sustainable and efficient poultry production system.

Keywords: Fermentation technology, poultry feeds, solid-state fermentation, submerged fermentation, immunity

Introduction
Fermentation
Fermentation is defined as a chemical transformation of complex organic substances into simple compounds by the action of enzymes which are produced by microorganisms such as bacteria, yeast, or molds. Enzymes facilitate this process by hydrolysis and breaking down complex organic substances into smaller, more easily digestible compounds and nutrients, particularly in case of food.

According to reports from Canibe and Jensen (2012) [4], fermentation outcomes are highly variable and seem to be influenced by the nature and characteristics of the substrate.

Various factors can influences the fermentation rate and fermented product quality. These factors encompass conditions like temperature, pH, levels of dissolved CO2 and CO2, composition of medium and the type of operational system used (continuous or batch type), the addition of different precursors, mixing processes, fermenter shear rates and the duration of the fermentation process (Renge *et al*., 2012) [36].

Depending on the microorganism used for the fermentation, yield of various end products such as acetic acid, lactic acid, or ethanol. Different microorganisms exhibit distinct reactions to each substrate. According to Couto and Sanroman (2006) [6], the Lactobacillus produces lactic acid, molds yields citric acid, while yeasts produce ethanol and CO_2.

History of fermentation

Fermentation is a natural phenomenon that predates our full understanding of the biochemical processes involved. Our ancestors harnessed this natural process to create a variety of products such as beer, wine, mead (fermented honey & water based drink), cheese. Back in the 1850-1860s, Louis Pasteur was the pioneering zymurgist, delving into the study of fermentation and demonstrating that it was caused by living cells. Around 1837 to 1838, C. Cagniard de la Tour, T. Swann, and F. Kuetzing published works that provided compelling evidence supporting the living nature of yeast. Through their independent microscopic investigations, they confirmed that yeast was a living organism capable of reproducing through budding. Interestingly, the term "yeast" traces its origins back to a Sanskrit word meaning "boiling," likely because bread and wine, two staple foods of Europe, were produced using yeast. As research progressed, scientists discovered the presence of bacteria in the fermentation process as well.

Types of fermentation

In general, fermentation techniques are divided into 2 types based on type of substrate used: solid-state fermentation (SSF) and submerged fermentation (SmF).

Solid state fermentation

Solid-state fermentation is a specific process in which microorganisms thrive on a solid substrate under the controlled conditions, typically in absence of free water. Over time, the advancement of these techniques has enabled large-scale production of a diverse range of bioactive compounds, including antibiotics, pigments, antioxidants, antitumor agents, bio-surfactants, bioactive peptides, and more. Generally, SSF is utilized for production of fermented dry feed, which can be incorporated into basic feed mixes like whole grains. Alternatively, the fermented dry feed can be transformed into a powder form.

Moisture essential for the process is absorbed within the solid matrix, as outlined by Pandey (2003) [21]. Nevertheless, it is crucial for the substrate to maintain sufficient moisture to facilitate growth and metabolic activity of micro-organisms.

Selection of Micro-organism

Microorganism selection is a crucial factor in enhancing product yields. The SSF approach, characterized by its low moisture content, is applicable to a restricted range of microorganisms, primarily fungi like Rhizopus spp. and Aspergillus spp. and some bacteria, such as Bacillus spp. and Lactobacillus spp. also employed in this method (Supriyati *et al*., 2015) [27].

