

vetmeduni

Bioactive compound research in ruminant nutrition **From Reductionism to Holism**

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Photo: Ratchaneewan Khiaosa-ard

PART 1

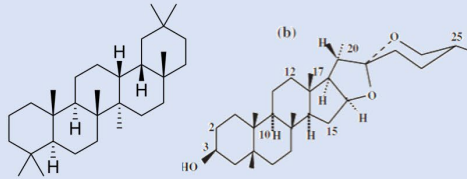
What do we know so far?



Plant bioactive compounds in ruminant research

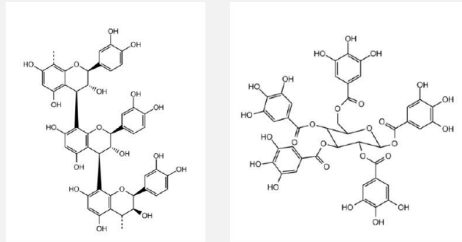
Saponins

- Terpenoids



Tannins

- Phenolic compounds



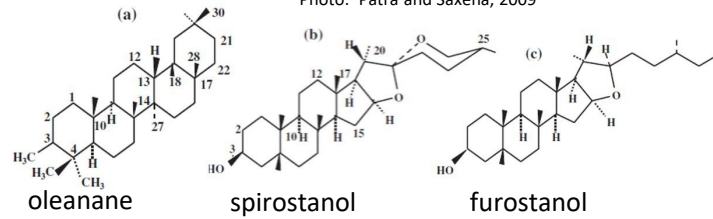
Essential oils

- Terpenes and more



Saponins

Photo: Patra and Saxena, 2009



University of Veterinary Medicine, Vienna

- Triterpenoid or steroidal aglycones linked to oligosaccharide moieties
- Found in plants and some marine organisms
- Heat-stable, **amphiphilic**, glycosidic compounds
- **Soap-like properties**
- Widely used in the folk medicine and pharmaceutical industry

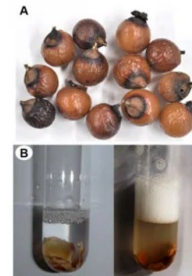
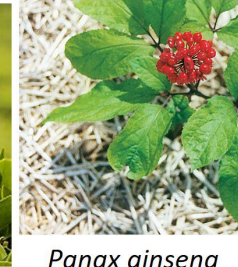


Photo: Rai et al., 2021



Photo credit: Wikipedia, or else stated

Saponins in ruminant research



Study (meta-analysis and systematic review)	Test forms	In vitro / In vivo	Publishing year	Animal	Rumen										Performance						Associated factors					
					Bacteria / archaea	Fungi	Protozoa	Rumen pH	Methane formation	N metabolism	SCFA production	acetate : propionate	Biohydrogenation	Feed digestion	Feed intake	Growth (ADG)	Milk yield	Milk fat	milk protein	Milk lactose	Dosage	Botanical origin/source	Extract vs Plant (Form)	Modification/Degradation in rumen	Duration (microbial adaptation)	Basal diet
Patra and Saxena 2009	Extract & Plants	Both	2009	multi				NR		NR	NR	NR	NR	NR	NR	NR	NR	X	(X)		(X)	(X)	(X)			
Jayanegara et al. 2014	Extracts	in vitro	2014	ND	NR	NR							NR		NR	NR	NR	X	X							
Dai and Ficiola 2019	Extracts	In vivo	2019	multi		NR							NR					X								
Almeida et al. 2021	Extracts	In vivo	2021	multi	NR	NR	NR	NR		NR	NR	NR	NR			NR	NR	NR	X							
Darabighane et al. 2021	Extracts	In vivo	2021	sheep	NR	NR	NR			NR			NR	NR	NR	NR	NR	X	X							
Kholif 2023	Extracts	in vivo	2023	multi	NR	NR		NR					NR	NR		NR		X	(X)		(X)	(X)	(X)			
Torres et al. 2023	Not mentioned	in vivo	2023	sheep	NR	NR					NR		NR			NR	NR	X	X				X			
Pepeta et al. 2024	Extract & Plants	in vivo	2024	multi	NR	NR	NR	NR		NR	NR	NR	NR	NR		NR	NR	X								
Yanza et al. 2024	Extracts	in vivo	2024	multi	NR	NR			NR				NR					X	X					X		

