

Technical Bulletin

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Lysophospholipids and their role in enhancing Digestion and Absorption

Modern day Poultry rearing has evidenced tremendous change over the past few decades in terms of genetics, nutrition, husbandry/ management and health/disease management. This is quiet evident with the broilers achieving 2.0 Kg in less than 38 days; 330 egg numbers in commercial layers and 165-170 progeny chicks in breeder birds. Of the above factors, nutrition plays a vital role in supporting the desired growth and production performance of birds. Provision of good quality feed with all the essential nutrients must be ensured in these circumstances. Also, the nutrients supplied through feed have to be effectively digested and absorbed to be efficiently utilized.

The digestion and absorption of nutrients is important in poultry because of relatively small GI tract which requires less passage time for the nutrients to travel through the tract. The table 1 would enable a better understanding of the transit time through different segments of GIT.

Table 1

GIT segment of poultry	Transit Time (Min)	Length of GIT (cm) ^a
Crop	50	10.0
Proventriculus/Gizzard	90	9.5
Duodenum	5-8	24.0
Jejunum	20-30	78.0
Ileum	50-70	37.5
Colon	25	7.7

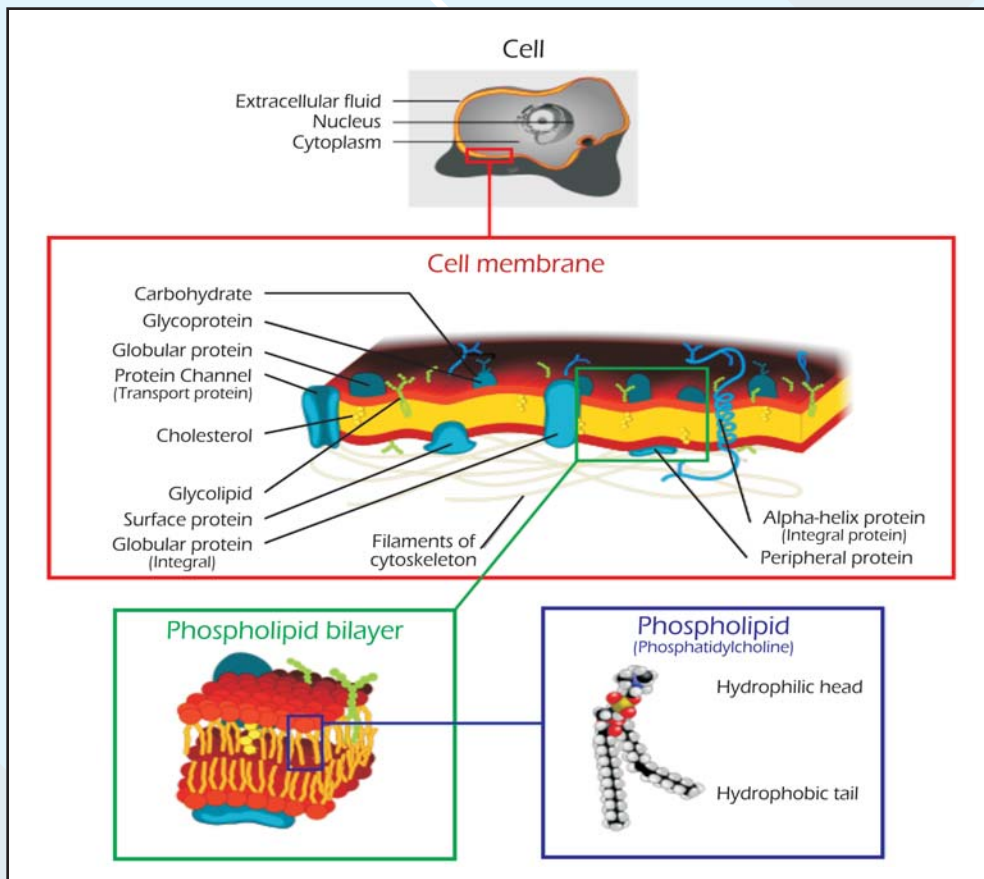
a – Yasar and Forbes (2000)

It takes only 4½ hours for the food to travel down from the mouth to the colon, within this short span, the nutrients have to be broken down and absorbed effectively. Also, the nutrients are primarily absorbed in the small intestine (duodenum, jejunum and ileum) where the feed stays only for a maximum duration of 2 hours. If the nutrients are not absorbed within the above time-limit, they are attacked by the bacteria in the large intestine or excreted as wastes into the environment, defeating the purpose for which they are fed and is reflected in terms of poor growth and productivity.