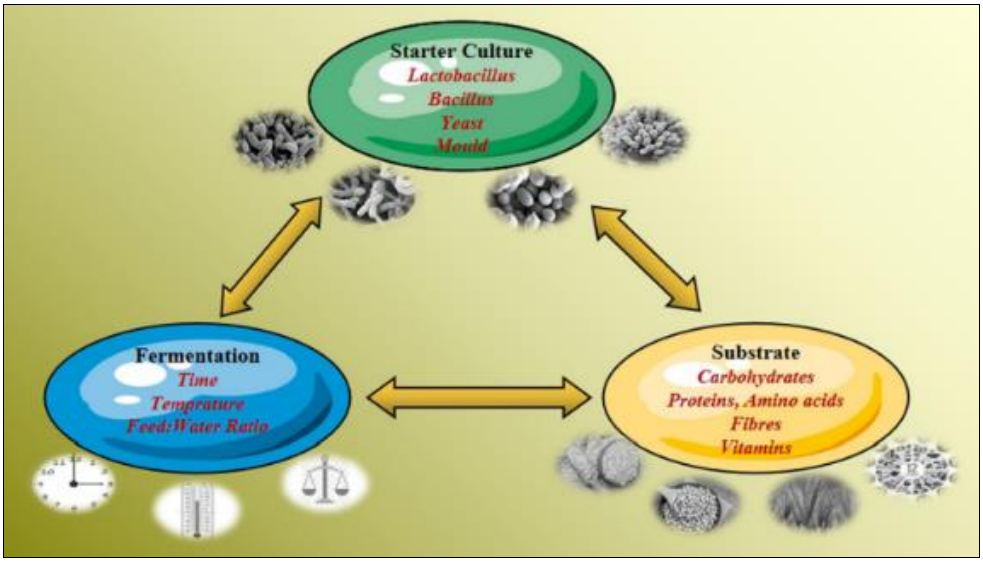

Fig 1: Interactions in fermented feed among micro-organisms present, fermentation parameters, and substrate quantity that influence final end products

Substrate

The choice of substrate plays an important role in micro organism growth and, consequently, increase the product yield. The selected substrate should ideally provide both physical support and essential nutrients to support the thriving culture.

In the SSF process, the selection of the substrate is crucial as it serves as both a physical support and a nutrient source for the growing culture. Commonly employed agro residues as substrates in SSF includes cassava & sugarcane bagasse, various cereal brans like wheat bran, rice bran, and oat bran, as well as soybean bran (Farinas, 2015) [9]. Additionally, coffee pulp and their husks, fruit peels & pulps, wood shavings, corn cobs, and straws from different sources are also used. These materials are mainly composed of NSP like celluloses, hemicelluloses, lignin, starch, pectins, and other fibers.

Depending on the composition of the substrate, the microorganism has to be selected. Lignocellulosic residues are best suited for the activity of wood-rotting fungi. Specifically, white rot fungi, which belong to the Basidiomycetes and some Ascomycetes groups, are adept at degrading lignin. On the other hand, brown rot fungi (Basidiomycetes) excel in breaking down celluloses and hemicelluloses. The soft-rot fungi, represented by species like *Aspergillus niger* and *Trichoderma reesei*, possess a comprehensive system of cellulases that they release into their environment.

Agro residues play a dual role in the process, serving not only as solid support for biomass growth and nutrient absorption, but also as a valuable source of nutrients and carbon for the organisms. In some cases, additional supplementation becomes necessary to ensure optimal growth. The medium is typically enriched with both macro and micronutrients, including elements like P, S, K, Mg, Ca, Zn, Mn, Cu, Fe, Co, and Iodine (Farinas, 2015) [9]. When selecting the substrate, availability and cost are the primary factors to be taken into consideration.

Bioreactor design for SSF

The SSF bioreactors can be categorized according to the type of mixing system utilized. There are several classifications based on the aeration method, which includes static bioreactors (e.g., fixed bed & perforated trays) and stirred bioreactors (such as stirred drum or horizontal drum or). Additionally, other categorizations are available depending on the presence of forced aeration.

Static bioreactors

A variety of static bioreactors are utilized in solid-state fermentation across different scales, ranging from laboratory setups to industrial applications. One common example of a static bioreactor used in SSF is the Erlenmeyer flask.

Erlenmeyer flasks are small perforated trays with fixed type of bed bioreactors (Also known as Raimbault columns), Roux bottles, jars, Petri dishes, and roller bottles are chosen for their simplicity and suitability for working with small volumes. One common feature of these bioreactors is the lack of agitation. Erlenmeyer flasks, particularly well-suited for laboratory-scale investigations and process optimization, are simple glass flasks with a restricted capacity, sealed with cotton plugs to allow aeration through the process of diffusion. Their advantages include ease of handling, cost-effectiveness, and the ability to conduct multiple simultaneous tests.