Decreased

Increased

Varied

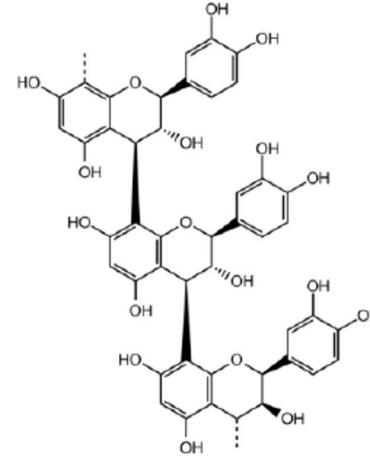
Unaffected

NR Not reported / not tested

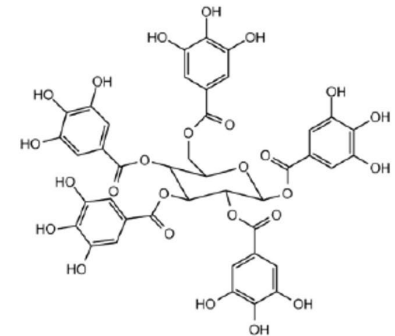
x Investigated
(x) Discussed

Tannins

- High molecular weight polyphenolic compounds of plant origin
- **Condensed tannins, Hydrolysable tannins**
- **Capable of complexing with other compounds such as proteins, fiber, and minerals**
- Heat stable
- Soluble in water and alcohols
- **Unabsorbed in the gut**
- Antioxidant properties
- Antimicrobial properties
- Decrease the palatability

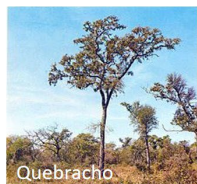


Condensed tannins
(flavonoid units)



Hydrolysable tannins
(Phenolic acid units)

Tannins in ruminant research



Quebracho



Acacia mearnsii



Castanea

Study	Test forms	In vitro / In vivo	Publishing year	Animal	Rumen									Feed digestion	Performance						Associated factors					
					Bacteria / archaea	Fungi	Protozoa	Rumen pH	Methane formation	N metabolism	SCFA production	acetate : propionate	Biohydrogenation		Feed intake	Growth (ADG)	Milk yield	Milk fat	milk protein	Milk lactose	Dosage	Botanical origin/source	Extract vs Plant (Form)	Modification/Degradation in rumen	Duration (microbial adaptation)	Basal diet
Patra and Saxena 2009	Extract & Plants	Both	2009	multi				NR	★	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	X	(X)		(X)	(X)		
Jayanegara and Palupi 2010	Extract & Plants	in vitro	2010	ND		NR		NR	NR		NR	NR	NR		NR	NR	NR	NR	NR	X						
Jayanegara and Palupi 2010	Extract & Plants	in vivo	2010	multi		NR		NR	NR		NR	NR	NR		NR	NR	NR	NR	NR	X						
Jayanegara et al. 2012	Extract & Plants	In vitro	2012	ND									NR		NR	NR	NR	NR	NR	X						
Jayanegara et al. 2012	Extract & Plants	In vivo	2012	multi		NR							NR		NR	NR	NR	NR	NR	X						
Dai and Ficiola 2019	Extracts	In vivo	2019	multi		NR							NR							X						
Herremans et al. 2020	Extract & Plants	In vivo	2020	Cows	NR	NR	NR	NR	NR		NR	NR	NR			NR				NR	X	X	X			X
Purba et al. 2020	Extract & Plants	Both	2020	multi	NR	NR	NR	NR	NR	NR				NR	NR	NR	NR	NR	NR	X						
Almeida et al. 2021	Extracts	In vivo	2021	multi	NR	NR	NR	NR		NR	NR	NR	NR			NR	NR	NR	NR	X						
Cardoso-Gutierrez et al. 2021	Plants	Both	2021	multi	NR	NR	NR	NR		NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	X	X				X	
Orzuna-Orzuna et al. 2021	Extract & Plants	In vivo	2021	Lambs	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR				NR	NR	X	X	X				
Orzuna-Orzuna et al. 2021	Extract & Plants	In vivo	2021	Beef cattle	NR	NR							NR	NR			NR	NR	NR	X	X	X		X	X	
Yanza et al. 2021	Extracts	In vivo	2021	multi		NR							NR	NR						X	X					
Friti et al. 2022	Extracts	In vivo	2022	multi	NR	NR							NR	NR						X	X					
Makmur et al. 2022	Extract & Plants	in vitro	2022	multi		NR								NR	NR	NR	NR	NR	NR	X						
Berça et al. 2023	Extract & Plants	In vivo	2023	Cattle	NR	NR	NR						NR		NR	NR	NR	NR	NR	X					X	
Berça et al. 2023	Extract & Plants	in vitro	2023	Cattle	NR	NR	NR								NR	NR	NR	NR	NR	X					X	
Brutti et al. 2023	Extract & Plants	In vivo	2023	Cattle	NR	NR	NR	NR	NR				NR	NR		NR	NR	NR	NR	X	X	X				
Torres et al. 2023	Not mentioned	in vivo	2023	Lambs	NR	NR	NR					NR					NR	NR	NR	X	X				X	
Fernandes et al. 2024	Not mentioned	in vivo	2024	Cattle	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR			NR			NR	X					
Pepeta et al. 2024	Extract & Plants	in vivo	2024	multi	NR	NR	NR	NR		NR	NR		NR	NR						X						
Susanto et al 2024	Extract & Plants	in vitro	2024	ND	(X)	NR		NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	X	X					

