

ESSENTIAL OILS AND FATTY ACIDS AS SILAGE ADDITIVES: A REVIEW

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Abstract

The search for natural alternatives to conventional silage additives has stimulated interest in plant-derived compounds capable of modulating microbial activity during ensiling. This review aimed to compile and critically analyse the available literature on the use of essential oils and fatty acids as additives in silage production. A systematic search of major scientific databases identified 28 studies evaluating essential oils applied to a variety of forage silages, whereas information on fatty acids was extremely limited. Most experiments tested crude plant essential oils, frequently with incomplete descriptions of their chemical composition, and only a few studies evaluated individual bioactive compounds. Overall, the responses to essential oil application varied considerably depending on the forage species, oil composition, application rate and storage conditions. Alfalfa silage appeared to respond more consistently to several essential oils, with reports of enhanced lactic acid production, reduced proteolysis and improved aerobic stability. In whole-plant corn silage, the most consistent effect across studies was the improvement of aerobic stability, whereas impacts on fermentation quality were variable and occasionally suggested compromised acidification. For other forage types, results were inconsistent and often limited by experimental design or lack of microbiological data. The literature on fatty acids as silage additives remains scarce, although available evidence suggests that these compounds may exert selective antimicrobial pressure capable of influencing fermentation processes. Overall, current knowledge remains insufficient to establish clear recommendations for the use of these compounds in silage systems. Future research should prioritise the evaluation of individual bioactive molecules rather than crude oils, incorporate molecular approaches to characterise microbial modulation, and include detailed assessments of the early fermentative phase to better understand the mechanisms through which these compounds influence silage fermentation and preservation.

Keywords: aerobic stability, essential oils, fatty acids, silage, additives

INTRODUCTION

Increasing emphasis has been placed on the adoption of natural compounds as replacements for synthetic chemical additives in various agribusiness systems. In the field of silage processing, the research on vegetable essential oils for use as additives is still quite modest and incipient. The available information is limited to some forage crops, and the reported effects still derive from crude oils extracted from plants and seeds. Potential antimicrobial activities on silage microbiota were summarized by Chen *et al.* (2023), suggesting that plant essential oils and their active components are more inhibitory against yeasts, fungi and pathogenic bacteria, including Gram-negative ones such as *Escherichia coli*, whereas far less inhibitory against lactic acid bacteria (LAB). These effects, found in *in vitro* studies, suggest that plant essential oils could be very promising as additives for silages, favouring conservation quality while inhibiting undesirable microorganisms. A meta-analysis by Susanto *et al.* (2023), however, did not corroborate these expectations in *in silo* studies. Unfortunately, the authors recognized that their conclusions were highly biased as all oils were considered as a single entity, regardless of their composition. The same approach was used for the evaluated silages across studies, whose diversity was not considered. Thus, the present review aimed to analyse the effects of each plant essential oil for each type of silage evaluated in the available literature so far.

Fatty acids, which are components of vegetable oils extracted from plant seeds, represent another group of natural compounds that have been even less investigated as silage additives. Although their ability to modulate microbial activity and fermentation is well established within the ruminal microbiota (Ibrahim *et al.*, 2021), their application for similar modulatory purposes in silages remains virtually unknown. Thus, the present review also summarized the available information regarding fatty acids as silage additives.

MATERIAL AND METHODS

A comprehensive search was performed using Scopus, Science Direct, Pubmed and Web of Science platforms through the keywords “essential oil” and “silage” (Figure 1). From the retrieved papers, 28 were related to essential oils being applied in silage processing (Table 1), published in 25 different journals of several impact factors. Most of the published papers were from Brazil (n=8, 28.6%), Iran (n=7, 25.0%), and Turkey (n=6, 21.4%), and four papers were not published in English, being two written in Persian (Maghsoudloo *et al.*, 2017, Besharati *et al.*, 2020a), one in Turkish (Önenç *et al.*, 2017) and one in Chinese (Zhou *et al.*, 2024). Twenty-one different plant essential oils were evaluated, being cinnamon (n=6, 21.4%) and oregano (n=6, 21.4%) the most explored, followed by cumin (n=4, 14.3%), lemongrass (n=3, 10.7%), lemon seed (n=3, 10.7%) and others. One paper focused on a commercial blend (Kung Jr. *et al.*, 2008) and only two papers evaluated individual essential oil compounds (Foskolos *et al.*, 2016, Sousa *et al.*, 2022). When not possible to work with essential oil compounds individually, recommendations have already been made in previous meta-analyses (Calsamiglia *et al.*, 2007, Cobellis *et al.*, 2016) to report a description as complete as possible of the composition of essential oils in future research. However, only seven studies presented a full description of components and their proportions in their essential oils, and other six at least described the main components. Eleven studies, however, did not report the composition of their essential oils.

Seventeen different silages were used in the trials, being alfalfa silage the most explored (n=8, 28.6%), followed by whole-plant corn silage (n=5, 17.9%), ryegrass silage (n=2, 7.1%), field pea silage (n=2, 7.1%) and other forages that were evaluated only once. It is noteworthy that to the moment only forage silages have been used in published studies, with the exception of Andrade *et al.* (2024) who ensiled a total mixed ration (TMR) containing whole plant sorghum and ground corn grains. Despite cereal plants such as whole plant corn, sorghum and barley also include grains when harvested for silage processing, no data for essential oils as additives on exclusive grain silages is available in peer reviewed papers.

Tab. 1: Compiled studies with essential oils as silage additives

Reference	Journal	Origin ¹	Silage	Essential oil/Compound	Composition data
Kung Jr. <i>et al.</i> (2008)	J. Dairy Sci.	USA	Whole plant corn	Commercial Blend	Main components listed
Chaves <i>et al.</i> (2012)	J. Sci. Food Agric.	Australia	Whole plant barley	Cinnamon, Oregano, Sweet orange	Main components described
Önenç <i>et al.</i> (2015)	Asian Australas. J. Anim. Sci.	Turkey	Field pea	Oregano, Cinnamon	Fully described
Foskolos <i>et al.</i> (2016)	Canadian J. Anim. Sci.	Spain	Ryegrass	Thymol, Eugenol, Cinnamaldehyde, Capsaicin, Carvacrol	Essential oil compounds
Hodjatpanah-Montazeri <i>et al.</i> (2016)	Iranian J. Anim. Sci.	Iran	Whole plant corn	Cinnamon, Thyme, Oregano, Mint, Cumin	Not reported
Maghsoudloo <i>et al.</i> (2017)	Iranian J. Anim. Sci. Res.	Iran	Whole plant corn	Rosemary, Fennel, <i>Carum copticum</i>	Not reported
Önenç <i>et al.</i> (2017)	J. Anim. Prod.	Turkey	Field pea	Oregano, Cinnamon	Fully described
Turan and Önenç (2018)	Asian Australas. J. Anim. Sci.	Turkey	Alfalfa	Cumin	Fully described
Önenç and Turgud (2019)	J. Agric. Faculty of Ege University	Turkey	Alfalfa	Oregano, Cumin, Cinnamon	Fully described
Cantoia Jr. <i>et al.</i> (2020)	Anim. Feed Sci. Technol.	Brazil	Sugarcane	Lemongrass	Not reported
Besharati <i>et al.</i> (2020a)	Research on Anim. Prod.	Iran	Alfalfa	Flaxseed	Not reported
Besharati <i>et al.</i> (2020b)	Acta agriculturae Slovenica	Iran	Alfalfa	Flaxseed, Cinnamon, Lemon seed	Not reported
Besharati and Niazifar (2020)	J. Anim. Sci.	Iran	Alfalfa	Lemon seed	Not reported
Vaičiulienė <i>et al.</i> (2020)	J. Microbiol. Biotechnol.	Lithuania	Ryegrass, red clover, alfalfa	Oregano, Thyme	Main components described
Besharati <i>et al.</i> (2021a)	Semina Ciências Agrárias	Iran	Alfalfa	Lemon seed	Fully described
Besharati <i>et al.</i> (2021b)	South African J. Anim. Sci.	Iran	Alfalfa	Ricinoleic acid + cardol + cardanol	Main components described
Akinci and Önenç (2021)	J. Agric. Faculty of Ege University	Turkey	Vetch + oat	Cumin	Fully described
Li <i>et al.</i> (2022)	Frontiers	China	Paper mulberry	<i>Amomum villosum</i>	Not reported
Del Valle <i>et al.</i> (2022)	The Journal of Agricultural Science	Brazil	Whole plant shorgum	Lemongrass	-
Sousa <i>et al.</i> (2022)	Brazilian Anim. Sci.	Brazil	Sorghum + <i>Urochloa brizantha</i>	Rosemary, Thymol	Fully described
Garcia <i>et al.</i> (2022)	Anim. Prod. Sci.	Brazil	Mombasa grass	Lemongrass	-
Silva <i>et al.</i> (2024a)	Scientia Agricola	Brazil	Whole plant corn	Sweet orange	Main components described
Duru (2024)	J. Hell. Vet. Med. Soc.	Turkey	Vetch + wheat	<i>Hypericum perforatum</i>	Not reported
Andrade <i>et al.</i> (2024)	Agriculture	Brazil	Total Mixed Ration	Limonene-rich essential oil	Not reported
Silva <i>et al.</i> (2024b)	Scientia Agricola	Brazil	Whole plant corn	Blend from <i>Copaifera langsdorffii</i> and <i>Eucalyptus staigeriana</i>	Main components described
Aniceto <i>et al.</i> (2024)	Fermentation	Brazil	Whole plant sorghum	Rosemary, Tea tree, Citronella	Main components described
Zhou <i>et al.</i> (2024)	Acta Agrestia Sinica	China	<i>Stylosanthes guianensis</i> , Rice straw	<i>Neolamarckia cadamba</i> leaf	Not reported
Xu <i>et al.</i> (2023)	Letters in Applied Microbiology	China	<i>Broussonetia papyfera</i> , <i>Pennisetum sinense</i>	<i>Amomum villosum</i>	Not reported

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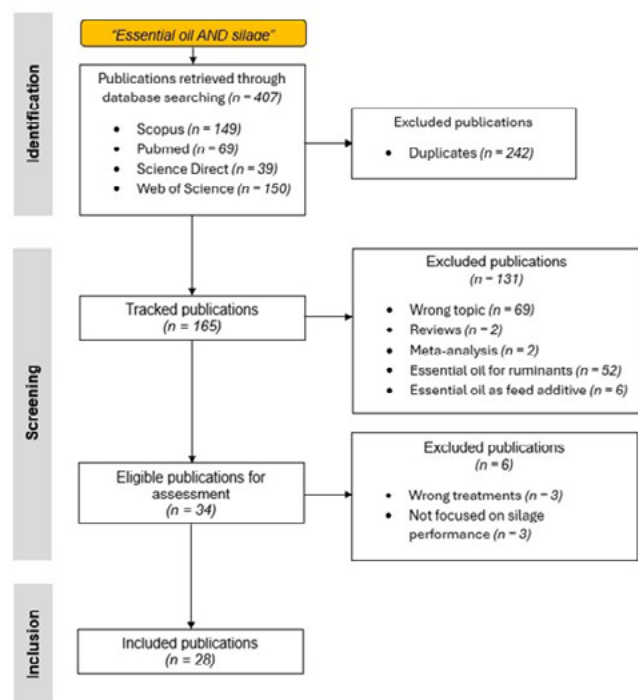


Fig. 1: Flowchart of selection of the studies included in the review of essential oils as additives for silages

RESULTS AND DISCUSSION

Alfalfa silage

Alfalfa has been the relatively most explored forage silage to evaluate essential oils as additives. As a legume forage, it presents some particularities that usually challenge the conservation process, such as the high protein content, which confers buffering capacity and makes the forage more susceptible to extensive proteolysis in the silo. Eight studies were published with seven different essential oils, ranging from 60 to 120 days of storage and application rates from 60 to 600 mg/kg fresh matter (FM). Chemical composition, fermentative profile, microbial counts and aerobic stability results are summarized in Supplementary Tables S1, S2, S3 and S4, respectively. Oregano essential oil was evaluated by Önenç and Turgud (2019) with very promising results, including increased LAB population and greater yield of lactic acid, which favored silage acidification, greater maintenance of crude protein (CP) content and lower dry matter (DM) losses. There was considerable inhibition of yeasts and fungi, although no information on aerobic stability was made available. Vaičiulienė *et al.* (2020) also reported lower silage pH with oregano and thyme essential oils, although only oregano essential oil was able to reduce yeast and fungi counts. With cumin essential oil, two studies reported similar findings, in addition to decreased enterobacteria counts and improved aerobic stability (Turan and Önenç, 2018, Önenç and Turgud, 2019). Lemonseed essential oil at application rates of 60 and 120 mg/kg FM (Besharati and Niazifar, 2020, Besharati *et al.*, 2020b, Besharati *et al.*, 2021a) also enhanced lactic acid production, promoted lower silage pH values, preserved CP content and improved aerobic stability. Cinnamon essential oil was tested at application rates of 60 (Besharati *et al.*, 2020b) and 600 mg/kg FM (Önenç and Turgud, 2019) on alfalfa silage stored for 60 days. Both application rates enhanced lactic acid production, increased LAB population, reduced pH and DM losses, preserved the CP content and reduced ammonia nitrogen (NH₃-N) concentration. Inhibition

of yeasts and fungi counts and improved aerobic stability were also reported.