Agitated bioreactors

An alternative approach to bioreactors for SSF involves continuous and intermittent agitation of the solid medium. This method led to the development of bioreactors, such as rotating drums or horizontal paddle mixers, which may or may not include water jacket. If the continuous mixing is employed, these bioreactors are designed to achieve greater uniformity of solid medium and enhances O_2 transfer to the microorganisms.

Steps involved in SSF (Yang *et al*., 2021) [32]

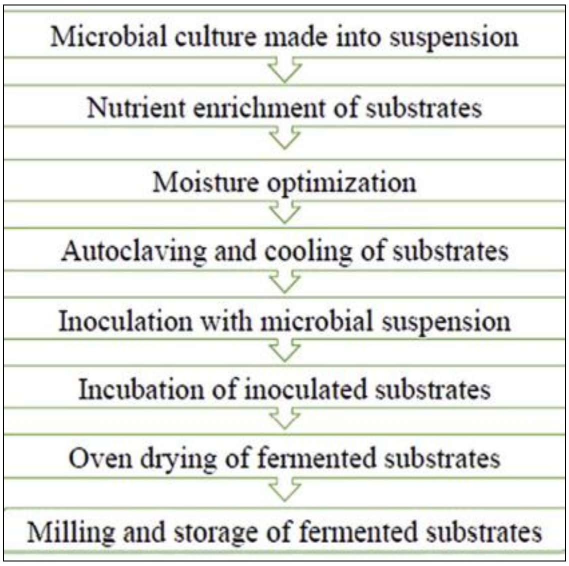

Fig 2: Schematic representation of steps involved in solid-state fermentation of substrates

The SSF Advantages

- Simple and cost-effective, possible to be carried out on farm
- Enhanced efficiency and cost-effective media
- Reduced efforts in downstream processing.
- Resembles the natural environment suitable for various microorganisms, particularly fungi and molds.
- Due to Low moisture content, less susceptibility to contamination, and ease of transportation of the fermentation products
- Possesses numerous environmental benefits.
- Less effluent release, generation of less wastewater and reduce pollution
- By enabling the utilization of solid agro-industrial byproducts as substrates and facilitating solid waste management (Pandey, 2003) [21], it offers the advantage of sustainability and environmental friendliness.
- Greater product stability & yield
- Substrates are utilized in a gradual and consistent manner, allowing for an extended fermentation period using the same substrate.

Disadvantages of SSF

- Difficulties with scale-up
- Difficulties in regulating of process parameters, including temperature, moisture, nutrient content, and others.
- Difficulty in the purification of end products and biomass estimation
- Microorganisms capable of thriving in low moisture conditions can be employed.
- Heat production poses challenges, and regulating the growth environment becomes exceedingly difficult.

Submerged fermentation

Submerged fermentation is a cultivation technique involving microorganisms grown in a liquid broth. This process breaks down the nutrients and releasing of desired bioactive compounds into the solution. In this approach, the fermentation substrate stays in a liquid form, offering essential nutrients for microbial proliferation. It makes use of free-flowing liquid substrates such as molasses, broths, soluble sugars, fruit and vegetable juices, whey, liquid media, wet distillers' grains, and sewage/wastewater (Missotten *et al*., 2016) [17].

In this approach, specific microorganisms are cultivated within enclosed containers filled with a nutrient-rich broth and a high oxygen concentration.

In Submerged Fermentation (SmF), the substrate is consumed rapidly, necessitating constant replacement or supplementation with nutrients. The fermentor, housing the substrate, operates continuously, and the product biomass is harvested continuously using various technical methods an finally the product is filtered or centrifuged and then dried. The most suitable bacteria for submerged fermentation are those that thrive in conditions with high moisture content or high water activity. Submerged fermentation (SmF) is employed to produce fermented liquid feed using these dried bacteria.

Advantages of SmF

- Improved monitoring of temperature, dissolved oxygen, pH and concentration of water-soluble molecules
- Separation of biomass after the fermentation process was simple
- Smooth mixing and aeration

- Straightforward scaling-up process

Phases during microbial growth

A) Lag phase

Following inoculation, there is an initial period with no significant increase in the number of microbial cells, and this phase is referred to as the "lag phase."

