Mini Review

A review on the role of EPA and DHA through goat nutrition to human health: could they be effective both to animals and humans?

Abstract

Goat’s milk is high digestable and useful for a number of human health problems, but this product could be further increased in its beneficial effects on immune response both at animal and human level through the use of dietary n-3 polyunsaturated fatty acids (PUFAs) sources in goat diet.

In the last decade, the interest on n-3 PUFAs, in particular eicosapentaenoic acid (EPA, C20:5) and docosahexaenoic acid (DHA, C22:6), has increased due to their proved positive effects on human health. These two PUFAs must be provided with the diet both in human and animals. Anyway EPA and DHA can be successfully transferred into milk and thus can be available to human consumption with positive related effects on immune response. But what is the best dietary source of EPA and DHA in goat diet? The most promising and investigated raw material seems to be fish oil, but the interest in alternative n-3 PUFAs-rich sources, such as algae, is increasing. The EPA and DHA mechanisms of action, affecting metabolic and pathological pathways, must be still elucidated in goats, especially for what concern the use of dietary algae. The inclusion of fish oil and algae in the diet of dairy goat can increase the healthy properties of milk with consequential health benefit in human.

Keywords: polyunsaturated fatty acids, fish oil, algae, goat, human, immune response

Abbreviations: UFA, unsaturated fatty acids; PUFA, polyunsaturated fatty acids; NEFA, non esterified fatty acids; EPA, eicosapentaenoic acid; DHA, docosahexaenoic acid; CLA, conjugated linoleic acid; DMI, dry matter intake; NRC, national research council; PGE₂, prostaglandin E₂; TXB₂, thromboxane B₂; LTB₄, leukotriene B₄; LTE₄, leukotriene E₄; MUFAs, monounsaturated fatty acids; ROS, reactive oxygen species; PMN, polymorphonuclear; LPS, lipopolysaccharide; CLA, conjugated linoleic acid

Introduction

High nutritional value of goat’s milk and its higher digestibility than cow’s milk due to the smaller fat globules size are evident since years and represent a good challenge in resolving a number of human health problems such as malabsorption syndrome or high total and LDL cholesterol levels in human blood. Goat’s milk has a low content of polyunsaturated fatty acids (PUFAs; 2-6% of fatty acid composition) and a relatively high amount of saturated fatty acids (SFAs; 53-72%); the remaining part is represented by monounsaturated fatty acids (MUFAs). The rising question is: how can we further improve this unique animal product and contribute to human health beside goat’s milk natural properties?

One of the major chances we have is to increase n-3 PUFAs milk content since there is evidence that these fatty acids have the potential to improve long-term human health. Moreover the western diet is very high in n-6 fatty acids relative to n-3 fatty acids. The ratio n-6/n-3 PUFAs recommended is less than 5 and today this ratio in animal products is between 10 and 15. This is the point, but what kind of n-3 PUFAs do we want to increase? And more: what is the most interesting source of these fatty acids?

Among all n-3 PUFAs, eicosapentaenoic acid (EPA, C20:5) and docosahexaenoic acid (DHA, C22:6), have been shown to be essential in mammals species leading to nutritional and health beneficial actions, often with anti-inflammatory properties.

If we account for human health, DHA leads to higher neuronal development in foetal and early infants, but the endogenous production of this fatty acid by the mother from α-linoleic acid (ALA), as example, is very poor due to the competition with linoleic acid (LA, 18.2 n-6) for the same metabolizing enzymes, particularly at the first Δ6-desaturation step. In human both EPA and DHA must then be introduced with the diet and the consumption of milk with increased EPA and DHA content can be a good chance. Unfortunately the content of these two fatty acids is minimal in traditional ruminant diets and consequently milk has a very low amount of them (<0.1% of total fatty acids) but improving EPA and DHA content in the diet of dairies could lead to an higher available content of these fatty acids in milk for human consumption.

What is the best source of EPA and DHA in animal nutrition? During years a big interest has been pointed out on fish oil that seems the most promising raw material for this purpose due to the fact that it mostly contains EPA and DHA among all the other n-3 PUFAs. Anyway, fish oil is a costly supplementation of lipids compared to other more conventional fatty acid sources and there is increasing concern on the sustainability of the use of fish products in animal diet, which leads to consider alternative sources of long-chain n-3 PUFAs.
One of these alternatives is the use of a source of α-linoleic acid (ALA) as a precursor for EPA and DHA such as linseed, that anyway is affected by two main disadvantages:

i. The low digestibility of linseed due to the fact that the digestive enzymes cannot penetrate the seed coat, usually solved with treatments like extrusion and expansion;

ii. The presence of anti nutritional factors, which however can be destroyed by thermo-extrusion in some extent.11

