Effects of Plant Extracts on Immune Response, Oxidative Stress, and Insulin Regulation in Ruminants: Postprint

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Abstract

Plant extracts are products derived from plant materials via chemical or physical methods for the directional extraction and concentration of one or more active ingredients without modifying their structural integrity. In recent years, plant extracts have garnered considerable attention as natural feed additives. Plant extracts are also widely applied in research and production related to ruminant nutrition. This review synthesizes domestic and international research progress, focusing primarily on three aspects: the effects of plant extracts on immune response, oxidative stress, and insulin regulation in ruminants.

Full Text

Effects of Plant Extracts on Immune Response, Oxidative Stress, and Insulin Regulation in Ruminants

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Abstract: Plant extracts are products derived from plant materials through chemical or physical methods that directionally obtain and concentrate one or more active ingredients without altering their chemical structures. In recent years, plant extracts have attracted widespread attention as natural feed additives and have been extensively studied and applied in ruminant nutrition research and production. This review summarizes current research progress on

the effects of plant extracts on immune response, oxidative stress, and insulin regulation in ruminants.

Keywords: plant extracts; immune response; oxidative stress; insulin; ruminants

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Plant extracts contain bioactive components such as alkaloids, saponins, essential oils, tannins, and polysaccharides, which possess antimicrobial, growth-promoting, and antioxidant functions. These extracts are considered natural alternatives to antibiotic drugs and have been widely investigated as potential rumen function modulators in ruminants. Since most nutrients required by ruminants originate from rumen microbial fermentation, which directly affects animal performance, plant extract additives can improve palatability while masking odors from manure or fertilizer. These characteristics align with future trends in agriculture and food production, demonstrating significant development potential.

Lu Dexun proposed the theory of "Nutritional Bioactive Substances Omics," offering a new perspective for plant extract research. This theory focuses on studying the interactions among naturally occurring nutritional bioactive substances and the optimized combinations of artificially formulated ones, emphasizing their integrated functions rather than isolated effects of single compounds. This approach has shifted research on nutritional bioactive substances, particularly plant extracts, from traditional nutrition to systematic animal nutrition, greatly advancing application studies. Research indicates that various plant extracts, including eugenol, cinnamaldehyde, allicin, and capsaicin, can activate transient receptor potential (TRP) channels in neurons, intestines, pancreas, immune cells, and other tissues in ruminants, triggering cellular activation and various physiological functions such as neuropeptide release and ion channel activation. Plant extracts modulate pro- or anti-inflammatory responses by altering inflammatory cytokines, leukocytes, and oxidative stress. Additionally, they regulate pancreatic hormones, particularly insulin secretion and sensitivity. In ruminants, plant extracts exert greater physiological effects on host animals than on rumen microorganisms. Studies infusing garlic oil, turmeric, and capsaicin into the intestine demonstrated significant increases in T helper cells associated with adaptive immunity without affecting gastrointestinal microbial communities in dairy cows. Furthermore, rumen-protected capsaicin reduced insulin secretion during glucose tolerance tests and alleviated acute-phase immune responses in LPS-induced inflammatory models in dairy cows. This review examines the effects of plant extracts on immune response, oxidative stress, and insulin regulation in ruminants to provide references for developing new plant extracts and elucidating their molecular mechanisms.