During this stage, the organisms acclimate to the new environment into which they have been introduced.

B) Acceleration phase

The acceleration phase refers to the period when the cells have just begun to multiply and their numbers start to increase.

C) Log phase

The log phase, also known as the exponential phase, is characterized by a steady and rapid increase in cell numbers.

D) Deceleration phase

The deceleration phase is the period when the previously constant growth rate starts to decline.

E) Stationary phase

During the stationary stage, the population of microbial cells reaches a state of equilibrium, with no significant growth occurring. This stage is reached either when the carbon source is exhausted or when there is an accumulation of end products.

F) Death phase

In the death phase, cell numbers steadily decline as a result of reduced metabolic activity and depletion of energy resources, leading to cell death.

The choice of maintaining specific phases of cell growth depends on the desired product. In microbial mass production, the logarithmic (log) phase is commonly favoured.

Importance of fermented feeds in poultry

1. Substitute to antibiotics

In the realm of animal production, antibiotics are incorporated into animal feeds through two approaches: utilizing sub-therapeutic levels as growth promoters (AGPs) or administering therapeutic levels to address diseases. As growth promoters, antibiotics assist in reducing the competition between gastro-intestinal microflora and the host for nutrients (Dibner and Richards 2005) [35].

Regrettably, significant apprehension has arisen regarding the utilization of AGPs due to their prolonged and widespread application in animal production, leading to the emergence of resistant bacterial spp. or strains and also due to the accumulation of antibiotic residues in eggs and meat. Many trials are conducted and proved that probiotics, prebiotics, and organic acids are used as antibiotic alternatives. But in recent days, fermented feeds become more popular. Because these fermented feeds are rich in probiotics, prebiotics & organic acids and have similar actions to them (Jazi *et al*., 2019) [13].

2. To decrease the feed cost

In the commercial poultry sector, the cost of broiler feed constitutes a significant portion, up to 70%, of the total expenses. Due to the increasing global feed prices, the poultry industry is progressively investigating alternative/unconventional feed ingredients. Nonetheless, this transition is confronted with several challenges, including fluctuating fiber and protein levels, along with the presence of antinutritional factors (ANF) in these unconventional ingredients. These factors have a negative impact on feed digestibility.

Earlier research studies have demonstrated that fermentation results in an increase in crude protein (CP) content while decreasing crude fiber content (Sugiharto *et al*., 2015a, 2016a) [24-25]. Furthermore, fermentation plays a role in decreasing the occurrence of diverse ANF and toxic compounds in feed ingredients (Xu *et al*., 2012) [30].

Besides the improved nutritional attributes, fermentation is associated with a considerable population of LAB (lactic acid bacteria), leading to the establishment of a low-pH environment enriched with high levels of organic acids (Canibe and Jensen, 2012) [4]. Research indicates that these specific characteristics, either individually or in combination, can act as a protective barrier, guarding the feed against pathogen contamination before being consumed (Niba *et al*., 2009) [18]. Moreover, these features can be advantageous for the gastrointestinal health of chickens.

Solid-state fermentation (SSF) of feed

It is a process where microorganisms degrade feed substrates to facilitate their growth and metabolite production, simultaneously breaking down anti-nutritional factors and toxins present in the feed. This method involves fermenting a feed substrate using naturally occurring or introduced microorganisms under controlled conditions, usually maintaining a water content below 70%.

Adopting SSF of feed has the potential to modify the feed's nutritional characteristics, digestibility, palatability, and safety. Furthermore, SSF shows promise as a viable alternative to antibiotics in animal feed (Wang *et al*., 2017) [29]. The nutritional properties of these fermented feeds are influenced by various factors, such as the type of fermentation starter (the type of bacterial culture), the composition of substrates used, and the specific fermentation conditions like temperature and incubation time (Niba *et al*., 2009) [18].

Fermented feeds characteristics

Fermentation is widely recognized for its positive impact on the nutritional and microbial properties of specific feedstuffs (Sugiharto *et al*., 2015a) [24].