The above facts call for the need of an external agent that is capable of facilitating the digestion and enhancing the absorption of nutrients.

I. Lipid Bilayer Membrane

All biological membranes are composed of lipid bilayer (40% lipids) along with membrane proteins (60% proteins). The arrangement of hydrophilic heads and hydrophobic tails of the lipid bilayer along with the membrane proteins are held together by non-covalent bonds and by calcium ions, which separates the internal components of the cell from the extracellular milieu (Singer and Nicholson, 1972). The membrane lipids are composed of phospholipids, being the most abundant, glycolipids and cholesterol. The different fractions of phospholipids are phosphotidyl choline (PC), phosphotidyl ethanolamine (PE), phosphotidyl inositol (PI), phosphotidyl serine (PE) and phosphatidic acid (PA). Of these, PC and PE are the predominant phospholipids in poultry (Hermier *et al.*, 1999). The



protein components of the lipid bilayer membrane act as channels, pores or gates that facilitate the flux of digested nutrients across the cell.

II. Mechanisms of Absorption

There are four basic mechanisms of nutrient absorption across the lipid bilayer.

1. **Diffusion** – Hydrophobic and water molecules diffuse across the membrane.
2. **Passive Transport** – Hydrophilic substances cannot pass across the membrane and their transport is mediated through channels/pores/gates. It is also known as facilitated diffusion.
3. **Active Transport** – Certain polar compounds are pumped across the membrane proteins with expense of energy (ATP is used up)
4. **Endocytosis and Exocytosis** – Some molecules too large or too hydrophilic are moved across the cell membrane through fusion or budding of vesicles.

The rate of transfer of molecules across the membrane depends on the membrane characteristics and size and charge of the molecule and the length of the hydrophobic acyl chain (carbon chain).

III. Factors affecting Absorption of Nutrients

1. Age

Younger birds are less efficient than adult birds in absorption of nutrients. This might be due to the developing GIT, unavailability of sufficient amounts of digestive enzymes, unavailability of sufficient receptors or carriers for absorption.

2. Rate of passage of ingesta

The slower the transit time, the better is the digestibility and absorption of nutrients. Factors that increase the viscosity of the digesta and the gastric emptying have a retarding effect on the absorption of nutrients.

3. Diet Composition

The feed supplied to the birds should be free of antinutrients and toxic substances which interfere with nutrient absorption. Feed high in fibre and other antinutritional factors such as phytates trap some of the nutrients affecting their digestion as well as absorption. Presence of pesticides and mycotoxins in feed also hamper the digestibility and absorption of most of the nutrients.

4. Feed Type

The feed type has a significant effect on the

digesta absorbability. Pelleting improves the digestibility and absorption of most of nutrients such as energy yielding nutrients, protein, fat, vitamins and minerals when compared with mash feed.

5. **Ingredient Quality**

Ingredient quality has a direct relationship with the absorption of nutrients. Eg. Underprocessed or overprocessed soybean meal affects the protein and amino acid digestion and absorption.

6. **Gut ecology**

A perfect balance between commensal and harmful bacteria aids in improved absorption of nutrients by maintaining the optimum pH (acidic) of the gut. Any disturbance in the gut microbiota decreases the nutrient absorption. Erratic/overuse of antibiotics or antibiotic growth promoters in feed also disturbs the microbial balance of the gut disturbing the nutrient absorption.

Stress conditions also misbalance the gut ecology.

7. **Disease Conditions**

Diseases that damage the epithelial cells of intestine leading to enteritis such as necrotic enteritis, coccidiosis, malabsorption syndrome, colibacillosis etc reduce the digestion and absorption of nutrients.

8. **Genetics**

Hereditary conditions that lead to an impaired intestinal development or conditions that cause reduced absorption or that cause reduced secretion of hormones such as PTH, corticosterone and thyroxine also lead to impaired absorption.

IV. Ways to Improve Absorption

A multitude of factors such as genetics, nutrition, disease conditions, physiology, feed type etc are involved in the digestion and absorption of nutrients. When health and management of the bird is good, nutrition has an essential role in improving the nutrient digestion and absorption. The nutrient digestion and absorption can be improved by the usage of,

1. Agents that improve the permeability of the biological membranes.
2. Bile salts and lipases

Supplementation of Bile salts and Lipase in poultry diets improve the emulsification and digestibility of fats but do not play any role in their subsequent absorption. Lipases are commonly used in various

exogenous enzyme formulations, whilst bile salt usage is not common due to its economic non-viability.