1. Immunomodulatory Effects of Plant Extracts on Ruminants

The immunomodulatory effects of plant extracts depend primarily on their chemical properties, host factors, and dosage, exhibiting bidirectional regulation. Tea saponin, for instance, modulates immune function in dairy cows by increasing serum immunoglobulin (Ig) levels, immune-related cytokine content, and interleukin-6 (IL-6) expression. Plant extracts activate TRP channels expressed in neurons, intestines, pancreas, and immune cells in mammalian tissues, enabling cellular activation or secondary transduction of ion transporters. Upon binding to TRP channels on cell membranes, plant extracts induce calcium ion (Ca^{2+}) influx, allowing nuclear factor kappa B (NF- B) and nuclear factor of activated T cells (NF-AT) to enter the nucleus and stimulating Ca^{2+} release from the endoplasmic reticulum through TRP channel activation. Eugenol exhibits anti-inflammatory and antioxidant properties after binding to TRPV1 and TRPV3, while cinnamaldehyde acts as an immune enhancer via TRPA1 mediation, and allicin activates TRPA1 and TRPV1 to modulate immunity. Capsaicin binds to TRPV1 to regulate immune responses and improve gastrointestinal mucosal blood flow, likely by modifying immune cells—including macrophages, neutrophils, and T and B cells—through stimulating or suppressing cytokines and antibodies. Capsaicin also indirectly affects immune cell function by binding to TRPV1 on afferent neurons and modulating neuropeptide release such as calcitonin gene-related peptide (CGRP), substance P, and tachykinins. CGRP demonstrates anti-inflammatory effects, while substance P and tachykinins are associated with vasodilation, plasma extravasation, and pro-inflammatory actions.

Plant extract-mediated receptor responses relate to immune function, gastrointestinal health, and host immunity. Ahmed et al. found that dietary supplementation with green tea by-products at various concentrations (0, 0.5%, 1.0%, and 2.0%) increased T and B cell populations and activated concanavalin A expression in goat spleen cells. Catechins, the main active components in green tea by-products, exhibited pro-inflammatory effects in goat spleen, though the mechanism remains unclear. Direct abomasal infusion of garlic oil, turmeric oleoresin, and capsaicin (2 g/d per head) in dairy cows significantly increased $CD4^+$ cell numbers and doubled CD25α proportions (an activation molecule on T lymphocytes), indicating T helper cell activation. In the same study, garlic oil tended to increase the neutrophil-to-lymphocyte ratio, suggesting an acute stress response. Subsequent research reported that dietary capsaicin increased neutrophil and eosinophil counts and elevated the neutrophil-to-lymphocyte ratio in peripheral blood of dairy cows, while also enhancing neutrophil phagocytosis, demonstrating promotion of acute-phase immune cell activation.

Plant extracts exhibit both pro-inflammatory and anti-inflammatory effects. Pro-inflammatory actions may stimulate immune responses, potentially beneficial for immunosuppressed cows during the dry period and early lactation. Anti-inflammatory effects reduce pro-inflammatory cytokine formation and acute-

phase immune responses. In dairy cows, feeding a mixture of curcumin, carnosic acid, cinnamaldehyde, and eugenol reduced milk somatic cell counts and improved udder health. Dietary rumen-protected capsaicin decreased haptoglobin expression and plasma cortisol concentration elevation induced by LPS stimulation in dairy cows. Yuan Liguo et al. demonstrated that dietary supplementation with 25 mg/kg oregano oil and 8 mg/kg flavomycin significantly prevented subclinical mastitis in dairy cows. Ye Wenchu et al. isolated and cultured pathogens from mastitic milk samples for in vitro antibacterial testing of *Sophora alopecuroides* extract, which showed significant antibacterial activity. However, the immunomodulatory effects of plant extracts depend on the animal' s immune status and dosage. Oh et al. found that low and medium doses of capsaicin were more effective than the highest dose for increasing neutrophil activity and total counts in dairy cows.

2. Regulation of Oxidative Stress by Plant Extracts in Ruminants

Plant extracts act as antioxidants by scavenging reactive oxygen species. One molecule of capsaicin can scavenge two peroxyl radicals through its phenolic hydroxyl group, while phenolic compounds can enhance endogenous antioxidant capacity. Plant secondary metabolites undergo detoxification through Phase I, II, and III metabolism, including hydrolytic biotransformation, conjugation reactions, and transporter-mediated efflux into the intestinal lumen or bile. Due to this detoxification, plasma and tissue concentrations of exogenous phenolic compounds are typically lower than endogenous antioxidants, which may explain the limited oxidative effects observed in animal studies. However, phenolic plant extracts can induce upregulation of genes associated with endogenous antioxidants and transcription factors, including aryl hydrocarbon receptors and nuclear factor E2-related factor. Garlic oil has been shown to scavenge free radicals and reduce lipid peroxidation.