An unidentified oil containing ricinoleic acid, cardol and cardanol at 500 mg/kg FM preserved CP content in alfalfa silage stored for 90 days and enhanced aerobic stability, although without changes in silage pH (Besharati *et al.*, 2021b). No information on microbial counts and organic acids production was made available in that study. Flaxseed essential oil increased lactic acid production and reduced silage pH, managing to preserve the CP content and to enhance aerobic stability (Besharati *et al.*, 2020a, 2020b). No information on microbial modulation was made available. As flaxseed essential oil also contains fatty acids in its composition, mainly unsaturated ones such as oleic and α -linolenic acids, the reported effects may be confounded by multiple classes of bioactive molecules. Unsaturated fatty acids are known to promote microbial modulation on several strains of LAB (Kodicek and Worden, 1945, Talon *et al.*, 2000, Kankaanpää *et al.*, 2001, Endo *et al.*, 2006), either by favoring or inhibiting their growth and activity.

Therefore, essential oils of different compositions have been consistently yielded desirable outcomes when tested as additives for alfalfa silage, including favored LAB development, silage acidification, protein preservation and improved aerobic stability. Only cinnamon essential oil was also associated with decreased deamination. Future research would benefit from molecular techniques to better understand the microbial succession during the early fermentative stage of alfalfa silage treated with essential oils.

Whole-plant corn silage

Five studies were available with whole-plant corn silage, evaluating 11 different plant essential oils and one commercial blend, with application rates varying from 40 to 600 mg/kg FM, and storage lengths from 3 to 256 days. Each essential oil was evaluated in only one study, so it was not possible to compare findings among trials. Chemical composition and fermentative profile are presented in Supplementary Tables S5 and S6, respectively. Although the commercial blend at 40 and 80 mg/kg FM had no effect on whole-plant corn silage after 256 days of storage (Kung Jr. *et al.*, 2008), individual essential oils showed results within 60 days (Hodjatpanah-Montazeri *et al.*, 2016, Maghsoudloo *et al.*, 2017, Silva *et al.*, 2024a, 2024b). The CP content was kept higher than control silage when applying mint and oregano essential oils, which might suggest decreased proteolysis. However, mint, oregano and cumin essential oils favored deamination and hampered the silage acidification. Only thyme essential oil reduced NH₃-N content. Rosemary and fennel essential oils managed to consistently preserve the content of soluble carbohydrates relative to the control silage at application rates of 125 and 250 mg/kg FM up to 45 days of storage. Fluctuations in CP and NH₃-N contents were reported in the first days after ensiling, although without a consistent pattern (Maghsoudloo *et al.*, 2017). The reduced proteolysis and lower carbohydrate consumption reported may suggest a compromised fermentation activity by some oils. The increase in NH₃-N concentration may suggest little or no antimicrobial effect by essential oils on undesirable microorganisms responsible for deamination, which are more active in an insufficiently acidified silage environment. Sweet orange essential oil was the only one able to reduce DM losses in whole-plant corn silage, as reported by Silva *et al.* (2024a) with application rates at 400 and 600 mg/kg FM. Cinnamon, thyme, oregano, cumin, sweet orange, eucalyptus and copaiba essential oils markedly enhanced the aerobic stability of whole-plant corn silage (Hodjatpanah-Montazeri *et al.*, 2016, Silva *et al.*, 2024a, 2024b). This enhancement has been the most consistent effect

reported among trials (Supplementary Table S8). The antimicrobial effects of essential oils are usually more pronounced against yeasts and fungi than against bacteria (Chen *et al.*, 2023), although the available data on microbial counts do not consistently support this hypothesis (Supplementary Table S7). Silva *et al.* (2024a, 2024b) reported increased acetic acid production by sweet orange essential oil applied at 600 mg/kg FM and by blends of *Eucalyptus staigeriana* and *Copaifera langsdorffii* applied at 500 mg/kg FM, which may contribute to enhanced silage stability during aerobic exposure.

Overall, essential oils appear to consistently enhance the aerobic stability of whole-plant corn silage, although the conservation quality may not be guaranteed. A decrease in proteolysis would be of interest in forage silages, although a slower acidification rate would not, as it could result in greater DM losses. There is clearly a need for more information on essential oils as additives for whole-plant corn silage. Maghsoudloo *et al.* (2017) were the only ones to report findings across multiple storage lengths including the early fermentative stage, and this approach would allow better understanding of how essential oils affect fermentation dynamics in future research. The microbial modulation associated to essential oils may also be more relevant at the early fermentative stage of silages, and molecular tools such as metagenomics and metabolomics might help to better describe and understand the microbial succession.

Other silages

A variety of other forages have been explored to test the effect of essential oils on silage quality and aerobic stability. Fifteen studies were compiled including twelve different forages and four combined silages and testing twelve different essential oils and five essential oil compounds. The available results for chemical composition, fermentative profile, microbial counts and aerobic stability are presented in Supplementary Tables S9, S10, S11 and S12, respectively. Besides alfalfa, other legume forages such as field pea and red clover were evaluated. Field pea was used by Önenç *et al.* (2015, 2017) to test oregano and cinnamon essential oils. Oregano essential oil, contrary to observations in alfalfa silage, apparently compromised the fermentation of field pea silage at an application rate of 400 mg/kg FM. Higher $\text{NH}_3\text{-N}$ contents and decreased lactic and acetic acids production were reported, resulting in higher silage pH and greater DM losses after 120 days of storage. Cinnamon essential oil inhibited deamination in field pea silage stored for 60 days and reduced DM losses, despite the decreased lactic and acetic acids production and higher silage pH. At 120 days, however, greater $\text{NH}_3\text{-N}$ concentration was observed, and DM recovery was no longer improved. Both oregano and cinnamon essential oils increased yeast counts, an effect completely opposite to the commonly reported on alfalfa silage. Overall, the positive effects consistently reported by applying essential oils in alfalfa silage do not appear to occur to the same extent in field pea silage. Red clover silage was used by Vaičiulienė *et al.* (2020), in which thyme essential oil increased silage pH and both oregano and thyme essential oils reduced LAB counts after 96 days of storage, whereas no effect was observed on yeast and fungi counts. It remains to be clarified whether legume forages other than alfalfa may benefit from essential oil-based treatments.

In addition to whole-plant corn silage, other cereal forages were evaluated. Chaves *et al.* (2012) tested cinnamon leaf, oregano and sweet orange essential oils at 125, 250 and 400 mg/kg FM on whole plant barley. However, the reported findings were obtained after only one year of storage, which may have limited the detection of significant effects. Only

a preserved CP content was reported with cinnamon and sweet orange essential oils, whereas no effects on microbiota or fermentation were observed for any of the tested oils. Del Valle *et al.* (2022) tested lemongrass essential oil at application rates between 1 and 4 mL/kg DM on a whole plant sorghum silage stored for 167 days. Increasing application rates linearly reduced butyric acid content and lactic-to-acetic acid ratio. However, $\text{NH}_3\text{-N}$ concentration increased along with greater CP disappearance in treated silages. Despite reducing gas losses, lemongrass essential oil had no effect on DM losses and aerobic stability of whole plant sorghum silage.

Grass silages were evaluated in five studies. Garcia *et al.* (2022) tested lemongrass essential oil at application rates up to 2 mL/kg FM on Mombasa grass silage stored for 60 days. Despite not improving DM recovery, desirable outcomes were obtained including reduced gas losses and $\text{NH}_3\text{-N}$ concentration, increased degradation of acid detergent fibre, and a lower silage pH after aerobic exposure that might suggest an enhanced aerobic stability. Foskolos *et al.* (2016) worked with individual essential oil compounds on ryegrass silage stored for 35 days. Capsaicin had no effect, whereas eugenol, cinnamaldehyde, carvacrol and thymol reduced $\text{NH}_3\text{-N}$ concentration, although none of them had any effect on CP content. Carvacrol and thymol, main components of oregano essential oil, reduced lactic acid production and increased silage pH, although carvacrol achieved these effects at a lower application rate. Only thymol reduced LAB counts. Vaičiulienė *et al.* (2020) reported contrasting results in which oregano and thyme essential oils reduced pH on ryegrass silage without significant effect on LAB counts. Unfortunately, no other fermentative parameter, such as acids production or $\text{NH}_3\text{-N}$ concentration, was assessed in that study. These essential oils also reduced yeast and fungi counts on ryegrass silage, although aerobic stability was not assessed. Zhou *et al.* (2024), testing an essential oil from *Neolamarkia cadamba* leaf, reported increased lactic acid production, decreased pH and reduced $\text{NH}_3\text{-N}$ content on *Stylosanthes guianensis* silage stored for 30 days, although LAB counts were reduced relative to the control silage. No information on DM losses and aerobic stability was made available. It is noteworthy in that trial that the essential oil was applied at 5 and 10 g/kg FM, considerably higher application rates than those usually reported in the literature. Xu *et al.* (2023) tested *Amomum villosum* essential oil on *Pennisetum sinense* silage and reported decreased $\text{NH}_3\text{-N}$ content and increased production of lactic and acetic acids, although without changes in silage pH. Again, no information on DM losses and aerobic stability was made available.

Only one study evaluated sugarcane silage by applying lemongrass essential oil at 1, 2 and 3 mL/kg FM. Cantoia Jr. *et al.* (2020) reported inhibition of yeast and fungi counts, decreased ethanol production and enhanced aerobic stability. There was evidence, however, of compromised fermentation, including decreased lactic and acetic acids yield and lower consumption of soluble carbohydrates, greater $\text{NH}_3\text{-N}$ concentration and increasing pH with increasing application rates of the essential oil. However, lower DM losses and CP degradation were also reported. Li *et al.* (2022) ensiled paper mulberry treated with *Amomum villosum* essential oil at 1 mL/kg FM. Despite consuming less soluble carbohydrates, the silage presented increased production of lactic and acetic acids, decreased $\text{NH}_3\text{-N}$ concentration and lower pH, suggesting enhanced fermentation by desirable bacteria, although no microbial assessment was made available.

Four studies tested essential oils on combined silages. Sousa *et al.* (2022) used sorghum intercropped with *Urochloa brizantha* ensiled for 15, 30 and 45 days to evaluate Lippia

sidoides essential oil and nanoformulated thymol (62% and 100%) at 400 mg/kg FM. Treatment with pure thymol was able to enhance aerobic stability after 15 days of storage, whereas at 30 and 45 days only the 62% nanoformulated thymol managed to promote this effect. Thymol reduced LAB counts and increased silage pH. Despite being more effective at inhibiting clostridia, pure thymol increased DM, gas and effluent losses relative to the control silage. However, the 62% nanoformulated thymol reduced DM and effluent losses. *L. sidoides* essential oil was able to reduce DM, gas and effluent losses, but did not inhibit clostridial growth. Aerobic stability was enhanced by thymol at both nanoformulations, whereas *L. sidoides* was not able to promote this effect. Akinci and Önenç (2021) reported several findings for a vetch + oat combined silage treated with 200, 300 and 500 mg/kg FM of cumin essential oil. Despite the increased lactic acid production and LAB counts, maintenance of CP content and decreased deamination, no effect was observed on silage pH and DM losses. Yeast, fungi and enterobacteria counts were reduced, and the treated silage presented a lower pH after a period of aerobic exposure, which might suggest enhanced aerobic stability. Duru (2024) reported improved fermentation quality when applying *Hypericum perforatum* essential oil between 20 and 160 mg/kg FM on a vetch + wheat combined silage. Xu *et al.* (2023), testing *A. villosus* essential oil on *Broussonetia papyfera* intercropped with *Pennisetum sinense* ensiled for 3 and 30 days, reported increased CP content relative to the control silage, decreased $\text{NH}_3\text{-N}$ content and increased production of lactic and acetic acids. No information on DM losses and aerobic stability was made available.