Effects of Dietary Tannins' Supplementation on Growth Performance, Rumen Fermentation, and Enteric Methane Emissions in Beef Cattle: A Meta-Analysis

José Felipe Orzuna-Orzuna ¹, Griselda Dorantes-Iturbide ¹, Alejandro Lara-Bueno ^{1,*}, Germán David Mendoza-Martínez ², Luis Alberto Miranda-Romero ¹ and Pedro Abel Hernández-García ³

23 % of data used
plants and forages,
77% tannin extracts

Effect on N metabolism

Sustainability 2021, 13, 7410

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Extracts are more powerful!

Dietary supplementation with tannins from studies using tannins (Figure 7). Figure 8 shows the effect of tannins on growth performance. However, when TANs were added to the diets in the form of extracts (SMD = −1.199; $p < 0.001$), when they came from a mixture of chestnut, *Quercus agrifolia*, and quebracho, CPD was not affected (SMD = −0.368; $p > 0.05$). However, when TANs were supplied as a part of the diet ingredients, CPD decreased significantly (SMD = −0.982; $p < 0.001$); however, when TANs were added to the diets in the form of extracts, CPD was not affected ($p > 0.05$; Figure 10). UNE decreased when TANs were added to the diets in the form of extracts (SMD = −0.558; $p < 0.001$). However, UNE increased when TANs were contained in the ingredients of the diets (SMD = 2.078; $p < 0.001$). On the other hand, UFE increased significantly when TANs were added to the diets in the form of extracts (SMD = $p < 0.001$); however, when TANs were supplied as a part of the diet ingredients, FNE was not affected (SMD = −0.368; $p > 0.05$).

Effect of dietary tannins on milk yield and composition, nitrogen partitioning and nitrogen use efficiency of lactating dairy cows: A meta-analysis

eterinary Medicine, Vienna

Sophie Herremans¹ | Frédéric Vanwindekens² | Virginie Decruyenaere¹ | Yves Beckers³ | Eric Froidmont¹

65% of data used
forage and byproducts
35% tannin extracts

1212

WILEY

HERREMANS ET AL.

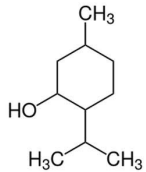
precision (Makowski, Piroux, & Brun, 2018) defined as $\frac{L}{SD_L} \frac{1}{SQ_L}$ with SD_L being the standard deviation of $L = \ln(R)$. If the intercept of the regression line differed from 0, a publication bias was detected. The trimfill function (Duval & Tweedie, 2000) with the default arguments was used on parameters subject to publication bias to evaluate the effect of this bias on the outcome.