Beside linseed, algae have high content of PUFA s and can be considered as an alternative to fish oil.20 They have several advantages: easy to cultivate, relatively low cost, high yielding and fast in growth. Moreover these compounds were found to have some interesting bioactive components such as antioxidants, sulfated polysaccharides, phlorotannins, diterpenes, minerals and vitamins, high protein content, valuable nutrients.5,12 Thus, the use of algae as a dietary additive in livestock diets could be of great interest22 especially with the aim of decrease SFA s content through an increased content of conjugated linoleic acid (CLA) and other PUFA s in the milk of ruminants.23

It is recognized that algal lipids have a higher PUFA s content than terrestrial plants24 although some differences in the fatty-acids composition among species and intra-species variations occurs also related to environmental and geographical factors,25,26 season,31 and algal parts.28

The use of algae as PUFA s source is a very new topic and the scientific investigations on algae in dairy goat nutrition are very few, but the interest is continuously increasing thanks to several health benefits found in other species through their prebiotic functions, antimicrobial activities, improved digestibility, antioxidant role and anti-inflammatory and immunomodulatory properties.34

Dietary n-3 PUFA s: milk yield, milk fatty acid composition, EPA and DHA Transfer efficiency

During years, goat milk yield and composition responses, when adding n-3 PUFA s in the diet, often produced contradictory results. Some studies27,28,30 found no effects on milk production when feeding fish oil, but the supplementation significantly reduced milk fat. Differently Kitesa et al.31 evidenced a significant milk yield decrease without changes in milk fat content in response to dietary tuna oil. To the author’s knowledge very few literature is available on the effect of dietary algae on production and quality of goat milk, but generally no differences in milk yield and a significant decrease in milk fat content in cow30 and sheep32,33 was observed.

In this view, one possible mechanism for diet-induced milk fat depression in dairy ruminants involves direct inhibition of lipid synthesis in the mammary gland in response to fatty acid intermediates such as trans-C18:1 and related metabolites, formed during partial biohydrogenation of unsaturated fatty acids in the rumen.33,34 The fatty acid composition of milk can be seen as the result of both the tissue fatty acid biosynthesis and the dietary fatty acid composition of the diet35 but, if in monogastri cans the relationship between dietary fatty acid profile and milk fatty acid content is quite stronger and linear, this is not the same for ruminants because rumen microorganisms are responsible for the biohydrogenation process of dietary fatty acids that leave the rumen and pass in the small intestine, where they can be absorbed.26,36,37

The administration of both fish oil and algae in ruminant are reported to strongly affect the fatty acid profile of milk where the observed results are a decrease in SFAs with increased PUFA s content. More in details, the fatty acid profile of mature goat milk was influenced by unprotected fish oil administration with a lower average concentration of C18:0 and higher content of C16:1, C18:3, and very long-chain n-3 PUFA s EPA and DHA.12

The use of algae in the diet of dairy goat are reported to decrease palmitic acid content in the milk with increased levels of oleic, linoleic and linolenic acid and a positive change in SFA :MUFA :PUFA with a reduced proportion of SFA and a tendency to a positive shift in n-6:n-3PUFA , with increasing proportion of n-3.38 The observed increase in PUFA s milk content15,33 in dairy sheep when feeding algae is relative to an higher content of very-long chain PUFA s and particularly EPA and DHA, according with what previously observed in cow.32

Thus, EPA and DHA content in milk can be significantly enhanced through goat nutrition, but what is the extent to which these fatty acids are transferred from the diet into small ruminants’ milk fat? The question is still debated because it mainly accounts for the source of n-3 PUFA s and if a rumen protection is applied over these compounds.

Beside the different dietary sources, the observed discrepancies in transfer efficiencies have been explained by the dosage applied, the duration of administration, and level/technological methods for rumen protection of the dietary fatty acid sources. With regard to this last point the transfer efficiency of EPA and DHA from diet into milk is however generally low when not rumen protected because of the high amount of rumen biohydrogenation together with the preferential incorporation into plasma phospholipids and cholesterol esters.32

Anyway, the alternative to introduce “inert” fats in the diet rather than the native forms of supplementation to escape or bypass fat rumen biohydrogenation is preferable because a more stable form can preserve fatty acids form oxidation and the inclusion in the diet of even poorly protected fats can reduce the negative effects of high levels of unprotected dietary fats on feed intake, ruminal fermentation, and milk fat content.28

At the moment it seems that the transfer efficiency to mature goat milk ranges from 7.0% to 14.0% for EPA and from 7.0% to 8.1% for DHA when a rumen unprotected form of dietary fish oil is used,19 although some works found lower EPA transfer efficiency ranging from 2.8% to 7.9% 30,31,40,41 and a greater variability for DHA rate of passage from 2.8%40 to 20.3%.40

Considering algae dietary supplementation, DHA transfer efficiency in dairy cow was 8.4% when using an unprotected form of algae, and 16.7% when a rumen-protected form was fed.12 Similarly, the DHA transfer rate into sheep milk from unprotected algae compound was between 5.0%31 and 8.0%, while 30% to 60% of dietary EPA was incorporated.21

Improving immune response with EPA and DHA

Fatty acids metabolism plays a significant role in immune cells functions, both in humans and in animals.42 The most powerful immunomodulatory effect is achieved by the n-3 PUFA s and, among them, by EPA, C20:5(n-3) and DHA C22:6(n-3).