Studies in ruminants have documented the effects of plant extracts on oxidative status. Dietary tannins (95.7 g/kg diet) enhanced antioxidant capacity in liver and blood of iron-reduced sheep. Zhong et al. found that feeding tea catechins to goats (0, 2,000, 3,000, or 4,000 mg/kg diet) improved antioxidant status by reducing oxidized glutathione (GSH) in blood. Catechins, the main components of tea polyphenols, exhibit pharmacological effects including lipid reduction, antithrombotic activity, blood pressure reduction, anti-mutation, anticancer properties, and antimicrobial effects, while improving animal metabolism and meat quality and promoting mammary gland development. These findings align with Ahmed et al.' s results showing that phenolic compounds increase endogenous antioxidant function in ruminants. Recent studies demonstrated that feeding juniper oil to goats (0.4–2.0 mL/kg diet, containing 89.7% α-pinene) increased superoxide dismutase (SOD) activity, while herbal powder containing hydrolyzable tannins, steroidal saponins, and glycoalkaloids (mixture of *Woodfordia fruticosa*, *Solanum nigrum*, and fenugreek) increased blood GSH content and catalase

(CAT) and glutathione S-transferase activities. In dairy cows, although garlic oil is known as an antioxidant, high doses may be harmful by increasing oxidative stress. Hashemzadeh-Cigari et al. reported that a plant extract mixture (including rosemary, cinnamon bark, turmeric, and clove bud) reduced blood thiobarbituric acid reactive substances (TBARS) concentration in postpartum dairy cows. Chestnut tannins (primarily hydrolyzable tannins) decreased blood malondialdehyde concentration and increased endogenous antioxidant enzyme activities (SOD and glutathione peroxidase) in dairy cows.

3. Regulation of Insulin by Plant Extracts in Ruminants

Insulin and glucagon are key hormones maintaining blood glucose homeostasis, and plant extracts can modulate insulin function. Research indicates that plant extracts enhance insulin activity through multiple mechanisms: increasing peroxisome proliferator-activated receptor-γ activity, upregulating insulin receptors and improving their function, increasing glycogen synthase activity and glycogen accumulation, attenuating inflammatory cytokine expression, accelerating fatty acid oxidation, and enhancing antioxidant status. Hashemzadeh-Cigari et al. reported that feeding dairy cows a plant extract mixture containing rosemary, cinnamon bark, turmeric, and clove buds increased insulin sensitivity. Intraduodenal administration of quercetin (18 mg/kg BW) twice daily significantly reduced blood glucose concentration and increased insulin concentration in dairy cows, demonstrating the hypoglycemic effect of the flavonoid quercetin in the intestine. In another dairy cow study, dietary capsaicin supplementation significantly reduced serum insulin concentration without affecting glucose concentration during glucose tolerance tests, consistent with results from rats and human subjects. Insulin plays a crucial role in energy partitioning in lactating dairy cows. While insulin is required by bone and muscle tissues, glucose uptake in the mammary gland depends primarily on glucose transporter 1 rather than insulin. Therefore, mammary glucose supply can be altered by insulin sensitivity in insulin-dependent tissues or pancreatic insulin secretion. Thus, plant extracts may redirect glucose for lactose synthesis and milk production through their regulatory effects on insulin sensitivity and secretion.

4. Conclusion

Natural plant extracts have become a research hotspot due to their natural properties, safety, non-polluting characteristics, and unique nutritional profiles that are incomparable to antibiotics and chemical additives. Although plant extracts exhibit multiple biological activities and some have been applied in animal production, several challenges remain. First, extraction of active components is largely limited to laboratory stages and often requires toxic reagents, causing environmental pollution and compromising extract safety. The complex extraction process also limits economic feasibility for animal production applications. Second, because plants contain multiple active components, the purity of plant extracts varies even within the same product, leading to inconsistent re-

search results and preventing the establishment of appropriate supplementation standards. Third, the mechanisms of action require further investigation. Most plant extract effects are documented only at the functional level, while molecular mechanisms affecting signaling pathways in animals remain unclear. Fourth, due to high costs of livestock experiments, particularly with large animals like dairy cows, many studies remain limited to in vitro tests, mouse models, or calf trials. Since the same plant extract may produce different effects across species and developmental stages, research findings are often inconsistent.