3 | RESULTS

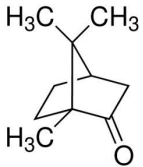
Through a literature search, 42 studies were identified. The treatments were divided into two groups: plants and forages (65% of the data) and tannin extracts (35% of the data). The present study showed that tannins had several impacts on dairy cows' milk production and nitrogen partitioning. The major findings are an absence of effect of tannins on corrected milk yield and NUE of dairy cows. Ruminant N-NH₃, MUN and urinary N excretion indi-

Essential oils

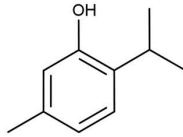
- Volatile oil, aromatic compounds in plants
- highly complex mixtures, including hydrocarbons such as terpenes (mainly monoterpenes) and sesquiterpenes, as well as oxygenated compounds (such as alcohols, aldehydes, ketones, acids, phenols, oxides, lactones, ethers, and ester)
- Soluble in water and alcohols
- antimicrobial and antioxidant functions



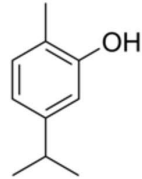
Menthol



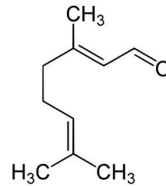
Camphor



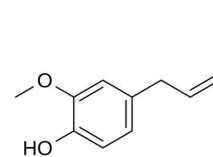
Thymol



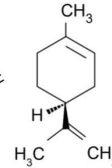
Carvacrol



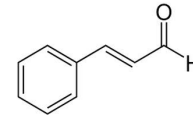
Citral



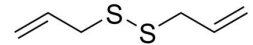
Eugenol



Limonene



Cinnamaldehyde



Diallyl disulfide



Essential oils in ruminant research

Study	Test forms	In vitro / In vivo	Publishing year	Animal	Rumen									Performance						Associated factors					
					Bacteria / archaea	Fungi	Protozoa	Rumen pH	Methane formation	N metabolism	SCFA production	acetate : propionate		Biohydrogenation	Feed digestion	Feed intake	Growth (ADG)	Milk yield	Milk fat	milk protein	Milk lactose	Dosage	Botanical origin/source	Extract vs Plant (Form)	Modification/Degradation in rumen
Patra and Saxena 2009	Extract & Plants	Both	2009	multi				NR		NR	NR		NR	NR	NR	NR	NR	X	(X)		(X)	(X)	(X)		
Klevenhusen et al. 2012	Single & blends	In vitro	2012	ND	NR	NR	NR	NR					NR	NR	NR	NR	NR	X					X		
Khiaosa-ard and Zebeli 2013	Single & blends	In vivo	2013	multi	NR	NR							NR		NR			X	X			(X)	(X)	X	
Dai and Ficiola 2019	Single & blends	In vivo	2019	multi									NR					X							
Belanche et al. 2020	Blend (Agolin®)	in vivo	2020	Cows	NR	NR				NR			NR	NR		NR		X				X			
Torres et al. 2020	Single & blends	In vivo	2020	Lambs	NR	NR					NR		NR				NR	X	X			X			
Almeida et al. 2021	Single & blends	In vivo	2021	multi	NR	NR	NR	NR		NR	NR	NR	NR			NR	NR	X							
Daning et al. 2021	Single & blends	In vivo	2021	Cows	NR	NR							NR	NR		NR		X							
Dorantes-Iturbide et al. 2022	Single & blends	In vivo	2022	multi (SR)		NR					NR							X	X			X			
Makmur et al. 2023	Single & blends	In vitro	2023	ND	NR	NR	NR		NR						NR	NR	NR	X							
Torres et al. 2023	Single & blends	in vivo	2023	Lambs		NR					NR		NR			NR	NR	X	X				X		
Fernandes et al. 2024	Single & blends	in vivo	2024	Cattle	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR			NR	X							
Pangesti et al. 2024	Single	In vitro	2024	multi		NR							NR		NR	NR	NR	X	X						
Permata et al 2024	Single	In vivo	2024	multi	NR	NR							NR			NR	NR	X							