An investigation into fermented maize kernels revealed a rise in organic acids during fermentation, resulting in a decrease in pH from 5.5 to 4.2. Additionally, the counts of lactose-negative enterobacteria, coliform bacteria, yeasts, and molds significantly lowered from 6 to 3 log CFU/g after the fermentation. Conversely, the counts of lactic acid bacteria (LAB) was increased from 7.5 to 8.2 log CFU/g (Ranjitkar *et al*., 2016) [36].

Generally, fermented products exhibit elevated populations of LAB and reduced no of Enterobacteriaceae members. Moreover, they contain higher concentrations of organic acids, particularly lactic acid, resulting in lower pH values compared to their raw material counterparts.

These characteristics make the fermented feeds particularly beneficial for promoting healthy gastro-intestinal functions and also enhancing the overall well-being of chickens (Sugiharto *et al*., 2016a, b) [24-25].

Fermentation offers several benefits, including improved microbiological metrics and the reduction of mycotoxins in feedstuffs. Studies conducted by Okeke *et al*. (2015) [19] demonstrated a decrease in mycotoxins in unsteeped maize grains through fermentation.

Under solid-state fermentation (SSF) conditions, Yang *et al*. (2018) [31] observed decreases in zearalenone levels, which is a mycotoxin primarily produced by Fusarium fungi. The decrease in mycotoxins is linked to the crucial function of bacteria engaged in fermentation, as they assist in breaking down and transforming the mycotoxins into non-toxic compounds.

In addition to the mentioned benefits, fermented feeds contain acetic acids and biogenic amines (such as Putrescine, Cadaverine and histamine), which could affect feed palatability. It is essential to note that fermentation might also compromise certain nutritional elements in the feed, such as degradation of free lysine, which could have adverse effects on host performance (Canibe and Jensen, 2003) [4].

Experts frequently recommend diverse strategies to enhance fermentation processes and optimize the final product. These approaches may include improving fermentation conditions and environments, incorporating acidifiers such as organic acids, utilizing concentrated starter strains like LAB (lactic acid bacteria), or employing enzymes (Canibe and Jensen, 2012) [4]. These measures not only accelerate the fermentation process but also result in improved the nutritional values & improve the palatability of the end product.

Sugiharto *et al*. (2015b) [25] proposed a strategy to preserve essential nutrients in fermented feeds by suggesting the fermentation of the grain fraction separately, before incorporating it into the compound diets, rather than fermenting the complete whole diets. Storage of poultry feed can lead to contamination by bacteria, molds, and fungi, resulting in spoilage and a decline in feed quality, while also increasing the risk of infections in poultry.

Fermented feeds containing elevated levels of lactic acid and maintaining a low pH have been found to impede the growth of bacteria like *E. coli* and Salmonella typhimurium in chicken diets (Niba *et al*., 2009) [18]. Additionally, Londero *et al*. (2014) [37] discovered that the inclusion of fermented whey in the diet of poultry enhanced their resistance to fungal contamination. Moreover, during fermentation, metabolites like organic acids and bacteriocins, produced by lactic acid bacteria, exhibit preservative effects and prolong the shelf-life of fermented feeds, leading to decreased bacterial and fungal contamination.

Effect of fermentation technique on the nutritive value of feed ingredients

The fermentation process enhance the nutritive value of feedstuffs by increasing their CP content. This improvement is attributed due to formation of microbial protein and reduction in non-protein compounds (NPN), such as fiber, during fermentation. Additionally, fermentation leads to a decrease in crude fiber, anti-nutritional factors, and toxic compounds, which is facilitated by the enzymes produced by microorganisms. The decrease in pH value observed in fermented feed can be attributed to the production of organic acids, such as lactic acid and acetic acid, as well as short-chain fatty acids (SCFA) by anaerobic bacteria during fermentation (Sugiharto *et al*., 2015a, 2016a) [24-25].