Further research and evaluation are needed to comprehensively understand the physiological regulatory mechanisms of plant extracts in ruminants and to explore their broad application prospects. It is necessary to combine effective active components with modern research techniques in nutrition, immunology, and molecular biology to investigate mechanisms from perspectives of nutrient metabolic pathways, immune regulation, and hormone secretion control.

References

[1] Sun Jie. Regulatory effects of plant extracts on rumen acidosis in ruminants[J]. China Feed, 2017(16):16-19,23.
[2] CALSAMIGLIA S,BUSQUET M,CARDOZO P W,et al.Invited review:essential oils as modifiers rumen microbial fermentation[J].Journal Dairy Science,2007,90(6):2580-2595.
[3] PATRA A K.Dietary phytochemicals and microbes[M].Dordrecht:Springer,2012:371-389.
[4] OH J,HRISTOV A N.Effects of plant-derived bio-active compounds on rumen fermentation,nutrient utilization,immune response,and productivity ruminant animals[M].2016.
[5] Lu Dexun. New concepts in feed science—interpreting theoretical and technical issues in feed science with systematic thinking[J]. Feed Industry, 2012, 33(1):1-7.
[6] HOLZER P.Transient receptor potential (TRP) channels as drug targets for diseases of the digestive system.[J].Pharmacology & Therapeutics,2011,131(1):142-170.
[7] VENNEKENS R,VRIENS J,NILIUS B.Herbal compounds and toxins modulating TRP channels[J].Current Neuropharmacology,2008,6(1):79-96.
[8] ZENG Z K,ZHANG S,WANG H L,et al.Essential oil and aromatic plants as feed additives in non-ruminant nutrition:a review[J].Journal Animal Science Biotechnology,2016,6(1):7.
[9] VAN DE WALL E H E M,GRAM D X,STRUBBE J H,et al.Ablation of capsaicin-sensitive afferent nerves affects insulin response during an intravenous glucose tolerance test[J].Life Sciences,2005,77(11):1283-1292.
[10] QIN B L,PANICKAR K S,ANDERSON R A.Cinnamon:potential role in the prevention of insulin resistance,metabolic syndrome,and type 2 diabetes[J].Journal of Diabetes Science and Technology,2010,4(3):685-693.
[11] OH J,HRISTOV A N,LEE C,et al.Immune and production responses of