Lipids in ruminant research

Study	Test forms	In vitro / In vivo	Publishing year	Animal	Rumen									Performance					Associated factors								
					Bacteria / archaea	Fungi	Protozoa	Rumen pH	Methane formation	N metabolism	SCFA production	acetate : propionate		Biohydrogenation	Feed digestion	Feed intake	Growth (ADG)	Milk yield	Milk fat	milk protein	Milk lactose	Dosage	Botanical origin/source	Extract vs Plant (Form)	Modification/Degradation in rumen	Duration (microbial adaptation)	Basal diet
Beauchemin et al. 2008	Oils, oilseeds and myristic ac	in vivo	2008	multi	NR	NR	★	NR		NR	NR	NR	NR		NR		NR	NR	NR	NR	X	X			(X)		
Eugene et al. 2008	Oils,Oilseeds and SFA	in vivo	2008	Cows	NR	NR	NR	NR		NR	NR	NR	NR	NR			NR				X						
Patra 2013	Oils, Oilseeds and FA	in vivo	2013	Cattle	NR	NR	NR	NR					NR		Q	NR	Q	NR	NR	NR	X		(X)			X	
Knapp et al. 2014	Seeds, oils and inert lipids	Both	2014	ND		NR		NR		NR	NR	NR	NR			NR	NR	NR	NR	NR	X	X			(X)	(X)	
Patra 2014	Oils, Oilseeds and FA	in vivo	2014	multi	NR	NR	NR	NR					NR			NR	NR	NR	NR	NR	X						X
Dai and Ficiola 2019	Oil (MCFA) & Oilseeds	In vivo	2019	multi		NR								NR							X						
Yanza et al. 2020	Oils and FA (MCFA)	in vitro	2020	ND		NR				Q			NR		NR	NR	NR	NR	NR	NR	X	X					
Yanza et al. 2020	Oils and FA (MCFA)	in vivo	2020	multi		NR					Q		NR		Q	NR	NR	NR	NR	NR	X	X					
Almeida et al. 2021	Oil (MCFA and PUFA)	in vivo	2021	multi	NR	NR	NR	NR		NR	NR	NR	NR				NR	NR	NR	NR	X					(X)	
Torres et al. 2023	FA & Oil	in vivo	2023	sheep		NR					NR		NR	NR	NR		NR	NR	NR	NR	X	X				X	



Other Compounds

Study	Test forms	In vitro / In vivo	Publishing year	Animal	Rumen									Performance						Associated factors							
					Bacteria / archaea	Fungi	Protozoa	Rumen pH	Methane formation	N metabolism	SCFA production	acetate : propionate		Biohydrogenation	Feed digestion	Feed intake	Growth (ADG)	Milk yield	Milk fat	milk protein	Milk lactose	Dosage	Origin/Source (Type)	Form	Modification/Degradation in rumen	Duration (microbial adaptation)	Basal diet
Ungerfeld et al. 2008	Fumarate	in vitro	2008	ND	NR	NR	NR	NR		NR	NR		NR	NR	NR	NR	NR	NR	NR	X			(X)	X			
Ungerfeld and Forster 2011	Malate	in vitro	2011	multi	NR	NR	NR	NR		NR	NR		NR	NR	NR	NR	NR	NR	NR	X			X	X			
Desnoyers et al. 2009	Yeast	in vivo	2009	multi	NR	NR	NR	NR	NR	NR			NR			NR			NR	X				X			
Sales 2011	Yeast (dry live yeast)	in vivo	2011	sheep	NR	NR			NR				NR				NR	NR	NR	NR	X			X	X		
Torres et al. 2022	Yeast and yeast products	in vivo	2022	Beef cattle	NR	NR			NR			NR					NR	NR	NR	NR	X		X		X	X	
Ogbuewu and Mbajorgu 2023	Yeast	in vivo	2023	Goats	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR			NR	NR	NR	NR	X		X				
Ogbuewu and Mbajorgu 2023	Yeast	in vivo	2023	multi (SR)	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR					X		X				X
Fernandes et al. 2024	Yeast-based additives	in vivo	2024	Cattle	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR			NR	NR	NR	X						
Almeida et al. 2021	Seaweeds (macroalgae)	in vivo	2021	multi	NR	NR	NR	NR		NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR	X						
Sofyan et al. 2022	Macroalgae	in vitro	2022	multi		NR	NR						NR		NR	NR	NR	NR	NR	NR	X	X					X
Sofyan et al. 2022	Macroalgae	in vivo	2022	multi	NR	NR	NR						NR			NR					X	X					X
Pepeta et al. 2024	Seaweeds (macroalgae)	in vivo	2024	multi	NR	NR	NR			NR	NR		NR	NR						NR	X						
Pepeta et al. 2024	Biochar	in vivo	2024	multi	NR	NR	NR	NR		NR	NR	NR	NR	NR	NR					NR	X						
Qomariyah et al. 2023	Biochar	in vitro	2023	ND	NR	NR							NR		NR	NR	NR	NR	NR	NR	X						
Qomariyah et al. 2023	Biochar	in vivo	2023	multi	NR	NR	NR						NR				NR	NR	NR	NR	X						
Harahap et al. 2020	Chitosan	in vitro	2020	ND												NR	NR	NR	NR	NR	X						
Sadarman et al. 2021	Black cumin seed	in vivo	2021	multi (SR)	NR	NR	NR	NR	NR	NR	NR	NR	NR					NR	NR	NR	X					X	X
Orzuna-Orzuna et al. 2023	Flavonoids: single or blend	in vivo	2023	Cattle	NR	NR			NR			NR									X	X	X		X	X	X
Orzuna-Orzuna et al. 2024	Capsaicin (Terpene alkaloids)	in vivo	2024	Cows	NR	NR	NR		NR				NR				NR				X						
Fernandes et al. 2024	Propolis	in vivo	2024	Cattle	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR				NR	NR	NR	NR	X					
Rezaei Ahvanooei et al. 2024	Monensin	in vivo	2024	Cows	NR	NR	NR					NR		NR			NR	NR	NR	NR	X						
Fernandes et al. 2024	Ionophores	in vivo	2024	Cattle	NR	NR	NR	NR	NR	NR	NR	NR	NR	NR					NR	NR	X						