Table 1: Effect of fermentation on nutritive value of feed ingredients

S. No	Fermentation Substrate	Organisms used	Output	Reference
1	Feed Mixture (containing Maize 60%, SBM 20% & wheat bran)	Bacillus, Lactobacillus S. Cerevisiae @ (2×10^9, 3×10^9 & 5×10^8 CFU/g) respectively for 5 days	Significant decrease in phytic acid, trypsin inhibitors, beta-glucans and pH value Increase in CP value after fermentation.	Liu *et al*. (2021) [16]
2	Cotton seed meal	B. Subtilis BJ (10^6 CFU/g) for 48 hours	Increased the CP, ash & total P content Lowered the crude fiber content	Tang *et al*. (2012) [28]
3	Basal substrate with rapeseed meal (RSM), wheat bran and brown sugar (75%, 24% & 1%) respectively	L. fermentum, B. subtilis Enterococcus faecium, Saccharomyces cerevisiae for 30 days	Reduction in pH and isothiocyanates Increase in C and lactobacilli counts	Chiang *et al*. (2010) [38]
4	Total feed (corn, SBM, corn gluten meal, DDGS, and wheat bran)	LAB 5 days	Improvement in the lactic acid bacterial count and total acid concentration	Li *et al*. (2022) [15]
5	Soybean meal	L. acidophilus, L. Plantarum, B. subtilis and Aspergillus oryzae for 7 days	Improvement in the crude protein, LAB count Lowered the crude fibre and ANF of soybean meal (Phytic acid, trypsin inhibitors, B-conglycin, glycinin)	Jazi *et al*. (2017) [12]
6	Canola meal	Lactobacillus salivarius for 30 days	Increasing CP content, Decreased CF & glucosinolates	Aljubori *et al*. (2017) [1]

Effect of fermented feeds on the growth performance

Fermented feed demonstrates probiotic properties because of its abundant population of lactic acid-producing bacteria, which contributes to acidification. This process lowers the gut pH and creates an environment that hinders the establishment of pathogenic organisms, ultimately leading to enhanced performance (Jazi *et al*., 2017) [12].

Microorganisms play an important role in the breaking down the complex macromolecular organic compounds in the feed into smaller, easily digestible substances. These reduced antinutritional factors lead to a decrease in gut inflammation and an improvement in nutrient bioavailability and digestibility, consequently enhancing growth performance (Feng *et al*., 2007a) [39].

Table 2: Effect of fermented feeds on the growth performance in poultry

S. No	Fermentation Substrate	Inclusion level	Output	Reference
1	Fermented Soybean meal (FSBM)	Total replacement of SBM with FSBM, Broiler	Higher weight gain Better FCR	Jazi *et al*. (2018) [11]
2	Fermented Rapeseed meal	10% FRSM Broiler	Higher weight gain, better FCR than unfermented group	Chiang *et al*. (2009) [5]
3	Fermented Cottonseed meal	10 and 20% FCSM Broiler	Improved growth performance	Jazi *et al*. (2017) [12]
4	Fermented Rapeseed cake	20% of FRSM to layers	Improvement in laying performance, sensory qualities of eggs & albumen quality	Kopacz *et al*. (2021) [14]
5	Fermented canola meal	Broiler	Improvement in body. weight gain, FCR and nutrient digestibility	Elbaz AM (2021) [8]

Effect on gut microbiota & intestinal morphology

The intestinal microflora serves as the initial line of defense, safeguarding the host from diseases that arise due to the colonization of pathogens in the gut. The fermented feed inclusion cause the acidification of the upper GIT. So, this provides appropriate environment for the establishment of beneficial bacteria & prevents the pathogenic bacteria (Niba *et al*., 2009) [18].

Lactobacillus has the ability to secrete lactobacilli and produce organic acids, CO_2, and H_2O_2, which possess the capability to inhibit the growth of pathogenic bacteria (Zhang, 2006) [33]. Furthermore, lactobacillin which is a bactericidal peptide, demonstrates selective entry into the bodies of pathogenic Gram-positive bacteria. Once inside, it acts by either destroying their genetic material or disrupting essential metabolic pathways, effectively inhibiting their growth. H_2O_2 activates the peroxidase-thiocyanate system, causing lactate peroxidase to react with hydrogen peroxide and thiocyanate, leading to the generation of oxidative intermediates. These intermediates play an important role in inhibiting the growth of pathogenic bacteria. Additionally, CO_2 has the capacity to control the growth of certain gram-negative bacteria (Yang *et al*., 2021) [32].