Long chain n-3 PUFA s modulate immune functions in several ways by replacing, for example, arachidonic acid during the

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eicosanoid signalling cascade,\textsuperscript{53} thus decreasing the production of inflammatory eicosanoids such as of PG\textsubscript{2}E\textsubscript{2},\textsuperscript{44} TXB\textsubscript{2},\textsuperscript{45} LTB\textsubscript{4},\textsuperscript{46} 5-hydroxyeicosatetraenoic acid\textsuperscript{47} and LTE\textsubscript{4}.\textsuperscript{48} Long chains n-3 PUFAs can also directly interfere with cytokine gene expression.\textsuperscript{49} Further regulatory pathways include regulation of cell surface expression of adhesion molecules,\textsuperscript{50} membrane fluidity and apoptosis rates.\textsuperscript{51} In addition, both EPA and DHA give rise to family of anti-inflammatory mediators called resolvins.\textsuperscript{52} Most of these activities directly target leukocyte function.\textsuperscript{53}

Monocytes are recognized as major effector cells of the immune system, playing a central role in the initiation, development and outcome of the innate immune response. Long chain n-3 fatty acids may affect monocyte/macrophage defensive functions in several ways. For example, both EPA and DHA can increase phagocytosis\textsuperscript{54} and decrease chemotaxis of human monocyes.\textsuperscript{55,56} Cytokine expression can be modulated as well, since both EPA and DHA can down-regulate the in vitro production of IL-1\textbeta, IL-6 and TNF-\alpha.\textsuperscript{57,58} Monocytes/macrophages, together with neutrophils, produce high amount of reactive oxygen species (ROS), thus contributing to the oxidative stress, a prominent and common feature of many disease processes. Generation of ROS is of paramount importance for the killing of micro-organism. Yet, if not properly regulated, such as in chronic inflammatory or immune diseases, ROS become hazardous for the organism by causing damage to cellular lipids, proteins and DNA, eventually impairing cells function. Long chain PUFAs modulated cellular production of ROS in macrophages,\textsuperscript{59} neutrophils\textsuperscript{60} and lymphocytes,\textsuperscript{61} but their activity is still debated, since some authors find an increase while others, on the contrary, found a decrease in ROS production.\textsuperscript{52}

Regulation of leukocytes apoptosis is another important homeostatic mechanism to control the activity of monocytes, by reducing or increasing their lifespan during inflammation, eventually determining the positive outcome of the inflammatory process. There is increasing evidence that PUFAs can cause cell death in normal and cancer cells.\textsuperscript{62,64} Several cell types, including macrophages,\textsuperscript{62} neutrophils\textsuperscript{66} and leukaemia cells,\textsuperscript{61} exhibit morphological features of apoptosis and necrosis after exposure to PUFAs at high doses. The mechanisms by which PUFAs cause apoptosis vary markedly with cell type and fatty acid species, and most of them are still debated.

As discussed previously, studies carried out in ruminants had the prominent aim to evaluate the possible higher EPA and DHA content in animal-derived products, but there is the increasing evidence from the few available investigations that feeding ruminants with n-3 PUFAs may affect fertility\textsuperscript{60} and modulate immune response, including for example, lipopolysaccharide (LPS)-induced hyperthermia.\textsuperscript{66}

Dietary fish oil in transition dairy goats was found to be effective on cell-mediated immune response,\textsuperscript{75} with modified mononuclear and polymorphonuclear (PMN) cells ratio as result. Treating cells with DHA exerted\textsuperscript{48} in increased PMN phagocytic activity and lower reactive oxygen species (ROS) production after in vitro challenged with EPA and DHA. A subsequent validation in vivo of the obtained results demonstrated that both EPA and DHA have beneficial effects on goats health by improving the defensive performances of neutrophils\textsuperscript{49} avoiding cellular and tissue damages by ROS. EPA and DHA affected also goat monocyes activities by up-regulating phagocytic activity and ROS production\textsuperscript{59} and by interfering with the formation of lipid droplets and by upregulating proteins belonging to PAT protein family.\textsuperscript{71}

**Conclusion**

The transfer of health-promoting PUFAs such EPA and DHA can be achieved from dairy goat nutrition to human through the milk with positive effects also at animal level. The increased EPA and DHA milk content represent a future challenge to better characterize an already high-nutritional and health-providing product. Among different EPA and DHA sources, at the present moment fish oil still represents the preferable source, but algae can be an alternative as soon experimental results will be consolidated.

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**Conflict of interest**

Author declares that there is no conflict of interest.

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