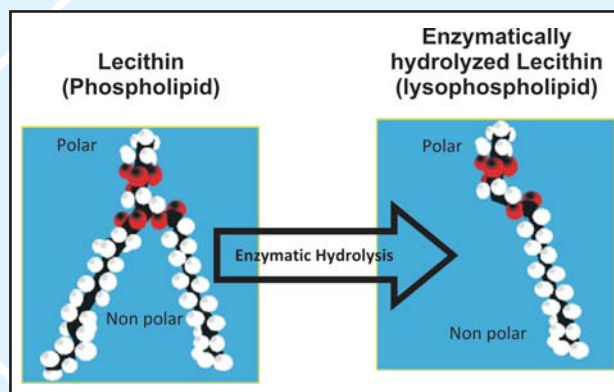
IV. (a) Agents that Improve the Membrane Permeability

Phospholipids (PLs) aid in the emulsification and hydrolysis of fat but have no effect on the absorption of fat and other nutrients (Saunders and Sillery, 1976). PLs when enzymatically hydrolysed help in improving the membrane permeability increasing the flux of various nutrients across the cell.

Enzymatic Modification of Phospholipids

Lecithin serves as the source of phospholipids. The lecithin can be obtained from a plant (soybean, sunflower and rape seeds) or animal source (egg yolk). Generally lecithin is sourced from soybean due to its greater yield, easy availability and feasibility. The lecithin obtained fluctuates greatly in its composition of PLs from source to source. The composition of lecithin for use in animal feeds has not been standardized and hence its utility in high quality end products is questionable. Also, it has its own disadvantages of being hindering the absorption of fat and fat soluble vitamins. (Koo and Noh, 2007)

Enzymatic hydrolysis of lecithin yields lysophospholipids (LPLs) that act as absorption enhancers. Hydrolysis of the PLs at the sn-2 position by phospholipase A2 (microbial origin – non-GMO) yields LPLs. Mere hydrolysis of the PLs would not serve the purpose as absorption enhancers, the enzymatic hydrolysis has to be standardized to yield consistent quantities of PLs and LPLs in the final product. Also, a defined ratio has to be maintained between the different LPLs (lysophosphatidyl choline (LPC), lysophosphatidyl ethanolamine (LPE), lysophosphatidyl inositol (LPI), lysophosphatidyl serine (LPS)) and amongst the PLs and the LPLs to observe consistent and desired end results with the product.



V. Mode of Action of Lysophospholipids

V. (a) As an emulsifier

1. LPLs have superior emulsification properties than PLs due to formation of smaller micelles. Smaller micelles have larger surface area and hence more is the emulsification.
2. LPLs have lower CMC (critical micellar concentration) than bile salts and hence are required in less quantity than bile to emulsify same amount of fat. High CMC enables lipophilic proteins to be included within the micelle. This would lead to the transmembrane proteins being included within the micelle which would reduce the pore size of membrane and reduce the flux of nutrients across the cell. Lower the CMC better the emulsification and further absorption.
3. LPLs reduce interfacial tension between water and oil forming more stable emulsion.
4. In living system, oil-in-water (o/w) emulsifier is required and LPLs serve the purpose as o/w emulsifiers due to low hydrophilic lipophilic balance (HLB). PLs serve as effective emulsifiers in water-in-oil (w/o) emulsions.
5. External supplementation of LPLs augments lysolecithin output of the Liver/ bile duct further improving the emulsification of fat and absorption of fat and other nutrients.