Essential oils as silage additives: general considerations

So far, alfalfa has been the only forage silage that appears to consistently benefit from several different essential oils as additives. Reported outcomes included enhanced lactic acid production and silage acidification, decreased storage losses and proteolysis, and consistent improvement of aerobic stability by inhibiting yeast and fungi presence. For whole-plant corn silage, the only consistent and desirable outcome among multiple essential oils was the enhancement of aerobic stability, whereas there was evidence of compromised fermentation along with extended activity of deaminating microorganisms. Unlike alfalfa silage, other legume silages

did not respond well to essential oil application, resulting in compromised fermentation, lower acidification, and greater $\text{NH}_3\text{-N}$ concentration. For whole plant cereal silages other than corn, the available findings were still inconclusive and probably limited by the extended storage lengths imposed by the trials. For grass silages, the decreased $\text{NH}_3\text{-N}$ concentration was the most consistent outcome, although evidence of compromised fermentation has also been reported, and no data was available on aerobic stability.

Oregano, cinnamon, and lemonseed essential oils appear to favor LAB activity in alfalfa silage, whereas in other silages they either suppressed lactic acid production or had no effect. Both oregano and cinnamon essential oils either favored and inhibited deamination in silages, depending on type of forage and storage length. Curiously, greater CP contents in treated silages have been consistently reported even in scenarios of increased deamination. As greater CP contents have also been reported in silages with reduced DM losses, protein concentration may be ruled out as an explanation. The most consistent and predictable effect of different essential oils on silages has been the enhancement of aerobic stability, either through inhibition of yeast and fungi growth or by increasing acetic acid production. There was only one contrasting report of favored yeast growth in field pea silage treated with cinnamon and oregano essential oils (Önenç *et al.*, 2015). Testing pure essential oil compounds using appropriate methodologies is required to unveil molecule-specific effects on silage fermentation kinetics. This would contribute to making the development of essential oil-based additives more rational, strategic, and performance-oriented.

Fatty acids and seed oils as silage additives

Fatty acids represent another group of natural compounds that have been even less investigated as silage additives. Although their ability to modulate microbial activity and fermentation is well established within the ruminal microbiota (Ibrahim *et al.*, 2021), their application for similar modulatory purposes in silage remains virtually unknown. The available literature is even more limited than that on essential oils (Table 2). Unlike essential oils, vegetable oils extracted from plant seeds have a high proportion of unsaturated fatty acids, mainly oleic, linoleic and α/γ -linolenic, whose fermentative modulation capacity through microbial inhibition has been reported in ruminal bacteria and protozoa (Ibrahim *et al.*, 2021).

Tab. 2: Compilation of studies on vegetable oils applied as silage additives and their main results

Reference	Silage ¹	Oil	Fatty acids	Application rate ²	Results ³
Jackson and Brown (1967)	Fescue (180 d)	Peanut	Oleic, Linoleic	49 g/kg DM	↓ pH, = CP, = Lac, = Ace, ↑ ME
Jackson (1971)	Ryegrass (120 d)	Peanut	Oleic, Linoleic	40 g/kg DM	= pH, = CP, ↓ NH_3 , = Lac, ↑ Ace, ↓ But, ↑ ME
Jackson and Anderson (1971)	Ryegrass (120 d)	Peanut	Oleic, Linoleic	40 g/kg DM	↓ SCFA, ↑ Ace, ↓ But
Besharati <i>et al.</i> (2020a)	Alfalfa (60 d)	Flaxseed	Linolenic, Linoleic	60 mg/kg DM	↓ pH, = NH_3 , ↑ CP, = SC, ↓ NDF, ↑ Lac
		Flaxseed	Linolenic, Linoleic	120 mg/kg DM	↓ pH, ↓ NH_3 , ↑ CP, ↑ SC, = NDF, ↑ Lac
Besharati <i>et al.</i> (2020b)	Alfalfa (60 d)	Flaxseed	Linolenic, Linoleic	60 mg/kg FM	↓ pH, = NH_3 , ↑ CP, = SC, ↓ NDF, ↑ Lac, ↑ AS

¹Forage + storage length in days.

²Application rates are presented in the units informed on each study.

³↑ = increase, ↓ = decrease, = = no effect, CP = crude protein, SC = soluble carbohydrates, SCFA = short chain fatty acids, NDF = neutral detergent insoluble fibre, Lac = lactic acid, Ace = acetic acid, But = butyric acid, AS = aerobic stability, ME = metabolizable energy

However, there is a considerable lack of information about microbial modulation by fatty acids in silage. Jackson and Brown (1967) reported no effect of treatment with peanut oil containing oleic (67.9%) and linoleic (22.5%) acids on the fermentation of fescue silage. On the other hand, Jackson (1971) and Jackson and Anderson (1971) reported significant changes in the fermentation of ryegrass silage treated with peanut oil. Although the production of organic acids was reduced, the treated silages presented reduced DM losses, lower extent of proteolysis and reduced presence of mold compared to the control silage. The application rates applied in these studies were about 40 g/kg of forage DM. Besharati *et al.* (2020a, 2020b) treated alfalfa silage with flaxseed oil, which is rich in linolenic and linoleic acids. Despite the considerably lower application rates of 60 and 120 mg/kg DM, a significant improvement in fermentation was observed, such as increased lactic acid production, enhanced silage acidification, reduced extent of proteolysis, and a marked improvement in aerobic stability compared to the control silage. To our knowledge, these are the only studies currently available in the literature that have applied fatty acids via vegetable oils in ensiled forages.

Unsaturated fatty acids can inhibit both Gram-positive and Gram-negative bacteria (Ivan *et al.*, 2013) through mechanisms related to changes in cell membrane permeability or the formation of a lipid film on the surface of the microorganism cell (Kodicek and Worden, 1945, Maia *et al.*, 2007). Since microbial species have variable susceptibility to these molecules, a selective pressure may occur within the microbiota (Maia *et al.*, 2007, 2010, Zhang *et al.*, 2008, Ivan *et al.*, 2013). As with essential oils, this selective pressure exerted by fatty acids may influence the classical acidification-based microbial control mechanism of silages, interfering with populations of LAB and undesirable microorganisms. Organic acid-based additives, for example, can improve nutrient conservation and aerobic stability of silages by exerting a non-selective antimicrobial effect on enterobacteria, yeasts, and LAB (Morais *et al.*, 2017).

Kodicek and Worden (1945) reported that *Lactobacillus helveticus* was inhibited when cultivated in a medium containing linoleic and linolenic acids at 16 µg/mL. Kankaanpää *et al.*

(2001) reported inhibition of strains of *Lactocaseibacillus rhamnosus*, *Lactobacillus delbrueckii* and *Lactocaseibacillus casei* when cultivated in a medium containing unsaturated fatty acids ranging from 18- to 22-carbon chain. Talon *et al.* (2000) also reported inhibition of five LAB species, including *Lactiplantibacillus plantarum*, with linoleic acid. Endo *et al.* (2006) observed inhibition of five LAB strains, including two from *L. plantarum* (JCM1149 and IFO15891) by α-linolenic, eicosapentaenoic (C20:5) and docosahexaenoic (C22:6) acids. However, *L. helveticus* was favored by saturated fatty acids, such as palmitic and stearic acids, and less inhibited by oleic acid in the work of Kodicek and Worden (1945). Endo *et al.* (2006) reported that all five LAB strains tested not only survived but also grew in medium containing oleic acid. Partanen *et al.* (2001) reported that *L. delbrueckii* incorporated oleic acid into its membranes, converting it to cyclopropane (C19) fatty acids. This process was also observed in *Lentilactobacillus buchneri* by Louesdon *et al.* (2014) and in *L. plantarum* strains by Wang *et al.* (2021). The addition of oleic acid to the culture medium may favor the growth of several LAB species (Johnsson *et al.*, 1995), and the conversion to cyclopropane fatty acids in the membrane appears to grant the bacteria the ability to survive under adverse conditions, such as freezing (Wang *et al.*, 2021) and low pH (Partanen *et al.*, 2001). Regarding clostridia, however, oleic acid appears to inhibit spore germination, an effect also observed with linoleic acid (Foster and Wynne, 1948). Skřivanová *et al.* (2005) reported inhibition of clostridia by linoleic acid, although the effect was more pronounced with lauric acid.

Fatty acids are not very susceptible to degradation in the anaerobic environment of silages due to the highly reduced nature of their carbon chains. Jackson and Anderson (1971) observed no evidence of chain breakage or biohydrogenation of unsaturated fatty acids after 120 days of storage. In addition to suppressing undesirable microorganisms after the early fermentative stage, the intact presence of exogenous fatty acids in the silage may enhance microbial control after silo opening, delaying the development of yeasts and spoilage bacteria and enhancing aerobic stability.

CONCLUSION

The available literature indicates that essential oils and fatty acids hold potential as natural silage additives due to their capacity to modulate microbial activity, suppress undesirable microorganisms and, in many cases, improve aerobic stability. However, the current evidence remains inconsistent across forage types, compounds and experimental conditions, reflecting the early stage of development of this approach. A major limitation of the existing research is the predominant use of crude essential oils with highly variable and often poorly characterised composition, which hampers comparisons among studies and obscures the mechanisms responsible for the observed effects. Future research should therefore prioritise the evaluation of individual bioactive molecules, or well-defined combinations of compounds, in order to identify molecule-specific effects on fermentation kinetics and microbial populations. In addition, the adoption of modern molecular approaches, including metagenomic and metabolomic techniques, would allow a more detailed understanding of microbial succession and metabolic shifts occurring in treated silages. Particular attention should also be given to the early fermentative stage, when microbial dynamics and acidification processes are most intense, through experimental designs that include temporal sampling throughout fermentation. For fatty acids, the scarcity of studies and the frequent reliance on whole seed oils highlight the need for targeted investigations into specific fatty acids and their mechanisms of action in silage ecosystems. Collectively, advancing this field will depend on moving from empirical testing of crude plant extracts towards mechanistically oriented studies capable of linking specific molecules to microbial modulation and fermentation outcomes in silage systems.

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APPENDIX SUPPLEMENTARY TABLES

Tab. S1: Chemical composition of alfalfa silage treated with essential oils

Reference	Storage length	Essential oil	Application rate	Δ% DM		Δ% CP		Δ% NDF		Δ% WSC	
Önenç and Turgud (2019)	60 d		Fresh forage	–		222.5 g/kg DM		446.6 g/kg DM			
			Control	254.2 g/kg FM		182.1 g/kg DM		416.8 g/kg DM		95.3 g/kg DM	
		Oregano	600 mg/kg FM	–0.1	ns	+1.5	*	+1.7	*	–5.6	*
		Cumin	600 mg/kg FM	+2.0	*	+3.5	*	+6.0	*	–23.1	*
		Cinnamon	600 mg/kg FM	+6.3	*	+3.1	*	+3.9	*	–58.0	*
Besharati and Niafifar (2020)	60 d		Fresh forage	222.0 g/kg FM		196.0 g/kg DM		248.0 g/kg DM		37.4 g/kg DM	
			Control	244.4 g/kg FM		116.2 g/kg DM		490.7 g/kg DM		40.8 g/kg DM	
		Lemonseed	60 mg/kg FM	–0.8	ns	+6.5	*	–2.1	ns	–35.5	*
			120 mg/kg FM	+5.6	*	+11.4	*	–4.5	ns	+20.3	*
Besharati <i>et al.</i> (2020a)	6 d		Fresh forage	222.0 g/kg FM		196.0 g/kg DM		248.0 g/kg DM		37.4 g/kg DM	
			Control	244.3 g/kg FM		116.2 g/kg DM		490.6 g/kg DM		40.8 g/kg DM	
		Flaxseed	60 mg/kg DM	+9.8	*	+5.2	*	–12.0	*	–0.7	ns
			120 mg/kg DM	+22.9	*	+6.4	*	–8.1	ns	+13.0	*
Besharati <i>et al.</i> (2020b)	60 d		Fresh forage	222.0 g/kg FM		196.0 g/kg DM		248.0 g/kg DM		37.4 g/kg DM	
			Control	244.4 g/kg FM		116.2 g/kg DM		490.7 g/kg DM		40.9 g/kg DM	
		Cinnamon	60 mg/kg FM	+5.1	*	+5.8	*	–13.5	*	+16.9	*
		Flaxseed	60 mg/kg FM	+9.7	*	+5.2	*	–12.0	*	–1.0	ns
		Lemonseed	60 mg/kg FM	+0.2	ns	+6.5	*	–2.1	ns	+13.2	*
		Mixture 1:1:1	180 mg/kg FM	–0.9	ns	+7.9	*	–19.3	*	+3.7	ns
Turan and Önenç (2018)	120 d		Fresh forage	–		239.0 g/kg DM		384.1 g/kg DM		–	
			Control	325.0 g/kg FM		230.6 g/kg DM		331.5 g/kg DM		–	
		Cumin	300 mg/kg FM	+2.4	ns	+6.3	*	–14.9	*	–	
			500 mg/kg FM	–0.2	ns	+2.2	*	–5.8	*	–	
Besharati <i>et al.</i> (2021b)	Alfalfa 90 d	Ricinoleic acid + cardol + cardanol	Fresh forage	350.0 g/kg FM		180.2 g/kg DM		418.0 g/kg DM		–	
			Control	292.9 g/kg FM		149.7 g/kg DM		380.9 g/kg DM		–	
			500 mg/kg FM	+4.8	ns	+12.1	*	–0.4	ns	–	
	3:1 Alfalfa/ apple pomace 90 d	Ricinoleic acid + cardol + cardanol	Fresh forage	352.5 g/kg FM		150.5 g/kg DM		414.6 g/kg DM		–	
			Control	297.0 g/kg FM		133.6 g/kg DM		407.1 g/kg DM		–	
			500 mg/kg FM	+3.4	ns	+14.3	*	–1.1	ns	–	
	1:1 Alfalfa/ apple pomace 90 d	Ricinoleic acid + cardol + cardanol	Fresh forage	355.0 g/kg FM		120.7 g/kg DM		411.1 g/kg DM		–	
			Control	294.0 g/kg FM		108.8 g/kg DM		410.5 g/kg DM		–	
			500 mg/kg FM	+9.2	ns	+5.4	*	+1.6	ns	–	
	1:3 Alfalfa/ apple pomace 90 d	Ricinoleic acid + cardol + cardanol	Fresh forage	357.5 g/kg FM		91.0 g/kg DM		407.7 g/kg DM		–	
			Control	271.0 g/kg FM		93.6 g/kg DM		408.7 g/kg DM		–	
			500 mg/kg FM	+17.7	*	–5.9	*	–0.7	ns	–	