Summary

- Extracts and purified forms of exotic plant origins
- Meta-analyses also show contradicting outcomes
- Overall several compounds show effects at the rumen level
 - Mainly affecting methanogenesis, SCFA pathways, and N metabolism
 - When protozoa are suppressed, methane likely goes down
 - Effects are often stronger in beef cattle than in dairy cattle
 - More successful with high concentrate diets
 - Microbial adaptation: weaker or no effect in the long term
 - They rarely influence rumen pH
- The effects post-rumen vary

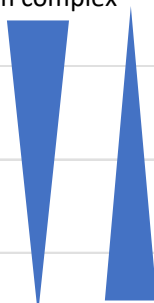
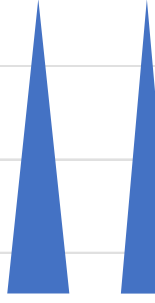
PART 2

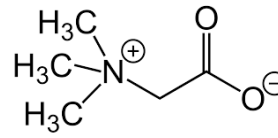
What should we know, too?



Things rarely studied

- Associated/dependent factors should be considered:
 - physicochemical properties under rumen and post rumen conditions, intestinal absorption, target tissues
 - Intact plant vs. extract, single bioactive compounds vs. blends
- Can we rely more on sustainable or economical sources?

Condition	Rumen	Abomasum	Small intestine	Large intestine	Direct effects (i.e., due to its presence)
Normal pH	pH 6-7	pH 2	pH 7-8	pH 6-7	
Enzyme actions	Microbial origin	Host origin	Host & Microbial origins	Microbial origin	
Condensed tannins (CT)	Form complex 	Complex released	Absorbed 	Form complex	Mainly within GIT, effects on biogas and soil
Hydrolysable tannins (HT)		Complex released		Form complex	Mainly within GIT, effects on biogas and soil
Flavonoids		Complex released		Biodegraded	Local and post GIT
Phenolic acids	Biodegraded	Complex released	Biodegraded	Biodegraded	Local and post GIT, excretion organs
Saponins	Partially biodegraded, Bloat forming	Low chemical or enzymatic hydrolysis (depend on type)	Biodegraded, Poorly absorbed	Biodegraded	Mainly within GIT, effects on biogas and soil
Essential oils	Biodegraded	?	Some can be greatly absorbed	Biodegraded	GIT, post GIT, excretion organs
Lipids	Freed and heavily transformed	Saponified fatty acids are freed	Greatly absorbed	not relevant	Local, post GIT, and tissues



Betaine addition as a potent ruminal fermentation modulator under hyperthermal and hyperosmotic conditions *in vitro*

Mubarik Mahmood,^{a,b} Renée Maxine Petri,^a Ana Gavráu,^c Qendrim Zebeli^a and Ratchaneewan Khiaosa-ard^a