dairy cows to postruminal supplementation with phytonutrients[J].Journal of Dairy Science,2013,96(12):7830-7843.
[12] OH J,HARPER M,GIALLONGO F,et al.Effects of rumen-protected Capsicum oleoresin on immune responses in dairy cows intravenously challenged with lipopolysaccharide[J].Journal of Dairy Science,2017,100(3):1902-1913.
[13] OH J,HARPER M,GIALLONGO F,et al.Effects of rumen-protected Capsicum oleoresin on productivity and responses to a glucose tolerance test in lactating dairy cows[J].Journal of Dairy Science,2017,100(3):1888-1901.
[14] Shi Ning, Xu Xiaokai, Fang Luoyun, et al. Effects of plant extracts on immune function and milk performance of dairy cows[J]. China Herbivore Science, 2017, 37(1):50-54.
[15] Chang Xiaoxiao, Zhang Yifu, Zhao Shiping, et al. Effects of tea saponin on immune function of dairy cows[J]. Chinese Journal of Animal Nutrition, 2017, 29(3):10.
[16] BERRIDGE M J,LIPP P,BOOTMAN M D.The versatility and universality of calcium signalling[J].Nature Reviews Molecular Cell Biology,2000,1(1):11-21.
[17] GEES M,COLSOUL B,NILIUS B.The role of transient receptor potential cation channels in Ca2+ signaling[J].Cold Spring Harbor Perspectives in Biology,2010,2(10):a003962.
[18] FRANCO-PENTEADO C F,DE SOUZA I A,LIMA C S P,et al.Effects of neonatal capsaicin treatment in the neutrophil production,and expression of preprotachykinin-I and tachykinin receptors in the rat bone marrow[J].Neuroscience Letters,2006,407(1):70-73.
[19] TAKANO F,YAMAGUCHI M,TAKADA S,et al.Capsicum ethanol extracts and capsaicin enhance interleukin-2 and interferon-gamma production in cultured murine Peyer' s patch cells ex vivo[J].Life Sciences,2007,80(17):1553-1563.
[20] NEVIUS E,SRIVASTAVA P K,BASU S.Oral ingestion of Capsaicin,the pungent component of chili pepper,enhances a discreet population of macrophages and confers protection from autoimmune diabetes[J].Mucosal Immunology,2012,5(1):76-86.
[21] TSUJI F,AONO H.Role of Transient receptor potential vanilloid 1 in inflammation and autoimmune diseases[J].Pharmaceuticals,2012,5(8):837-852.
[22] ZIMMERMAN B J,ANDERSON D C,GRANGER D N.Neuropeptides promote neutrophil adherence to endothelial cell monolayers[J].The American Journal of Physiology,1992,263(5 Pt 1):G678-G682.
[23] PENNEFATHER J N,PATAK E,PINTO F M,et al.Mammalian tachykinins and uterine smooth muscle:the challenge escalates[J].European Journal Pharmacology,2004,500(1/2/3):15-26.
[24] FURNESS J B,RIVERA L R,CHO H J,et al.The gut as a sensory organ[J].Nature Reviews Gastroenterology & Hepatology,2013,10(12):729-740.
[25] AHMED S T,LEE J W,MUN H S,et al.Effects of supplementation with green tea by-products on growth performance,meat quality,blood metabolites and immune cell proliferation in goats[J].Journal of Animal Physiology and Animal Nutrition,2015,99(6):1127-1137.

[26] WEISS D J,WARDROP K J.Schalm's veterinary hematology[M].Iowa,USA:Blackwell Publishing,2010.
[27] OH J,GIALLONGO F,FREDERICK T,et al.Effects of dietary Capsicum oleoresin on productivity and immune responses in lactating dairy cows[J].Journal of Dairy Science,2015,98(9):6327-6339.
[28] SORDILLO L M,AITKEN S L.Impact of oxidative stress on the health and immune function of dairy cattle.[J].Veterinary Immunology and Immunopathology,2009,128(1/2/3):104-109.
[29] HASHEMZADEH-CIGARI F,GHORBANI G R,KHORVASH M,et al.Supplementation of herbal plants differently modulated metabolic profile,insulin sensitivity,and oxidative stress in transition dairy cows fed various extruded oil seeds[J].Preventive Veterinary Medicine,2015,118(1):45-55.
[30] Yuan Liguo, Tong Hengmin, Shen Jianzhong. Study on application of oregano oil and flavomycin in dairy cow production[J]. Feed Industry, 2004, 25(1):38-42.
[31] Ye Wenchu, Yuan Fuwei, Ban Yun, et al. In vitro antibacterial activity of Sophora alopecuroides extract against mastitis pathogens in dairy cows[J]. Chinese Journal of Veterinary Science, 2016, 36(9):1568-1571.
[32] AMORATI R,FOTI M C,VALGIMIGLI L.Antioxidant activity of essential oils[J].Journal of Agricultural and Food Chemistry,2013,61(46):10835-10847.
[33] OKADA Y,TANAKA K,SATO E,et al.Kinetics and antioxidative sites of capsaicin in homogeneous solution[J].Journal of the American Oil Chemists' Society,2010,87(12):13.
[34] MASELLA R,DI BENEDETTO R,VARÌ R,et al.Novel mechanisms of natural antioxidant compounds in biological systems:involvement of glutathione and glutathione-related enzymes[J].The Journal of Nutritional Biochemistry,2005,16(10):577-586.
[35] GURLEY B J.Emerging Technologies for improving phytochemical bioavailability:benefits and risks[J].Clinical Pharmacology and Therapeutics,2011,89(6):915-919.
[36] SEYMOUR E M,BENNINK M R,BOLLING S F.Diet-relevant phytochemical intake affects the cardiac AhR and nrf2 transcriptome and reduces heart failure in hypertensive rats[J].The Journal of Nutritional Biochemistry,2013,24(9):1580-1586.
[37] MIRUNALINI S,DHAMODHARAN G,KARTHISHWARAN K.A natural wonder drug helps to prevent cancer:garlic oil[J].Notulae Scientia Biologicae,2010,2(1):14-19.
[38] LÓPEZ-ANDRÉS P,LUCIANO G,VASTA V,et al.Dietary quebracho tannins are not absorbed,but increase the antioxidant capacity of liver and plasma in sheep[J].British Journal of Nutrition,2013,110(4):632-639.
[39] ZHONG R Z,XIAO W J,REN G P,et al.Dietary tea catechin inclusion changes plasma biochemical parameters,hormone concentrations and glutathione redox status in goats [J].Asian-Austral Aslan Journal of Animal Sciences,2011,24(12):1681-1689.
[40] Deng Xiaowen, Xi Weibin, Zhang Junling. Effects of plant extracts on performance and rumen fermentation in ruminants[J]. Modern Journal of