Δ% – Percentage variation relative to the control mean; CP – crude protein; DM – dry matter; FM – fresh matter; NDF – neutral detergent insoluble fiber; WSC – water soluble carbohydrates; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$.

Tab. S2: Fermentative profile of alfalfa silage treated with essential oils

Reference	Storage length	Essential oil	Application rate	Δ% DM losses		Δ% pH		Δ% NH ₃ -N		Δ% Lactic acid	
Önenç and Turgud (2019)	60 d		Fresh forage	–		–		–		–	
			Control	19.1 g/kg		5.5		105.6 g/kg N		26.5 g/kg DM	
		Oregano	600 mg/kg FM	–16.8	*	–5.5	*	–6.0	ns	+111.7	*
		Cumin	600 mg/kg FM	–24.1	*	–4.2	*	–9.7	ns	+165.0	*
		Cinnamon	600 mg/kg FM	–27.2	*	–8.5	*	–27.3	*	+316.2	*
Besharati and Niazifar (2020)	60 d		Fresh forage	–		6.14		–		–	
			Control	–		4.66		84.93 g/kg N		69.4 g/kg DM	
		Lemonseed	60 mg/kg FM	–	–	–22.5	*	+0.3	ns	+9.9	*
			120 mg/kg FM	–	–	–18.7	*	0.0	ns	+14.5	*
Besharati <i>et al.</i> (2020a)	60 d		Fresh forage	–		6.14		–		–	
			Control	–		4.65		84.93 g/kg N		69.4 g/kg DM	
		Flaxseed	60 mg/kg DM	–	–	–19.4	*	–1.6	ns	+2.7	*
			120 mg/kg DM	–	–	–17.0	*	–14.3	*	+6.9	*
Besharati <i>et al.</i> (2020b)	60 d		Fresh forage	–		6.14		–		–	
			Control	–		4.65		84.93 g/kg N		69.4 g/kg DM	
		Cinnamon	60 mg/kg FM	–	–	–21.3	*	–6.0	*	+15.7	*
		Flaxseed	60 mg/kg FM	–	–	–19.8	*	–1.6	ns	+2.7	*
		Lemonseed	60 mg/kg FM	–	–	–22.4	*	+0.3	ns	+9.9	*
		Mixture 1:1:1	180 mg/kg FM	–	–	–18.5	*	–1.6	ns	+2.1	*
Turan and Önenç (2018)	120 d		Fresh forage	–		–		–		–	
			Control	–		4.88		–		–	
		Cumin	300 mg/kg FM	–	–	–2.3	*	–	–	–	–
			500 mg/kg FM	–	–	–10.5	*	–	–	–	–
Besharati <i>et al.</i> (2021b)	Alfalfa 90 d	Ricinoleic acid + cardol + cardanol	Fresh forage	–		8.4		–		–	
			Control	–		5.1		700.0 g/kg N		–	
			500 mg/kg FM	–	–	0.0	ns	+6.7	ns	–	–
	3:1 Alfalfa/ apple pomace 90 d	Ricinoleic acid + cardol + cardanol	Fresh forage	–		7.2		–		–	
			Control	–		4.2		490.0 g/kg N		–	
			500 mg/kg FM	–	–	0.0	ns	–12.2	ns	–	–
	1:1 Alfalfa/ apple pomace 90 d	Ricinoleic acid + cardol + cardanol	Fresh forage	–		6.0		–		–	
			Control	–		4.1		396.0 g/kg N		–	
			500 mg/kg FM	–	–	0.0	ns	–35.2	*	–	–
	1:3 Alfalfa/ apple pomace 90 d	Ricinoleic acid + cardol + cardanol	Fresh forage	–		4.7		407.7 g/kg DM		–	
			Control	–		3.8		408.7 g/kg DM		–	
			500 mg/kg FM	–	–	–2.6	ns	–0.7	ns	–	–

Δ% – Percentage variation relative to the control mean; CP – crude protein; DM – dry matter; FM – fresh matter; NDF – neutral detergent insoluble fiber; WSC – water soluble carbohydrates; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$.

Tab. S3: Microbial counts of alfalfa silage treated with essential oils

Reference	Storage length	Essential oil	Application rate	$\Delta\%$ Yeasts (log cfu/g)		$\Delta\%$ Molds (log cfu/g)		$\Delta\%$ Enterobacteria (log cfu/g)		$\Delta\%$ LAB (log cfu/g)	
Önenç and Turgud (2019)	60 d		Fresh forage	–		–		–		–	
			Control	5.52		5.31		–		4.87	
		Oregano	600 mg/kg FM	-40.9	*	-15.4	*	–	–	+7.8	*
		Cumin	600 mg/kg FM	-92.0	*	-15.1	*	–	–	+10.1	*
		Cinnamon	600 mg/kg FM	-87.9	*	-14.3	*	–	–	+28.7	*
Turan and Önenç (2018)	120 d		Fresh forage	–		–		–		–	
			Control	4.86		2.49		1.01		3.51	
		Cumin	300 mg/kg FM	-29.4	*	-100.0	*	-100.0	*	+76.1	*
			500 mg/kg FM	-22.6	*	-100.0	*	-100.0	*	+108.3	*

$\Delta\%$ – Percentage variation relative to the control mean; LAB – lactic acid bacteria; FM – fresh matter; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$

Tab. S4: Aerobic stability of alfalfa silage treated with essential oils

Reference	Storage length	Essential oil	Application rate	$\Delta\%$ Aerobic stability	
Besherati et al. (2020b)	60 d		Control	77 h	
		Cinnamon	60 mg/kg FM	+29.0	*
		Flaxseed	60 mg/kg FM	+19.5	*
		Lemonseed	60 mg/kg FM	+25.5	*
		Mixture 1:1:1	180 mg/kg FM	+45.5	*

$\Delta\%$ – Percentage variation relative to the control mean; LAB – lactic acid bacteria; FM – fresh matter; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$

Tab. S5: Chemical composition of whole plant corn silage treated with essential oils.

Reference	Storage length	Essential oil	Application rate	$\Delta\%$ DM		$\Delta\%$ CP		$\Delta\%$ NDF		$\Delta\%$ WSC	
Önenç and Turgud (2019)	256 d	Blend (Thymol + Eugenol + Vanillin + Limonene)	Fresh forage	289.9 g/kg FM		–		–		67.9 g/kg DM	
			Control	261.0 g/kg FM		–		–		23.4 g/kg DM	
			40 mg/kg FM	+1.1	ns	–	–	–	–	+14.1	ns
			80 mg/kg FM	-1.1	ns	–	–	–	–	-6.4	ns
Silva et al. (2024a)	60 d	Sweet orange	Fresh forage	323.2 g/kg FM		54.7 g/kg DM		554.0 g/kg DM		–	
			Control	293.4 g/kg FM		60.5 g/kg DM		546.6 g/kg DM		–	
			200 mg/kg FM	+2.3	*Q	+1.1	ns	+1.9	ns	–	–
			400 mg/kg FM	+5.8	*Q	+0.8	ns	-0.6	ns	–	–
			600 mg/kg FM	+3.5	*Q	+3.6	ns	-2.1	ns	–	–
Silva et al. (2024b)	60 d	Blend (<i>Eucalyptus staigeriana</i> : <i>Copaifera langsdorffii</i>) 500 mg/kg FM	Fresh forage	323 g/kg FM		54.7 g/kg DM		554 g/kg DM		–	
			Control	293 g/kg FM		60.5 g/kg DM		547 g/kg DM		–	
			1 : 3	+4.8	*	+4.6	ns	+0.7	ns	–	–
			1 : 1	+6.8	*	+0.5	ns	-5.1	ns	–	–
			3 : 1	+5.8	*	-3.8	ns	+0.5	ns	–	–

Reference	Storage length	Essential oil	Application rate	$\Delta\%$ DM		$\Delta\%$ CP		$\Delta\%$ NDF		$\Delta\%$ WSC	
Hodjatpanah–Montazeri <i>et al.</i> (2016)	45 d		Control	296.8 g/kg FM		74.7 g/kg DM		574.0 g/kg DM		–	
		Cinnamon	120 mg/kg DM	+12.9	*	+0.9	ns	+0.5	ns	–	–
			240 mg/kg DM	+8.1	*	+0.9	ns	–3.7	*	–	–
		Thyme	120 mg/kg DM	+5.0	ns	+0.7	ns	–4.2	*	–	–
			240 mg/kg DM	+1.0	ns	+2.4	ns	–0.7	ns	–	–
		Mint	120 mg/kg DM	–4.6	ns	+30.7	*	+10.1	*	–	–
			240 mg/kg DM	–4.3	ns	+34.5	*	+1.0	ns	–	–
		Oregano	120 mg/kg DM	–11.9	*	+21.3	*	+0.7	ns	–	–
			240 mg/kg DM	+2.7	ns	+37.3	*	+4.9	*	–	–
		Cumin	120 mg/kg DM	–0.5	ns	–4.0	ns	+1.2	ns	–	–
			240 mg/kg DM	+5.1	ns	+4.3	ns	–5.6	*	–	–
Maghsoudloo <i>et al.</i> (2017)	3 d		Control	295 g/kg FM		78.7 g/kg DM		560 g/kg DM		169.2 g/kg DM	
		Rosemary	125 μ L/kg FM	+2.5	ns	0.0	ns	0.0	ns	+46.8	*
			250 μ L/kg FM	+2.6	ns	+2.3	*	+7.1	*	+46.7	*
		Fennel	125 μ L/kg FM	+2.0	ns	–1.5	*	+18.4	*	+72.0	*
			250 μ L/kg FM	+4.6	ns	0.0	ns	–3.6	ns	+9.3	*
		<i>Carum copticum</i>	125 μ L/kg FM	+3.3	ns	0.0	ns	–1.8	ns	+8.1	*
			250 μ L/kg FM	+2.8	ns	0.0	ns	–5.4	ns	–1.9	*
Maghsoudloo <i>et al.</i> (2017)	3 d		Control	296.3 g/kg FM		78.7 g/kg DM		683.3 g/kg DM		156 g/kg DM	
		Rosemary	125 μ L/kg FM	+2.5	ns	0.0	ns	0.0	ns	+46.8	*
			250 μ L/kg FM	+2.6	ns	+2.3	*	+7.1	*	+46.7	*
		Fennel	125 μ L/kg FM	+2.0	ns	–1.5	*	+18.4	*	+72.0	*
			250 μ L/kg FM	+4.6	ns	0.0	ns	–3.6	ns	+9.3	*
		<i>Carum copticum</i>	125 μ L/kg FM	+3.3	ns	0.0	ns	–1.8	ns	+8.1	*
			250 μ L/kg FM	+2.8	ns	0.0	ns	–5.4	ns	–1.9	*
Maghsoudloo <i>et al.</i> (2017)	7 d		Control	296.3 g/kg FM		78.7 g/kg DM		683.3 g/kg DM		156 g/kg DM	
		Rosemary	125 μ L/kg FM	+0.8	ns	0.0	ns	–3.4	ns	+31.0	*
			250 μ L/kg FM	+1.6	ns	+11.2	*	+6.8	*	+41.0	*
		Fennel	125 μ L/kg FM	+1.5	ns	+4.4	*	–3.4	ns	+63.1	*
			250 μ L/kg FM	+4.0	ns	+2.3	*	–3.4	ns	+12.3	*
		<i>Carum copticum</i>	125 μ L/kg FM	+2.4	ns	+8.9	*	–9.3	*	–19.6	*
			250 μ L/kg FM	+2.1	ns	+6.7	*	–0.5	ns	+1.3	*
Maghsoudloo <i>et al.</i> (2017)	21 d		Control	300.1 g/kg FM		84 g/kg DM		613.3 g/kg DM		125.2 g/kg DM	
		Rosemary	125 μ L/kg FM	–0.3	ns	–2.1	*	+10.9	*	+17.0	*
			250 μ L/kg FM	–0.2	ns	+2.0	*	+1.1	ns	+67.9	*
		Fennel	125 μ L/kg FM	+0.1	ns	+2.0	*	+19.0	*	+63.2	*
			250 μ L/kg FM	+1.5	ns	–2.1	*	+3.3	ns	+18.6	*
		<i>Carum copticum</i>	125 μ L/kg FM	+1.1	ns	–2.1	*	–1.7	ns	–20.0	*
			250 μ L/kg FM	–0.7	ns	+2.0	*	+1.1	ns	+30.0	*