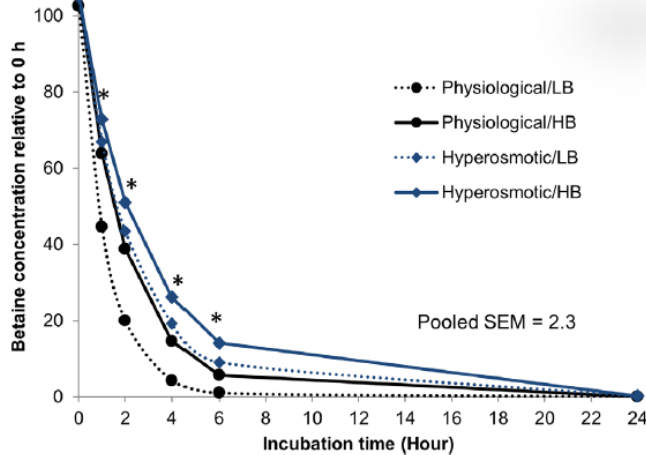
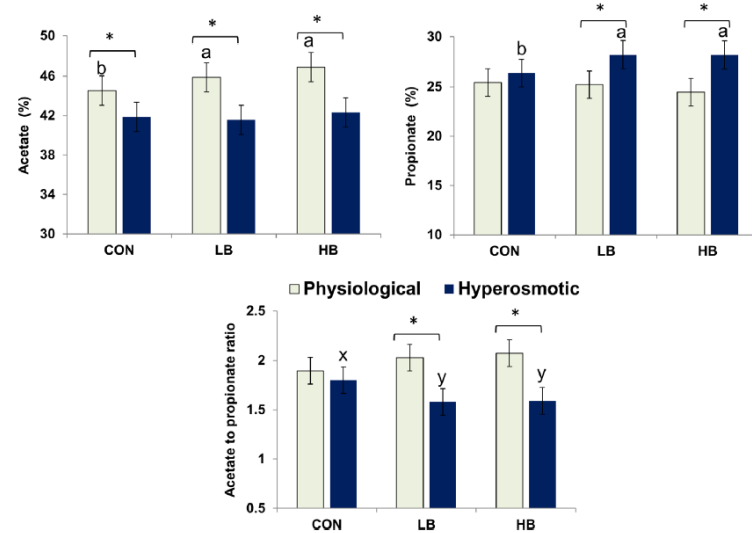


Figure 1. Ruminal disappearance of betaine (expressed as proportional change (%) of 0 h concentration) incubated under physiological and hyperosmotic rumen conditions *in vitro*. Target 0 h concentrations were 51 mg L⁻¹ (LB) and 286 mg L⁻¹ (HB). Asterisks indicate differences among treatments at each incubation hour ($P < 0.05$). [Color figure can be viewed at [wileyonlinelibrary.com](https://onlinelibrary.wiley.com)]



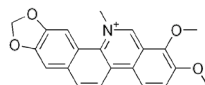
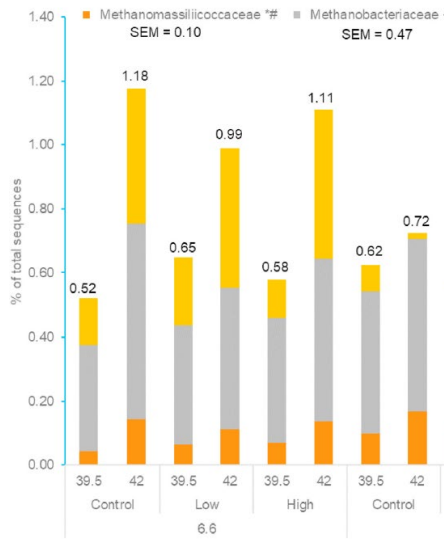
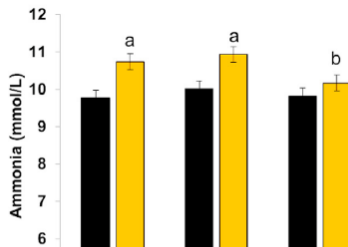
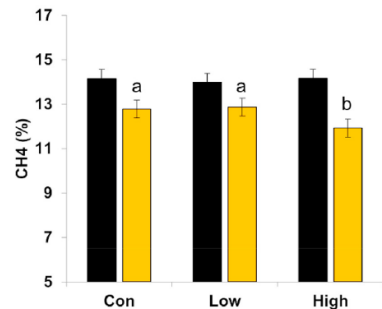
Some bacteria and archaea are more tolerant to stressors (heat, high osmolality, lower pH)



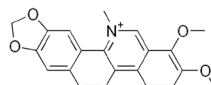
Anaerobes in the microbiome

Physicochemical stressors and mixed alkaloid supplementation modulate ruminal microbiota and fermentation *in vitro*