V. (b) As an absorption enhancer

1. LPLs form micelles with smaller radii in the gut

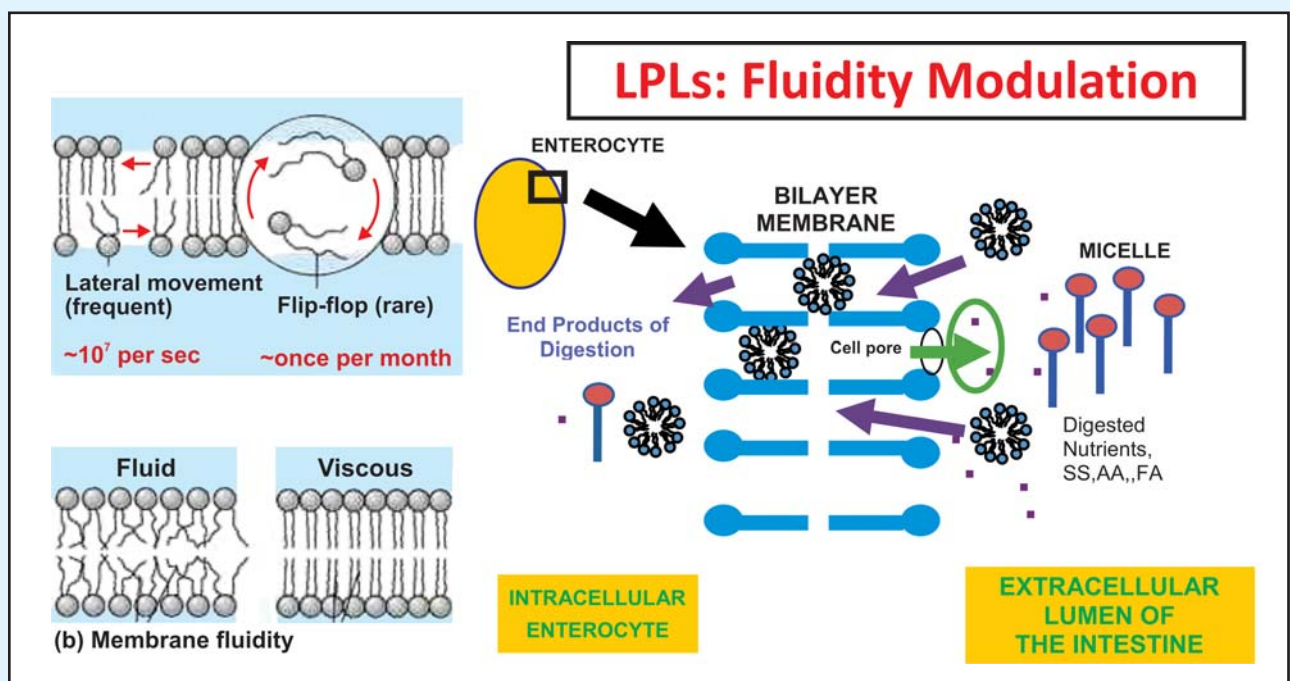
thereby increasing absorption.

2. LPLs have the ability to change the attraction between lipids and displace calcium ions. With this increased freedom of movement lipids can leave the membrane and remaining can aggregate closer together making existing holes larger so that larger molecules are preferentially absorbed.
3. When the membrane comes into contact with certain ratio of lysolipids, these exogenous lysolipids quickly get interdigitated into the bi-layer membrane. The close-packing between the PLs is disrupted ('membrane perturbation') and the lipids go from order to disorder state and the membrane becomes more fluid i.e. the gaps or pores in the membrane form big clusters or larger vacancies in the matrix causing an increase in number and size of the pores.

VI. Role of Different LPLs

VI. (a) In Livestock Sector

1. LPLs alter the protein channel (gramicidin channel) in membrane by altering the membrane deformation energy. The change in deformation energy is related to the molecular shape particularly the size of the head group of LPL. LPC and LPI are markedly more potent than LPE and LPS in increasing the channel activity, thus increasing the pore size of membrane and consequent flux of nutrients (Lundbaek and Andersen, 1994).



- LPLs produce specific and reversible activation of TREK-1 and TRAAK – the K⁺ channels. Opening of TREK-1 and TRAAK by LPLs plays an important role in the regulation of synaptic function and also plays a protective role during ischemia and inflammation. The LPLs modulate the channel function by either interacting with the channel protein or by partitioning into the lipid bilayer. The shape of the LPL rather than the charge plays a role in altering the membrane channels. LPI and LPC (large head groups) are much more potent openers of TREK-1 and TRAAK than LPE and LPS (small head groups). The activation of the channels also depends on the length of the acyl chain. LPC with C14 and C18 are strong activators than C6 and C10 (Maingret *et al.*, 2000).
 - LPC enhances the lymphatic absorption of α -Tocopherol and regulates the absorption of lipids and lipid soluble vitamins. On the other hand PC, inhibits the intestinal absorption of α -tocopherol (Koo and Noh, 2007).
 - LPLs play critical role in absorption of lipids and fat soluble vitamins (Noh and Koo, 2000).
 - Addition of LPLs on top of diet in three different trials has shown to increase the growth performance of broiler birds when compared to control group of birds (Table 2).
 - Experimentation in broilers fed diets reformulated by 2% with LPL (by scaling down all the nutrients by 2%) has shown to produce body weight and FCR better than or equivalent to control diet and also helped in reducing the feed cost per Kg body weight (Table 3).
- The varied trials with LPL on top as well as in reformulated diets suggest that the LPLs enhance the nutrient digestion and absorption by their ability to form comparatively smaller micelles/liposomes and by increasing the flux rate of various digested nutrients across the cell membrane by improving its permeability. This consequently accounts for improved growth performance of broilers. Also, it has been observed that LPL reduces liver fat percentage (Table 4) demonstrating better fat digestion and