Reference	Storage length	Essential oil	Application rate	Δ% DM		Δ% CP		Δ% NDF		Δ% WSC	
Maghsoudloo et al. (2017)	45 d		Control	295.4 g/kg FM		85.7 g/kg DM		650 g/kg DM		90.5 g/kg DM	
		Rosemary	125 µL/kg FM	+0.9	ns	-2.0	*	-6.2	*	+18.9	*
			250 µL/kg FM	+0.5	ns	-4.1	*	-3.1	ns	+90.4	*
		Fennel	125 µL/kg FM	+0.8	ns	-6.1	*	-1.5	ns	+99.0	*
			250 µL/kg FM	+1.3	ns	-2.0	*	-13.8	*	+2.4	*
		<i>Carum copticum</i>	125 µL/kg FM	-0.1	ns	-12.3	*	0.0	ns	+9.5	*
			250 µL/kg FM	+2.2	*	+4.1	*	0.0	ns	+66.6	*

Δ% – Percentage variation relative to the control mean; CP – crude protein; DM – dry matter; FM – fresh matter; NDF – neutral detergent insoluble fiber; WSC – water soluble carbohydrates; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$.

Tab. S6: Fermentative profile of whole plant corn silage treated with essential oils

Reference	Storage length	Essential oil	Application rate	Δ% DM losses		Δ% pH		Δ% NH ₃ -N		Δ% Lactic acid		Δ% Acetic acid	
Kung Jr. et al. (2018)	256 d	Blend (Thymol + Eugenol + Vanillin + Limonene)	Fresh forage	–		5.38		0.82 g/kg DM		–		–	
			Control	101.0 g/kg		3.72		2.29 g/kg DM		55.0 g/kg DM		22.0 g/kg DM	
			40 mg/kg FM	+34.7	ns	-0.3	ns	-1.3	ns	-2.9	ns	-10.0	ns
			80 mg/kg FM	+12.9	ns	-1.6	ns	+8.3	ns	+3.6	ns	-2.7	ns
Silva et al. (2024a)	60 d	Sweet orange	Fresh forage	–		–		–		–		–	
			Control	102.5 g/kg		3.62		0.94 g/kg CP		–		28.0 g/kg DM	
			200 mg/kg FM	+1.3	*Q	-1.9	ns	-3.2	ns	–	–	+79.1	*Q
			400 mg/kg FM	-42.8	*Q	-0.8	ns	-7.4	ns	–	–	-28.3	*Q
			600 mg/kg FM	-26.8	*Q	-2.2	ns	-10.6	ns	–	–	+277.3	*Q
Silva et al. (2024b)	60 d	Blend (<i>Eucalyptus staigeriana</i> : <i>Copaifera langsdorffii</i>)	Control	44 g/kg		3.6		7.3 g/kg CP		–		28 g/kg DM	
			1:3	-2.3	ns	-2.8	ns	-8.2	ns	–	–	+118.2	*
			1:1	-2.3	ns	-2.8	ns	-13.7	ns	–	–	+133.2	*
			3:1	-6.8	ns	-5.6	ns	-13.7	ns	–	–	+51.1	*
Hodjatpanah-Montazeri et al. (2016)	45 d	Cinnamon	Control	–		3.75		0.79 mg/dL		–		–	
			120 mg/kg DM	–	–	+0.5	ns	+3.8	ns	–	–	–	–
			240 mg/kg DM	–	–	+1.6	*	-3.8	ns	–	–	–	–
		Thyme	120 mg/kg DM	–	–	-0.5	ns	-11.4	*	–	–	–	–
			240 mg/kg DM	–	–	+0.3	ns	-22.8	*	–	–	–	–
		Mint	120 mg/kg DM	–	–	+7.5	*	+13.9	*	–	–	–	–
			240 mg/kg DM	–	–	-3.2	*	+5.1	ns	–	–	–	–
		Oregano	120 mg/kg DM	–	–	+8.3	*	+34.2	*	–	–	–	–
			240 mg/kg DM	–	–	+10.1	*	+10.1	*	–	–	–	–
		Cumin	120 mg/kg DM	–	–	+3.7	*	0.0	ns	–	–	–	–
			240 mg/kg DM	–	–	+2.9	*	+25.3	*	–	–	–	–
Maghsoudloo et al. (2017)	3 d	Rosemary	Control	–		3.72		3.37 mg/dL		–		–	
			125 µL/kg FM	–	–	-0.8	ns	-3.6	ns	–	–	–	–
			250 µL/kg FM	–	–	0.0	ns	-14.2	*	–	–	–	–
		Fennel	125 µL/kg FM	–	–	-1.1	ns	-6.2	*	–	–	–	–
			250 µL/kg FM	–	–	-0.8	ns	-7.7	*	–	–	–	–
		<i>Carum copticum</i>	125 µL/kg FM	–	–	-0.8	ns	-5.3	*	–	–	–	–
			250 µL/kg FM	–	–	0.0	ns	-8.6	*	–	–	–	–

Reference	Storage length	Essential oil	Application rate	Δ% DM losses		Δ% pH		Δ% NH ₃ -N		Δ% Lactic acid		Δ% Acetic acid	
Maghsoudloo et al. (2017)	7 d	Rosemary	Control	–		3.54		2.99 mg/dL		–		–	
			125 µL/kg FM	–	–	+0.6	ns	+10.7	ns	–	–	–	–
		Fennel	250 µL/kg FM	–	–	+0.6	ns	+24.7	*	–	–	–	–
			125 µL/kg FM	–	–	–0.3	ns	+22.4	*	–	–	–	–
		<i>Carum copticum</i>	250 µL/kg FM	–	–	+0.3	ns	+16.4	ns	–	–	–	–
			125 µL/kg FM	–	–	–0.3	ns	+15.4	ns	–	–	–	–
Maghsoudloo et al. (2017)	21 d	Rosemary	Control	–		3.56		2.85 mg/dL		–		–	
			125 µL/kg FM	–	–	+0.6	ns	–3.2	ns	–	–	–	–
		Fennel	250 µL/kg FM	–	–	+0.6	ns	–4.2	ns	–	–	–	–
			125 µL/kg FM	–	–	+0.3	ns	–9.8	ns	–	–	–	–
		<i>Carum copticum</i>	250 µL/kg FM	–	–	+0.3	ns	–15.8	ns	–	–	–	–
			125 µL/kg FM	–	–	0.0	ns	–16.8	ns	–	–	–	–
Maghsoudloo et al. (2017)	45 d	Rosemary	Control	–		3.41		2.93 mg/dL		–		–	
			125 µL/kg FM	–	–	+0.3	ns	–8.5	ns	–	–	–	–
		Fennel	250 µL/kg FM	–	–	+0.9	ns	–8.5	ns	–	–	–	–
			125 µL/kg FM	–	–	+0.3	ns	–3.4	ns	–	–	–	–
		<i>Carum copticum</i>	250 µL/kg FM	–	–	+1.8	ns	–9.6	ns	–	–	–	–
			125 µL/kg FM	–	–	+0.6	ns	–9.6	ns	–	–	–	–

Δ% – Percentage variation relative to the control mean; DM – dry matter; FM – fresh matter; NH₃-N – ammonia nitrogen; * – significant at P < 0.05; ns – not significant at P < 0.05.

Tab. S7: Microbial counts of whole plant corn silage treated with essential oils.

Reference	Storage length	Essential oil	Application rate	Δ% Yeasts (log cfu/g)		Δ% Molds (log cfu/g)		Δ% Enterobacteria (log cfu/g)		Δ% LAB (log cfu/g)	
Kung Jr. et al. (2018)	256 d	Blend (Thymol + Eugenol + Vanillin + Limonene)	Fresh forage	7.04		6.16		7.65		7.40	
			Control	5.16		4.47		–		–	
			40 mg/kg FM	+4.8	ns	+2.2	ns	–	ns	–	ns
			80 mg/kg FM	+0.4	ns	+0.9	ns	–	ns	–	ns
Silva et al. (2024a)	60 d		Fresh forage	–		–		–		–	
			Control	–		2.55		–		2.29	
		Sweet orange	200 mg/kg FM	–	–	+83.1	ns	–	–	–29.7	ns
			400 mg/kg FM	–	–	+12.5	ns	–	–	–3.9	ns
			600 mg/kg FM	–	–	–52.2	ns	–	–	–41.5	ns
Silva et al. (2024b)	60 d	Blend (<i>Eucalyptus staigeriana</i> : <i>Copaifera langsdorffii</i>)	Control	–		3.03		–		2.77	
			1 : 3	–	–	–89.1	*	–	–	–28.9	ns
			1 : 1	–	–	+43.6	*	–	–	+24.9	ns
			3 : 1	–	–	+27.4	*	–	–	–2.5	ns

Δ% – Percentage variation relative to the control mean; LAB – lactic acid bacteria; FM – fresh matter; * – significant at P < 0.05; ns – not significant at P < 0.05

Tab. S8: Aerobic stability of whole plant corn silage treated with essential oils

Reference	Storage length	Essential oil	Application rate	$\Delta\%$ Aerobic stability	
Kung Jr. <i>et al.</i> (2018)	256 d	Blend (Thymol + Eugenol + Vanillin + Limonene)	Fresh forage	–	
			Control	47.5 h	
			40 mg/kg FM	–4.6	ns
			80 mg/kg FM	+4.2	ns
Silva <i>et al.</i> (2024a)	60 d	Sweet orange	Fresh forage	–	
			Control	36 h	
			200 mg/kg FM	0.0	ns
			400 mg/kg FM	+16.7	ns
			600 mg/kg FM	+66.7	*
Silva <i>et al.</i> (2024b)	60 d	Blend (<i>Eucalyptus staigeriana</i> : <i>Copaifera langsdorffii</i>)	Control	36 h	
			1:3	+66.7	*
			1:1	+83.3	*
			3:1	+66.7	*
Hodjatpanah–Montazeri <i>et al.</i> (2016)	45 d	Cinnamon	Control	72 h	
			120 mg/kg DM	+129.6	*
			240 mg/kg DM	+300.0	*
		Thyme	120 mg/kg DM	+300.0	*
			240 mg/kg DM	+44.4	*
		Mint	120 mg/kg DM	–20.8	ns
			240 mg/kg DM	–3.7	ns
		Oregano	120 mg/kg DM	+300.0	*
			240 mg/kg DM	+300.0	*
		Cumin	120 mg/kg DM	+300.0	*
				+300.0	*

$\Delta\%$ – Percentage variation relative to the control mean; LAB – lactic acid bacteria; FM – fresh matter; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$