Animal Husbandry and Veterinary Medicine, 2010(6):44-47.
[41] YESILBAG D,BIRICIK H,CETIN I,et al.Effects of juniper essential oil on growth performance,some rumen protozoa,rumen fermentation and antioxidant blood enzyme parameters of growing Saanen kids[J].Journal of Animal Physiology & Animal Nutrition,2017,101(5):e67-e76.
[42] CHOUBEY M,PATTANAIK A K,BALIYAN S,et al.Dietary supplementation of a novel phytogenic feed additive:effects on nutrient metabolism,antioxidant status and immune response of goats[J].Animal Production Science,2015,56(10):1612.
[43] LAMBERT J D,ELIAS R J.The antioxidant and pro-oxidant activities of green tea polyphenols:a cancer prevention[J].Archives Biochemistry Biophysics,2010,501(1):65-72.
[44] LIU H W,ZHOU D W,LI K.Effects of chestnut tannins on performance and antioxidative status of transition dairy cows[J].Journal of Dairy Science,2013,96(9):5901-5907.
[45] ANDERSON R A.Chromium and polyphenols from cinnamon improve insulin sensitivity:plenary lecture[J].Proceedings of The Nutrition Society,2008,67(1):48-53.
[46] GOHLKE A,INGELMANN C J,NÜRNBERG G,et al.Influence of 4-week intraduodenal supplementation of quercetin on performance,glucose metabolism,and mRNA abundance of genes related to glucose metabolism and antioxidative status in dairy cows[J].Journal of Dairy Science,2013,96(11):6986-7000.
[47] AHUJA K D,ROBERTSON I K,GERAGHTY D P,et al.Effects of chili consumption on postprandial glucose,insulin,and energy metabolism[J].The American Journal of Clinical Nutrition,2006,84(1):63-69.
[48] VAN DE WALL E H E M,WIELINGA P Y,STRUBBE J H,et al.Neonatal capsaicin causes compensatory adjustments energy homeostasis rats[J].Physiology & Behavior,2006,89(1):115-121.
[49] DE KOSTER J D,OPSOMER G.Insulin resistance in dairy cows[J].Veterinary Clinics of North America: Food Animal Practice,2013,29(2):299-322.
[50] ZHAO F Q,KEATING A F.Expression and regulation of glucose transporters in the bovine mammary gland[J].Journal of Dairy Science,2007,90(Suppl.1):E76-E86.

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