Table 2

Effect of LPL supplemented on top of diet on the growth performance of broiler chicken

Parameter	Experiment I (Floor, 40d, Cobb 400 males)		Experiment II (Floor, 35 d, Hybro unsexed)		Experiment II (Cages, 39 d, Hybro unsexed)	
	Control	LPL on top	Control	LPL on top	Control	LPL on top
Body weight, Kg	2.529	2.543	1.698	1.799	1.872	1.937
FCR	1.794	1.775	1.764	1.693	1.802	1.690
Mortality, %	4.46	3.60	8.89	11.11	4.69	3.65
Feed cost/Kg BW, Rs	23.39	23.32	22.69	21.98	23.24	21.97
EPEF	337	345	251	270	254	283

EPEF : European Performance Efficiency Factor (Source: Nutrisys Center for Animal Nutrition)

Table 3

Effect of supplementation of LPL in diets reformulated by 2% on the growth performance of broiler chicken

Parameter	Experiment I (Floor, 40d, Cobb 400 males)		Experiment II (Floor, 35 d, Hybro unsexed)	
	Control	LPL in reformulated diet	Control	LPL in reformulated diet
Body weight, Kg	2.529	2.530	1.698	1.745
FCR	1.794	1.772	1.764	1.760
Mortality, %	4.46	4.46	8.89	9.44
Feed cost/Kg BW, Rs	23.39	22.91	22.69	21.75
EPEF	337	341	251	257

EPEF : European Performance Efficiency Factor (Source: Nutrisys Center for Animal Nutrition)

Table 4

Liver fat percentage of Hybro unsexed broiler chicken fed diet supplemented with LPL

Parameter	Experiment I (Floor, 35d)		Experiment II (Cages, 39d)		
	Control	LPL on top	LPL in reformulated diet	Control	LPL on top
Liver fat, %	12.25	7.42	9.18	11.19	6.53

(Source: Nutrisys Center for Animal Nutrition)

absorption which may lead to lean meat production in broiler birds.

7. Lysolecithins produce monodisperse o/w-microspheres and yield more stable emulsification process. Lecithins, on the other hand, could not produce monodisperse microspheres in water phase (Tong *et al.*, 2001).
8. Revnier *et al.* (1985) opined that for a given phospholipid concentration, cholesterol uptake is greater in the presence of lysolecithin than in the presence of lecithin.
9. LPLs in general are involved in regulating diverse processes such as development and repair of blood vessels (the acyl chain length of C < or equal to 14 leads to endothelial cell motility whereas C16 or C18 inhibit it reducing cell repair) (Murugesan and Fox, 1996).
10. LPC and LPE play a role in T-lymphocyte activation improving both cellular and humoral immunity (Yoshinori *et al.*, 1992).
11. Apart from increasing membrane permeability, LPLs also play a role in promoting the rapid influx of monocytes and activate the macrophages consequently causing phagocytosis of myelin debris (Ousman and David, 1999).

VI. (b) In Human Pharma

1. LPLs activate specific combination of G proteins by attaching to LPL receptor-G2A, enhances neutrophil function, promotes apoptosis (programmed cell death) and also facilitates calcium mobilization (Frasch *et al.*, 2007) and are involved in regulating diverse processes such as angiogenesis, cardiac development, neuronal survival and immunity (Hla *et al.*)

2. LPLs play a role in inflammation mediated immune regulation in human cancer by serving as ligands for CD1d restricted T cells (Chang *et al.*, 2008)
3. Skoura and Hla (2009) reported that LPL signaling is important in cancer, autoimmune and inflammatory diseases, multiple sclerosis and that in future would lead to novel therapeutic tools to safeguard humans against various diseases. LPLs are also indicated in the treatment of atherosclerosis and other inflammatory diseases (Matsumoto *et al.*, 2007).
4. Alkyl LPLs are used as chemotherapeutic agents in human solid tumors (Andreesen a, 1979) and leukemia (Andreesen b, 1979) and visceral leishmaniasis (Azzouz *et al.*, 2007).

Conclusion

LPLs are involved in the absorption of various nutrients by impacting the flux rate at which nutrients of various molecular weights pass across the membranes of the gut. Transport of nutrients as small as calcium ions and as large as polysaccharides is enhanced with LPLs due to modulation of the membrane channel by altering their mechanical properties or by increasing the number and size of pores within the membrane. Thus the absorption of breakdown products such as amino acids, simple sugars, fatty acids, vitamins, minerals and other additives is optimised leading to efficient growth and production performance of birds.

References are available with author and can be made available on request.

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