Tab. S9: Chemical composition of other silages treated with essential oils

Reference	Silage	Essential oil	Application rate	$\Delta\%$ DM		$\Delta\%$ CP		$\Delta\%$ NDF		$\Delta\%$ WSC	
Cantoia Jr. <i>et al.</i> (2020)	Sugarcane 60 d		Fresh forage	249.0 g/kg FM		27.0 g/kg DM		542.0 g/kg DM		–	
			Control	222.0 g/kg FM		21.4 g/kg DM		621.0 g/kg DM		129 g/kg DM	
		Lemongrass	1 mL/kg FM	+14.4	*Q	+22.0	*Q	–15.6	*Q	+27.1	*Q
			2 mL/kg FM	+20.7	*Q	+16.8	*Q	–20.1	*Q	+40.3	*Q
			3 mL/kg FM	+14.4	*Q	+29.2	*Q	–15.3	*Q	+29.5	*Q
Zhou <i>et al.</i> (2024)	<i>Stylosanthes guianensis</i> 30 d		Fresh forage	363.0 g/kg FM		99.0 g/kg DM		701.0 g/kg DM		18.1 g/kg DM	
			Control	365.0 g/kg FM		85.0 g/kg DM		–		–	
		<i>Neolamarkia cadamba</i> leaf	5 g/kg FM	+1.9	ns	–0.1	ns	–	–	–	–
			10 g/kg FM	+2.7	ns	–5.9	ns	–	–	–	–
	Rice straw 30 d		Fresh forage	279.0 g/kg FM		72.6 g/kg DM		611.0 g/kg DM		21.6 g/kg DM	
			Control	271.0 g/kg FM		82.8 g/kg DM		–		–	
		<i>Neolamarkia cadamba</i> leaf	5 g/kg FM	+10.7	ns	–11.2	*	–	–	–	–
			10 g/kg FM	+9.6	ns	–8.7	*	–	–	–	–

Reference	Silage	Essential oil	Application rate	$\Delta\%$ DM		$\Delta\%$ CP		$\Delta\%$ NDF		$\Delta\%$ WSC	
Li <i>et al.</i> (2022)	Paper mulberry 60 d		Fresh forage	288.7 g/kg FM		156.7 g/kg DM		667.0 g/kg DM		20.7 g/kg DM	
			Control	258.3 g/kg FM		149.8 g/kg DM		312.1 g/kg DM		7.1 g/kg DM	
		<i>Amomum villosum</i>	1 mL/kg FM	+2.5	ns	+3.5	ns	+27.8	*	+63.4	*
Aniceto <i>et al.</i> (2024)	Whole plant sorghum 60 d		Fresh forage	363.8 g/kg FM		47.8 g/kg DM		685.3 g/kg DM		–	
			Control	345.0 g/kg FM		42.3 g/kg DM		726.2 g/kg DM		–	
		Rosemary (Ros)	1 g/kg FM	+5.9	ns	+26.5	*	–7.7	ns	–	–
		Tea tree (TT)	1 g/kg FM	+12.2	ns	+13.0	*	–5.3	ns	–	–
		Citronella (Cit)	1 g/kg FM	+17.2	ns	+16.2	*	–3.4	ns	–	–
		Ros:TT 1:1	1 g/kg FM	+6.6	ns	+18.5	*	+1.7	ns	–	–
		Ros:Cit 1:1	1 g/kg FM	+9.8	ns	+20.6	*	–2.3	ns	–	–
		TT:Cit 1:1	1 g/kg FM	–3.9	ns	+9.8	ns	+1.0	ns	–	–
		Ros:TT:Cit 1:1:1	1 g/kg FM	+2.3	ns	+17.3	*	–0.4	ns	–	–
Chaves <i>et al.</i> (2012)	Whole-plant barley 365 d		Control	349.3 g/kg FM		117.4 g/kg DM		487.1 g/kg DM		40.9 g/kg DM	
		Cinnamon leaf	125 mg/kg FM	+0.3	ns	+4.3	*	–4.7	ns	+33.3	*
			250 mg/kg FM	–4.5	ns	+5.6	*	–3.8	ns	+46.2	*
			400 mg/kg FM	–0.3	ns	+6.0	*	+5.1	ns	+0.5	ns
		Oregano	125 mg/kg FM	–1.8	ns	+0.3	ns	+1.8	ns	–19.6	ns
			250 mg/kg FM	–3.8	ns	+0.9	ns	+3.0	ns	+24.0	ns
			400 mg/kg FM	–1.5	ns	+3.3	ns	+1.2	ns	+13.4	ns
		Sweet orange	125 mg/kg FM	–1.8	ns	+5.4	*	+2.6	ns	–2.7	ns
			250 mg/kg FM	–3.5	ns	+5.4	*	+2.5	ns	–23.7	ns
			400 mg/kg FM	–4.4	ns	+10.2	*	+3.9	ns	+21.3	ns
Foskolos <i>et al.</i> (2016)	Ryegrass 35 d		Control	–		38.8 g/kg DM		–		–	
		Eugenol	50 mg/kg FM	–	–	+3.4	ns	–	–	–	–
			500 mg/kg FM	–	–	+0.5	ns	–	–	–	–
			2000 mg/kg FM	–	–	–2.3	ns	–	–	–	–
			Control	–		48.9 g/kg DM		–		–	
		Thymol	50 mg/kg FM	–	–	+1.8	ns	–	–	–	–
			500 mg/kg FM	–	–	–3.9	ns	–	–	–	–
			2000 mg/kg FM	–	–	–12.5	ns	–	–	–	–
			Control	–		42.0 g/kg DM		–		–	
		Cinnamaldehyde	50 mg/kg FM	–	–	–2.9	ns	–	–	–	–
			500 mg/kg FM	–	–	–5.7	ns	–	–	–	–
			2000 mg/kg FM	–	–	–4.0	ns	–	–	–	–
			Control	–		38.6 g/kg DM		–		–	
		Carvacrol	50 mg/kg FM	–	–	+2.6	ns	–	–	–	–
			500 mg/kg FM	–	–	–4.1	ns	–	–	–	–
			2000 mg/kg FM	–	–	–2.1	ns	–	–	–	–
Önenç <i>et al.</i> (2015)	Field pea 60 d		Fresh forage	311.7 g/kg FM		233.7 g/kg DM		399.5 g/kg DM		38.0 g/kg DM	
			Control	235.3 g/kg FM		223.4 g/kg DM		375.6 g/kg DM		11.5 g/kg DM	
		Oregano	400 mg/kg FM	–3.8	*	+4.3	*	+4.4	*	+33.0	*
		Cinnamom	400 mg/kg FM	+8.3	*	+8.6	*	+3.4	*	–18.6	*
		Mixture 1:1	400 mg/kg FM	+2.0	*	–3.2	*	+4.0	*	+4.7	ns

Reference	Silage	Essential oil	Application rate	Δ% DM		Δ% CP		Δ% NDF		Δ% WSC	
Önenç et al. (2017)	Field pea 120 d		Fresh forage	311.7 g/kg FM		233.7 g/kg DM		399.5 g/kg DM		38.0 g/kg DM	
			Control	227.9 g/kg FM		226.0 g/kg DM		376.7 g/kg DM		6.9 g/kg DM	
		Oregano	400 mg/kg FM	+10.0	*	-3.1	*	+8.1	*	+7.3	*
		Cinnamom	400 mg/kg FM	+12.7	*	-3.3	*	+10.2	*	+33.7	*
		Mixture 1:1	400 mg/kg FM	+9.8	*	-8.9	*	+8.8	*	+22.6	*
Akinci and Önenç (2021)	Vetch + Oat 70 d		Fresh forage	312.4 g/kg FM		96.1 g/kg DM		607.6 g/kg DM		40.7 g/kg DM	
			Control	282.2 g/kg FM		97.3 g/kg DM		642.5 g/kg DM		14.1 g/kg DM	
		Cumin	200 mg/kg FM	-0.1	ns	+3.2	*	+1.0	*	-35.4	*
			300 mg/kg FM	+7.8	*	+5.3	*	-1.3	*	-15.8	*
			500 mg/kg FM	+4.9	*	+5.3	*	-0.8	*	-20.5	*
Duru (2024)	Vetch + Wheat 75 d		Fresh forage	369.2 g/kg FM		137.2 g/kg DM		384.0 g/kg DM		-	
			Control	384.6 g/kg FM		137.4 g/kg DM		356.8 g/kg DM		-	
		<i>Hypericum perforatum</i>	20 mg/kg	+4.9	*	-1.5	ns	+1.6	ns	-	-
			40 mg/kg	+2.9	*	-6.9	ns	+4.3	ns	-	-
			80 mg/kg	-0.9	ns	-4.4	ns	+11.0	*	-	-
			160 mg/kg	-1.7	ns	-4.9	ns	+9.3	*	-	-
Xu et al. (2023)	<i>Broussonetia papyfera</i> 3 d	<i>Amomum villosum</i>	Control	263.3 g/kg FM		152.6 g/kg DM		661.6 g/kg DM		7.4 g/kg DM	
			1 mL/kg FM	+3.0	ns	+3.7	ns	-2.2	ns	+4.1	ns
	<i>B. papyfera</i> : <i>P. sinese</i> 7:3 3 d		Control	232.7 g/kg FM		149.8 g/kg DM		587.0 g/kg DM		12.6 g/kg DM	
			1 mL/kg FM	+2.4	*	+5.1	ns	-4.3	*	+15.9	*
	<i>B. papyfera</i> : <i>P. sinese</i> 1:1 3 d		Control	210.8 g/kg FM		140.5 g/kg DM		553.9 g/kg DM		15.1 g/kg DM	
			1 mL/kg FM	+2.4	ns	+11.1	*	-9.8	*	+2.6	ns
	<i>B. papyfera</i> : <i>P. sinese</i> 3:7 3 d		Control	182.4 g/kg FM		128.9 g/kg DM		473.7 g/kg DM		10.8 g/kg DM	
			1 mL/kg FM	+4.8	*	+14.0	*	-14.0	*	+26.9	*
Xu et al. (2023)	<i>Pennisetum sinese</i> 3 d	<i>Amomum villosum</i>	Control	142.1 g/kg FM		115.1 g/kg DM		417.8 g/kg DM		15.2 g/kg DM	
			1 mL/kg FM	+10.9	*	+5.2	ns	-7.7	*	-5.9	ns
	<i>Broussonetia papyfera</i> 30 d		Control	258.3 g/kg FM		152.3 g/kg DM		613.3 g/kg DM		5.6 g/kg DM	
			1 mL/kg FM	+2.5	ns	+1.8	ns	-2.4	ns	-1.8	ns
	<i>B. papyfera</i> : <i>P. sinese</i> 7:3 30 d		Control	205.0 g/kg FM		147.0 g/kg DM		523.2 g/kg DM		10.8 g/kg DM	
			1 mL/kg FM	+9.5	*	+2.4	ns	-5.1	*	+26.9	*
	<i>B. papyfera</i> : <i>P. sinese</i> 1:1 30 d		Control	187.2 g/kg FM		132.6 g/kg DM		509.3 g/kg DM		12.6 g/kg DM	
			1 mL/kg FM	+6.1	*	+5.2	*	-16.1	*	+18.3	*
Andrade et al. (2024)	TRM 90 d		Fresh TMR	624.5 g/kg FM		181.0 g/kg DM		305.4 g/kg DM		-	
			Control	637.8 g/kg FM		165.8 g/kg DM		282.0 g/kg DM		-	
		Monensin	35 mg/kg DM	-2.9	*	+8.6	ns	+9.4	ns	-	-
			45 mg/kg DM	-2.0	*	+6.2	ns	+4.8	ns	-	-
		Limonene-rich essential oil	300 mg/kg DM	-2.6	*	+15.6	ns	+12.3	ns	-	-
			600 mg/kg DM	-2.9	*	+15.7	ns	+15.0	ns	-	-

Reference	Silage	Essential oil	Application rate	Δ% DM		Δ% CP		Δ% NDF		Δ% WSC	
Andrade <i>et al.</i> (2024)	TRM 90 d		Fresh TMR	410.0 g/kg FM		164.2 g/kg DM		395.3 g/kg DM		–	
			Control	362.8 g/kg FM		156.2 g/kg DM		412.1 g/kg DM		–	
		Monensin	35 mg/kg DM	+2.1	ns	+4.2	ns	–7.2	*	–	
			45 mg/kg DM	+0.2	ns	+7.9	ns	–7.8	*	–	
		Limonene-rich essential oil	300 mg/kg DM	–2.1	ns	+8.6	ns	–8.6	*	–	
			600 mg/kg DM	–2.0	ns	+5.2	ns	–10.0	*	–	

Δ% – Percentage variation relative to the control mean; CP – crude protein; DM – dry matter; FM – fresh matter; NDF – neutral detergent insoluble fiber; WSC – water soluble carbohydrate; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$; Q – quadratic effect

Tab. S10: Fermentative profile of whole plant corn silage treated with essential oils