The reduction in pathogenic bacteria can also be attributed to the decreased amount of substrates available for microbial fermentation in the gut. This is due to increased nutrient digestibility in the small intestine which is brought by fermentation of feed (Yang *et al*., 2021) [32].

The fermentation process degrades the antinutritional factors in feed and prevents their negative effects like villus atrophy and crypt hyperplasia in the small intestine. Fermented feed rich in SCFA, butyric acid, and acetic acid act as an energy source for the growth of epithelial cells and improves morphology.

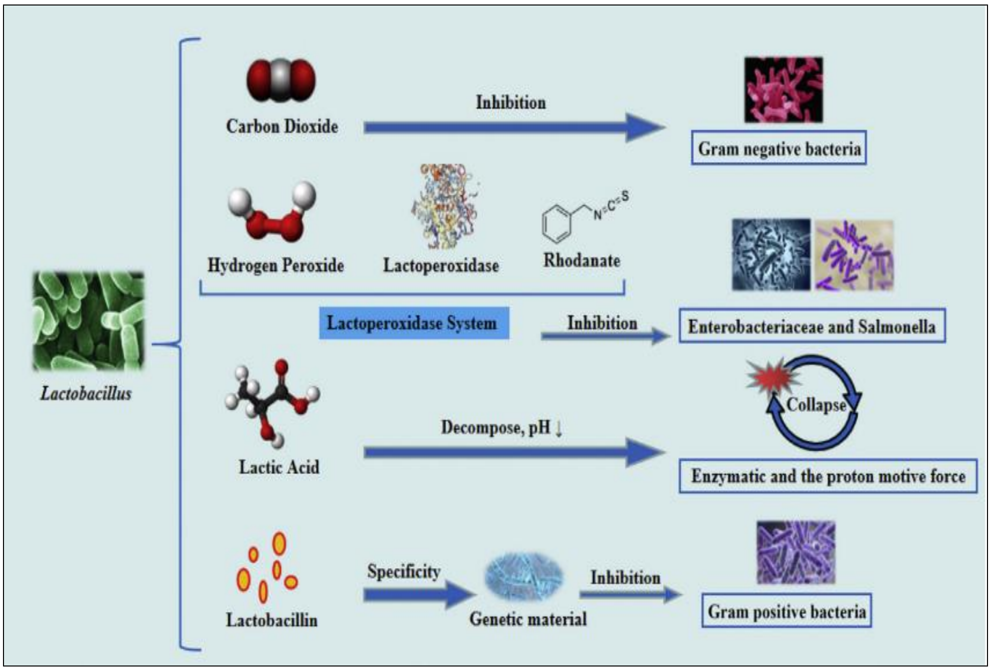

Fig 3: The regulation mechanism of lactobacillus in solid-state fermented feed (SFF) on gastrointestinal ecology.

Table 3: Effect of fermented feeds on gut microbiota & intestinal morphology in poultry

S. No	Fermentation Substrate	Inclusion level	Output	Reference
1	Fermented soybean-based diet (FSBM)	Total replacement of SBM with FSBM, Broiler	Low pH value Increase in LAB count Increased the villus height (VH) and VH:CD ratio and also decreased CD in jejunum	Jazi *et al*. (2018) [11]
2	Fermented cottonseed meal	10 & 20 percent FCSM	Increase in LAB count Decrease coliforms and pH in the ileum Increased VH and VH:CD ratio in duodenum and jejunum Reduced CD	Jazi *et al*. (2017) [12]
3	Fermented canola meal	Broiler	Increased LAB count and *E. coli* count was decreased in intestine Improved intestinal morphology by increasing villus height.	Elbaz AM (2021) [8]
4	Fermented soybean meal based diet	Broiler	Significant improvement in the VH, CD & VH:CD ratio of duodenum and jejunum Increased LAB count, lowered Ileal pH value	Supriya *et al*. (2022) [26]
5	Fermented sesame meal	Broiler	Increase in LAB count and decrease in E.coli and coliforms count in the ileum	Hajimohammadi .*et al*. (2020) [2]

Effect of fermentation on lipid profile

Fermented feeds are abundant in lactic acid bacteria (LAB), specifically Lactobacillus spp., which possess the ability to deconjugate bile salts. This action leads to the hydrolysis of bile salts, disrupting their reabsorption cycle and increasing their excretion in feces. Since cholesterol serves as a precursor for primary bile salts synthesized in liver, the increased excretion of bile salts is linked to higher cholesterol excretion as well. Furthermore, Lactobacilli exhibit hypolipidemic properties by inhibiting the activity of 3-hydroxy-3-methyl-glutaryl-coa, resulting in lower serum concentrations of triglycerides.

In a study carried out by Jazi *et al*. (2018) [11], the researchers investigated and compared the effects of different dietary interventions on serum lipid profiles in quails. The dietary treatments comprised a control diet based on corn and soybean meal (SBM), along with four experimental diets: 1) control diet + 0.1% dietary probiotic mixture (PM); 2) control diet + 0.2% organic acid (OA) mixture; 3) control diet + a combination of both PM and OA; and 4) an additives-free diet with fermented soybean meal (FSBM) replacing the SBM in the control diet. By day 35 of the experiment, the diets containing PM, PM+OA, and FSBM exhibited significant reductions in serum concentrations of triglycerides, cholesterol, LDL-C, and VLDL-C-when compared to both the control and OA-supplemented diets.

Hajimohammadi *et al*. (2020) [2] reported that fermented sesame meal inclusion @ 15% in the broiler diets significantly reduced blood cholesterol & triglycerides levels.

Effect of fermentation on antioxidant status

During the process of fermentation, there is an increase in the production of phenolic compounds, anthocyanins, and flavonoids in the substrates, all of which exhibit antioxidant activity. Moreover, the substrates generate numerous active peptides from protein hydrolysates, which possess the ability to counteract free radicals (Wang *et al*., 2017a) [29]. The fermentation process additionally yields lactic acid, amino acids and antioxidant vitamins, thereby significantly enhancing the overall antioxidant capacity in the fermented feed.

Hu *et al*. (2016) [10] fermented the basal substrate (80% Rape seed meal, 10% wheat bran and 10% corn flour) with B. subtilis, E. faecalis and Candida utilis studied the effect of fermentation on antioxidant status. The 3 dietary treatments are corn-soya-based CD, CD+10.9% RSM and CD + 9.41% FRSM. Significant improvement in total antioxidant capacity and total superoxide dismutase levels in birds fed with fermented rapeseed meal.

Effect of Fermentation on Immunity

As part of the intestinal resident flora, Lactobacillus present in solid-state fermentation (SFF) has the ability to bind to specific receptors on the surfaces of intestinal epithelial cells. This allows them to colonize and function effectively as a protective mucosal barrier. Lactobacillus also fulfills a crucial function by promoting the proliferation of B-cells within the small intestinal lymphoid tissue, thereby enhancing mucosal immune responses. It stimulates plasmocytes to produce a substantial amount of IgA, which contributes to enhanced immune function. Moreover, probiotics and their metabolic products, through solid-state fermentation, activate lymphocytes in the intestinal mucosa, fostering the production of interleukin, tumor necrosis factor, and interferons, thus providing further support to the immune system (Liu *et al*., 2014) [40].

In an experiment conducted by Xu *et al*. (2012) [30], the effect of fermented rapeseed meal supplementation on the antioxidant status of broilers was evaluated. The L. fermentum and B. subtilis cultures were used for fermentation. The dietary treatments included fermented rapeseed meal replacing soybean meal at 0%, 5%, 10%, and 15%, respectively. A significant increase in IgG and IgM levels were observed in the groups which are fed with 10% and 15% fermented rapeseed meal.

Conclusion

Microbial fermentation plays a key role in reducing anti-nutritional factors and enhancing the nutritional value of feed ingredients. The incorporation of fermented products into animal feed brings about several advantages, including improved performance and gastrointestinal health due to their probiotic effects. As a result, they could be considered as an alternative to AGPs (antibiotic growth promoters). Additionally, fermented feedstuffs have positive effects on antioxidant status, immunity, and serum lipid profile.

Furthermore, substituting expensive conventional feedstuffs like yellow corn and soybean meal with more affordable unconventional fermented feedstuffs not only reduces feed costs but also contributes to overall cost efficiency in animal production.

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