Reference	Silage	Essential oil	Application rate	Δ% DM losses		Δ% pH		Δ% NH ₃ -N		Δ% Lactic acid		Δ% Acetic acid	
Cantoia Jr. <i>et al.</i> (2020)	Sugarcane 60 d	Lemongrass	Fresh forage	–		–		–		–		–	
			Control	311.0 g/kg		3.89		146.0 g/kg N		23.3 g/kg DM		14.4 g/kg DM	
			1 mL/kg FM	–37.9	*Q	–2.1	*Q	+1.4	*L	–10.3	*L	–40.1	*Q
			2 mL/kg FM	–51.8	*Q	–1.3	*Q	+23.3	*L	–27.9	*L	–49.6	*Q
			3 mL/kg FM	–32.8	*Q	3.6	*Q	+41.1	*L	–43.8	*L	–33.3	*Q
Zhou <i>et al.</i> (2024)	<i>Stylosanthes guianensis</i> 30 d	<i>Neolamarkia cadamba</i> leaf	Control	–		5.66		86.8 g/kg N		5.29 g/kg DM		3.45 g/kg DM	
			5 g/kg FM	–	–	–5.3	*	–37.3	*	+54.8	*	ND	*
			10 g/kg FM	–	–	–9.4	*	–57.9	*	+100.4	*	ND	*
	Rice straw 30 d	<i>Neolamarkia cadamba</i> leaf	Control	–		5.18		98.2 g/kg N		4.44 g/kg DM		3.23 g/kg DM	
			5 g/kg FM	–	–	–14.5	*	–59.1	*	+74.8	*	+22.3	*
			10 g/kg FM	–	–	–14.5	*	–64.6	*	+95.7	*	+42.1	*
Aniceto <i>et al.</i> (2024)	Whole plant sorghum 60 d		Control	64.1 g/kg		4.41		0.65 g/kg CP		35.1 g/kg DM		58.0 g/kg DM	
		Rosemary (Ros)	1 g/kg FM	–9.8	ns	+3.4	ns	+36.9	ns	+86.3	ns	–5.0	ns
		Tea tree (TT)	1 g/kg FM	–10.0	ns	+4.3	ns	+4.6	ns	+15.9	ns	–77.2	*
		Citronella (Cit)	1 g/kg FM	–27.8	*	+2.3	ns	–10.8	ns	–39.3	ns	–50.5	*
		Ros:TT 1:1	1 g/kg FM	–2.3	ns	+0.2	ns	+23.1	ns	+75.9	ns	–37.4	*
		Ros:Cit 1:1	1 g/kg FM	–19.7	ns	+3.6	ns	–9.2	ns	+6.4	ns	–34.6	*
		TT:Cit 1:1	1 g/kg FM	–26.5	*	–0.5	ns	+3.1	ns	–10.5	ns	–79.5	*
		Ros:TT:Cit 1:1:1	1 g/kg FM	–25.3	ns	+0.5	ns	+13.8	ns	+19.7	ns	–34.5	*
Chaves <i>et al.</i> (2012)	Whole plant barley 365 d		Control	–		4.10		1.20 g/kg DM		69.8 g/kg DM		16.8 g/kg DM	
		Cinnamon leaf	125 mg/kg FM	–	–	–2.4	ns	0.0	ns	+10.6	ns	–1.8	ns
			250 mg/kg FM	–	–	–2.4	ns	0.0	ns	+19.1	ns	–23.8	*
			400 mg/kg FM	–	–	0.0	ns	+8.3	ns	+12.8	ns	+2.4	ns
		Oregano	125 mg/kg FM	–	–	0.0	ns	+8.3	ns	+2.6	ns	+7.1	ns
			250 mg/kg FM	–	–	–2.4	ns	–8.3	ns	+5.6	ns	–14.3	ns
			400 mg/kg FM	–	–	–2.4	ns	0.0	ns	+7.2	ns	–1.2	ns
		Sweet orange	125 mg/kg FM	–	–	–2.4	ns	+8.3	ns	+8.3	ns	+5.4	ns
			250 mg/kg FM	–	–	0.0	ns	+16.7	ns	+13.9	ns	+8.9	ns
			400 mg/kg FM	–	–	–2.4	ns	0.0	ns	+14.2	ns	–18.5	ns

Reference	Silage	Essential oil	Application rate	Δ% DM losses		Δ% pH		Δ% NH ₃ -N		Δ% Lactic acid		Δ% Acetic acid	
Foskolos <i>et al.</i> (2016)	Ryegrass 35 d	Eugenol	Control	–		5.81		1.74 g/kg DM		43.3 g/kg DM		–	
			50 mg/kg FM	–	–	–1.4	ns	–3.4	ns	+8.5	ns	–	–
			500 mg/kg FM	–	–	+2.4	ns	–17.8	ns	–18.2	ns	–	–
			2000 mg/kg FM	–	–	+11.5	ns	–44.8	*	–24.2	ns	–	–
		Thymol	Control	–		5.6		1.60 g/kg DM		39.9 g/kg DM		–	
			50 mg/kg FM	–	–	–2.5	ns	+6.3	ns	+4.3	ns	–	–
			500 mg/kg FM	–	–	+12.9	ns	+6.3	ns	–1.8	ns	–	–
			2000 mg/kg FM	–	–	+15.7	*	–53.1	*	–92.2	*	–	–
		Cynnamal-dehyde	Control	–		5.63		1.85 g/kg DM		43.7 g/kg DM		–	
			50 mg/kg FM	–	–	–0.7	ns	+5.4	ns	+28.4	ns	–	–
			500 mg/kg FM	–	–	+3.2	ns	–1.1	ns	+28.6	ns	–	–
			2000 mg/kg FM	–	–	+6.9	ns	–33.5	*	–17.4	ns	–	–
		Carvacrol	Control	–		5.77		1.81 g/kg DM		48.6 g/kg DM		–	
			50 mg/kg FM	–	–	+4.0	ns	–7.7	ns	–26.7	ns	–	–
			500 mg/kg FM	–	–	+12.0	*	–33.7	*	–39.5	*	–	–
			2000 mg/kg FM	–	–	+13.9	*	–40.3	*	–80.0	*	–	–
Önenç <i>et al.</i> (2015)	Field pea 60 d		Fresh forage	–		6.06		–		–		–	
			Control	15.2 g/kg		4.31		99.48 g/kg N		56.7 g/kg DM		23.8 g/kg DM	
		Oregano	400 mg/kg FM	+9.9	ns	+2.1	ns	+2.9	*	–17.6	*	–3.5	ns
		Cinnamom	400 mg/kg FM	–18.4	*	+3.7	*	–58.9	*	–54.1	*	–18.7	*
		Mixture 1:1	400 mg/kg FM	+19.7	*	+3.2	*	–31.4	*	–46.4	*	–27.6	*
Önenç <i>et al.</i> (2017)	Field pea 120 d		Fresh forage	–		6.06		–		–		–	
			Control	17.2 g/kg		5.18		99.08 g/kg N		45.3 g/kg DM		27.0 g/kg DM	
		Oregano	400 mg/kg FM	+98.3	*	+6.2	*	+32.4	*	–32.6	*	–27.2	*
		Cinnamom	400 mg/kg FM	+12.2	ns	+6.0	*	+38.1	*	–47.0	*	–14.9	*
		Mixture 1:1	400 mg/kg FM	+99.4	*	+6.4	*	+45.5	*	–37.9	*	–28.6	*
Li <i>et al.</i> (2022)	Paper mulberry 3 d	<i>Amomum villosum</i>	Control	–		5.77		–		44.9 g/kg DM		7.7 g/kg DM	
			1 mL/kg FM	–	–	–20.5	*	–	–	+23.6	ns	+83.1	*
	Paper mulberry 7 d		Control	–		5.51		–		58.4 g/kg DM		7.7 g/kg DM	
			1 mL/kg FM	–	–	–18.3	*	–	–	+3.1	*	+172.7	*
	Paper mulberry 15 d		Control	–		5.45		–		65.5 g/kg DM		10.9 g/kg DM	
			1 mL/kg FM	–	–	–18.0	*	–	–	–32.2	*	+149.5	*
	Paper mulberry 30 d		Control	–		5.32		–		31.4 g/kg DM		15.4 g/kg DM	
			1 mL/kg FM	–	–	–18.0	*	–	–	+140.8	*	+52.6	*
Sousa <i>et al.</i> (2022)	Sorghum + <i>Urochloa brizantha</i> 15 d		Control	–		4.07		–		–		–	
		<i>Lippia sidoides</i>	400 mg/kg FM	–	–	–1.7	ns	–	–	–	–	–	–
		62% Thymol	400 mg/kg FM	–	–	–2.0	ns	–	–	–	–	–	–
		100% Thymol	400 mg/kg FM	–	–	+0.7	ns	–	–	–	–	–	–
	Sorghum + <i>Urochloa brizantha</i> 30 d		Control	–		3.99		–		–		–	
		<i>Lippia sidoides</i>	400 mg/kg FM	–	–	–0.8	ns	–	–	–	–	–	–
		62% Thymol	400 mg/kg FM	–	–	–1.3	ns	–	–	–	–	–	–
		100% Thymol	400 mg/kg FM	–	–	+3.5	*	–	–	–	–	–	–
	Sorghum + <i>Urochloa brizantha</i> 45 d		Control	–		3.99		–		–		–	
		<i>Lippia sidoides</i>	400 mg/kg FM	–	–	–0.8	ns	–	–	–	–	–	–
		62% Thymol	400 mg/kg FM	–	–	0.0	ns	–	–	–	–	–	–
		100% Thymol	400 mg/kg FM	–	–	+4.5	*	–	–	–	–	–	–

Reference	Silage	Essential oil	Application rate	Δ% DM losses		Δ% pH		Δ% NH ₃ -N		Δ% Lactic acid		Δ% Acetic acid	
Akinci and Önenç (2021)	Vetch + Oat 70 d	Cumin	Fresh forage	–		6.20		–		–		–	
			Control	11.4 g/kg		4.46		56.54 g/kg N		33.6 g/kg DM		–	
			200 mg/kg FM	–4.4	ns	–0.7	ns	–4.1	*	+20.3	*	–	
			300 mg/kg FM	–17.5	ns	–0.7	ns	–14.1	*	+15.4	*	–	
			500 mg/kg FM	–5.3	ns	–0.7	ns	–14.3	*	+2.5	*	–	
Duru (2024)	Vetch + Wheat 75 d	Hypericum perforatum	Control	–		4.71		–		8.1 g/kg DM		2.7 g/kg DM	
			20 mg/kg	–	–	–3.2	*	–	–	+4.9	ns	+7.4	ns
			40 mg/kg	–	–	–3.8	*	–	–	+4.9	ns	+25.9	ns
			80 mg/kg	–	–	–4.2	*	–	–	+9.9	ns	+33.3	ns
			160 mg/kg	–	–	–4.2	*	–	–	+6.2	ns	+22.2	ns
Xu et al. (2023)	<i>Broussonetia papyfera</i> 3 d	<i>Amomum villosum</i>	Control	–		5,77		103,5 g/kg N		19,2 g/kg DM		8,9 g/kg DM	
			1 mL/kg FM	–	–	–20,5	*	–4,6	ns	+64,1	*	–13,5	ns
	<i>B. papyfera</i> : <i>P. sinese</i> 7:3 3 d		Control	–		5,68		102,6 g/kg N		14,7 g/kg DM		13,3 g/kg DM	
			1 mL/kg FM	–	–	+1,1	*	–7,9	*	+23,1	*	+12,8	ns
	<i>B. papyfera</i> : <i>P. sinese</i> 1:1 3 d		Control	–		5,49		108,0 g/kg N		19,5 g/kg DM		15,3 g/kg DM	
			1 mL/kg FM	–	–	–1,3	*	–13,1	*	+46,7	*	+31,4	*
	<i>B. papyfera</i> : <i>P. sinese</i> 3:7 3 d		Control	–		5,31		116,6 g/kg N		10,4 g/kg DM		11,6 g/kg DM	
			1 mL/kg FM	–	–	0,0	ns	–21,9	*	+47,1	*	+83,6	*
	<i>Pennisetum sinese</i> 3 d		Control	–		5,29		108,3 g/kg N		17,7 g/kg DM		21,3 g/kg DM	
1 mL/kg FM		–	–	0,0	ns	–8,2	ns	+33,9	ns	–11,3	ns		
Xu et al. (2023)	<i>Broussonetia papyfera</i> 30 d	<i>Amomum villosum</i>	Control	–		5,25		93,2 g/kg N		25,6 g/kg DM		22,1 g/kg DM	
			1 mL/kg FM	–	–	–15,2	*	+1,5	ns	+39,1	*	+57,9	*
	<i>B. papyfera</i> : <i>P. sinese</i> 7:3 30 d		Control	–		5,21		102,0 g/kg N		17,9 g/kg DM		24,2 g/kg DM	
			1 mL/kg FM	–	–	+0,6	ns	–10,4	*	+39,7	*	+26,4	*
	<i>B. papyfera</i> : <i>P. sinese</i> 1:1 30 d		Control	–		5,10		92,4 g/kg N		24,5 g/kg DM		33,5 g/kg DM	
			1 mL/kg FM	–	–	–1,0	*	–8,0	*	+38,4	*	+45,1	*
	<i>B. papyfera</i> : <i>P. sinese</i> 3:7 30 d		Control	–		5,04		99,2 g/kg N		17,8 g/kg DM		30,0 g/kg DM	
			1 mL/kg FM	–	–	0,0	ns	–13,5	*	+80,9	*	+67,0	*
	<i>Pennisetum sinese</i> 30 d		Control	–		4,95		104,8 g/kg N		26,4 g/kg DM		35,0 g/kg DM	
1 mL/kg FM		–	–	–6,9	ns	–10,6	*	+32,2	*	+23,7	*		
Andrade et al. (2024)	TMR 90 d		Control	26.2 g/kg		4.75		104.8 g/kg N		36.1 g/kg DM		7.6 g/kg DM	
		Monensin	35 mg/kg DM	–30.5	ns	0.0	ns	–15.6	*	+5.0	ns	+27.6	*
			45 mg/kg DM	–20.6	ns	+0.2	ns	–22.5	*	+16.1	ns	+38.2	*
		Limonene-rich essential oil	300 mg/kg DM	–21.8	ns	–0.2	*	–20.9	*	+40.2	*	+47.4	*
			600 mg/kg DM	–29.8	ns	–0.4	*	–26.0	*	+34.1	*	+42.1	*
Andrade et al. (2024)	TMR 90 d		Control	54.2 g/kg		4.01		84.6 g/kg N		55.0 g/kg DM		8.8 g/kg DM	
		Monensin	35 mg/kg DM	–13.1	*	+1.5	ns	+1.2	ns	+16.5	*	–12.5	*
			45 mg/kg DM	–25.3	*	+1.0	ns	+1.4	ns	+19.6	*	–13.6	*
		Limonene-rich essential oil	300 mg/kg DM	+9.6	ns	–1.2	ns	+1.4	ns	+30.7	*	+10.2	*
			600 mg/kg DM	+35.4	*	+4.0	*	+10.4	*	+5.3	*	+75.0	*

Tab. S11: Microbial counts of other silages treated with essential oils

Reference	Silage	Essential oil	Application rate	$\Delta\%$ Ethanol		$\Delta\%$ Yeasts (log cfu/g)		$\Delta\%$ Molds (log cfu/g)		$\Delta\%$ Enterobacteria / Clostridium (log cfu/g)		$\Delta\%$ LAB (log cfu/g)	
Cantolia Jr. <i>et al.</i> (2020)	Sugarcane 60 d	Lemongrass	Fresh forage	–		–		–		–		–	
			Control	101.0 g/kg DM		4.87		–		–		14.4 g/kg DM	
			1 mL/kg FM	–67.0	*Q	+3.7	*L	–	–	–	–	–40.1	*Q
			2 mL/kg FM	–92.1	*Q	–21.4	*L	–	–	–	–	–49.6	*Q
			3 mL/kg FM	–83.1	*Q	–36.1	*L	–	–	–	–	–33.3	*Q
Önenç <i>et al.</i> (2015)	Field pea 60 d		Fresh forage	–		–		–		–		–	
			Control	–		0.33		ND		–		–	
		Oregano	400 mg/kg FM	–	–	+384.8	*	ND	–	–	–	–	–
		Cinnamom	400 mg/kg FM	–	–	+663.6	*	ND	–	–	–	–	–
		Mixture 1:1	400 mg/kg FM	–	–	+557.6	*	ND	–	–	–	–	–
Zhou <i>et al.</i> (2024)	<i>Stylosanthes guianensis</i> 30 d		Fresh forage	–		5.06		4.46		7.59		6.29	
			Control	–		< 2.00		< 2.00		4.20		7.78	
		<i>Neolamarkia cadamba</i> leaf	5 g/kg FM	–	–	< 2.00		< 2.00		< 3.00		–18.3	*
			10 g/kg FM	–	–	< 2.00		< 2.00		< 3.00		–44.0	*
	Rice straw 30 d		Fresh forage	–		4.33		3.56		6.40		6.18	
			Control	–		< 2.00		< 2.00		6.00		8.14	
		<i>Neolamarkia cadamba</i> leaf	5 g/kg FM	–	–	< 2.00		< 2.00		< 2.00		–10.6	*
			10 g/kg FM	–	–	< 2.00		< 2.00		< 2.00		–18.7	*
Chaves <i>et al.</i> (2012)	Whole plant barley 365 d	Cinnamon leaf	Control	4.7 g/kg DM		3.50		1.20		–		7.30	
			125 mg/kg FM	+44.7	*	–25.7	ns	–58.3	ns	–	–	–1.4	ns
			250 mg/kg FM	+21.3	ns	–11.4	ns	–8.3	ns	–	–	–16.4	ns
			400 mg/kg FM	+10.6	ns	–42.9	ns	–16.7	ns	–	–	–1.4	ns
		Oregano	125 mg/kg FM	+10.6	ns	+5.7	ns	+41.7	ns	–	–	0.0	ns
			250 mg/kg FM	+8.5	ns	+31.4	ns	+125.0	ns	–	–	–13.7	ns
			400 mg/kg FM	+2.1	ns	–82.9	ns	0.0	ns	–	–	–2.7	ns
		Sweet orange	125 mg/kg FM	+55.3	*	–22.9	ns	+58.3	ns	–	–	–4.1	ns
			250 mg/kg FM	+27.7	ns	–34.3	ns	+41.7	ns	–	–	0.0	ns
			400 mg/kg FM	+6.4	ns	–34.3	ns	–16.7	ns	–	–	–4.1	ns
			400 mg/kg FM	+6.4	ns	–34.3	ns	–16.7	ns	–	–	–4.1	ns
Akinci and Önenç (2021)	Vetch + Oat 70 d	Cumin	Fresh forage	–		2.26		1.80		0.89		2.11	
			Control	–		1.87		1.87		0.85		3.46	
			200 mg/kg FM	–		–72.7	*	–81.3	*	–92.9	*	+82.1	*
			300 mg/kg FM	–		–84.0	*	–94.7	*	–92.9	*	+53.2	*
			500 mg/kg FM	–		–84.0	*	–94.7	*	–92.9	*	+48.8	*

Reference	Silage	Essential oil	Application rate	Δ% Ethanol		Δ% Yeasts (log cfu/g)		Δ% Molds (log cfu/g)		Δ% Enterobacteria / Clostridium (log cfu/g)		Δ% LAB (log cfu/g)	
Foskolos <i>et al.</i> (2016)	Ryegrass 35 d	Eugenol	Control	–		–		–		0.89		8.57	
			50 mg/kg FM	–	–	–	–	–	–	0.0	ns	–2.5	ns
			500 mg/kg FM	–	–	–	–	–	–	–21.3	ns	–2.7	ns
			2000 mg/kg FM	–	–	–	–	–	–	–13.5	ns	–7.9	*
		Thymol	Control	–		–		–		1.05		7.88	
			50 mg/kg FM	–	–	–	–	–	–	–15.2	ns	+2.2	ns
			500 mg/kg FM	–	–	–	–	–	–	+16.2	ns	+5.3	ns
			2000 mg/kg FM	–	–	–	–	–	–	–26.7	ns	–33.9	*
		Cynnamal-dehyde	Control	–		–		–		0.85		8.58	
			50 mg/kg FM	–	–	–	–	–	–	+70.6	ns	–1.7	ns
			500 mg/kg FM	–	–	–	–	–	–	+8.2	ns	–0.6	ns
			2000 mg/kg FM	–	–	–	–	–	–	+14.1	ns	–4.3	ns
		Carvacrol	Control	–		–		–		0.90		8.04	
			50 mg/kg FM	–	–	–	–	–	–	+6.1	ns	+3.1	ns
			500 mg/kg FM	–	–	–	–	–	–	–14	ns	+2.6	ns
			2000 mg/kg FM	–	–	–	–	–	–	+8.4	ns	–11.6	ns

Δ% – Percentage variation relative to the control mean; LAB – lactic acid bacteria; DM – dry matter; FM – fresh matter; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$; L – linear effect; Q – quadratic effect; ND – below detection limit.

Tab. S12: Aerobic stability of other silages treated with essential oils

Reference	Silage	Essential oil	Application rate	Δ% Aerobic stability		Δ% pH after aerobic exposure		Δ% DM losses by aerobic exposure	
Cantoia Jr. <i>et al.</i> (2020)	Sugarcane 60 d	Lemongrass	Control	52.2 h		4.24		14.1 g/kg/d	
			1 mL/kg FM	+15.3	*L	–4.2	*Q	+51.8	ns
			2 mL/kg FM	+66.7	*L	–5.7	*Q	+39.0	ns
			3 mL/kg FM	+103.1	*L	–2.6	*Q	–3.5	ns
Chaves <i>et al.</i> (2012)	Whole-plant barley 365 d	Cinnamon leaf	Control	–		5.50		–	
			125 mg/kg FM	–	–	–23.6	ns	–	–
			250 mg/kg FM	–	–	–20.0	ns	–	–
			400 mg/kg FM	–	–	–25.5	ns	–	–
		Oregano	125 mg/kg FM	–	–	+1.8	ns	–	–
			250 mg/kg FM	–	–	–5.5	ns	–	–
			400 mg/kg FM	–	–	–25.5	ns	–	–
		Sweet orange	125 mg/kg FM	–	–	–5.5	ns	–	–
			250 mg/kg FM	–	–	–12.7	ns	–	–
			400 mg/kg FM	–	–	–1.8	ns	–	–

Reference	Silage	Essential oil	Application rate	Δ% Aerobic stability		Δ% pH after aerobic exposure		Δ% DM losses by aerobic exposure	
Sousa <i>et al.</i> (2022)	Sorghum + <i>Urochloa brizantha</i> 15 d		Control	30.2 h		–		–	
		Lippia sidoides	400 mg/kg FM	+21.9	ns	–	–	–	–
		62% Thymol	400 mg/kg FM	+27.2	ns	–	–	–	–
		100% Thymol	400 mg/kg FM	+74.2	*	–	–	–	–
	Sorghum + <i>Urochloa brizantha</i> 30 d		Control	31.0 h		–		–	
		Lippia sidoides	400 mg/kg FM	+36.1	ns	–	–	–	–
		62% Thymol	400 mg/kg FM	+61.9	*	–	–	–	–
		100% Thymol	400 mg/kg FM	+23.2	ns	–	–	–	–
	Sorghum + <i>Urochloa brizantha</i> 45 d		Control	40.6 h		–		–	
		Lippia sidoides	400 mg/kg FM	–0.5	ns	–	–	–	–
		62% Thymol	400 mg/kg FM	+29.6	*	–	–	–	–
		100% Thymol	400 mg/kg FM	0.0	ns	–	–	–	–
Akinci and Önenç (2021)	Vetch + Oat 70 d	Cumin	Control	–		8.07		–	
			200 mg/kg FM	–	–	–7.1	*	–	–
			300 mg/kg FM	–	–	–7.1	*	–	–
			500 mg/kg FM	–	–	–2.5	*	–	–
Andrade <i>et al.</i> (2024)	TMR 90 d		Control	213.7 h		–		–	
		Monensin	35 mg/kg DM	+21.4	*	–	–	–	–
			45 mg/kg DM	+13.7	*	–	–	–	–
		Limonene-rich essential oil	300 mg/kg DM	+15.0	*	–	–	–	–
			600 mg/kg DM	+39.4	*	–	–	–	–
Andrade <i>et al.</i> (2024)	TMR 90 d		Control	83.9 h		–		–	
		Monensin	35 mg/kg DM	+9.1	*	–	–	–	–
			45 mg/kg DM	+6.2	*	–	–	–	–
		Limonene-rich essential oil	300 mg/kg DM	+6.0	*	–	–	–	–
			600 mg/kg DM	+10.0	*	–	–	–	–

Δ% – Percentage variation relative to the control mean; DM – dry matter; FM – fresh matter; * – significant at $P < 0.05$; ns – not significant at $P < 0.05$; L – linear effect; Q – quadratic effect.