Ratchaneewan Khiao-ard ^{a, *}, Mubarik Mahmood ^{a, b}, Frederike Lerch ^a, Franz-Pius Trautinger ^a, Renée Maxine Petri ^a, Matthias Münich ^c, Qendrim Zebeli ^a



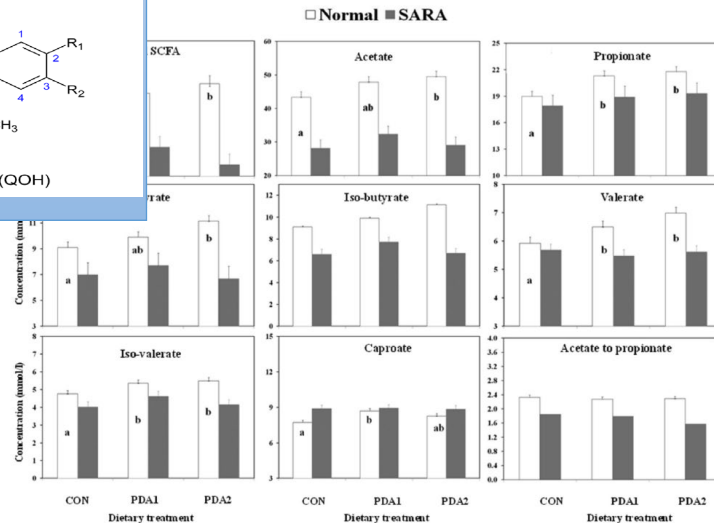
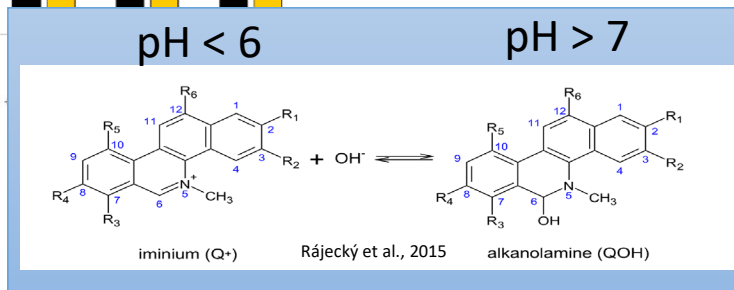
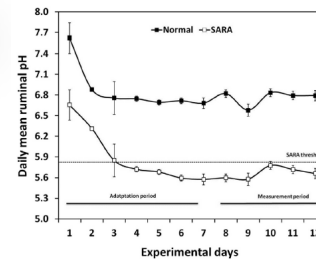
sanguinarine



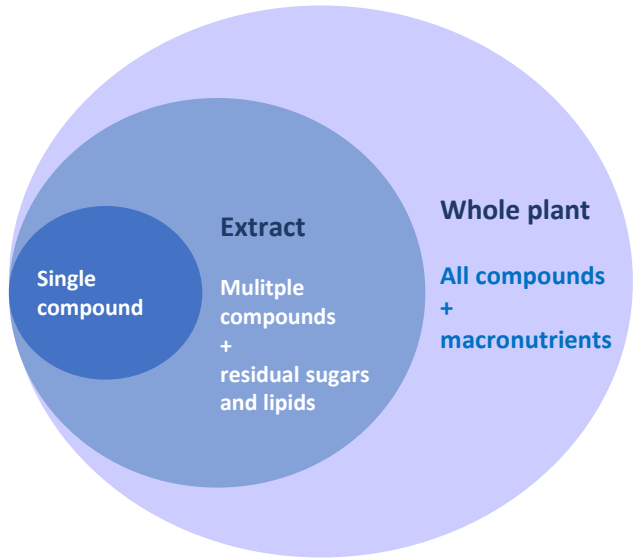
chelerythrine

Rumen microbial abundance and fermentation profile during severe subacute ruminal acidosis and its modulation by plant derived alkaloids *in vitro*

Elsayed Mickdam ^{a, b, d}, Ratchaneewan Khiao-ard ^{a, d}, Barbara U. Metzler-Zebeli ^{c, d}, Fenja Klevenhusen ^{a, d}, Remigius Chizzola ^a, Qendrim Zebeli ^{a, d, *}



Can we get similar effects from the whole plant matrix?



Functional compound → **Functional Feed**



pomace



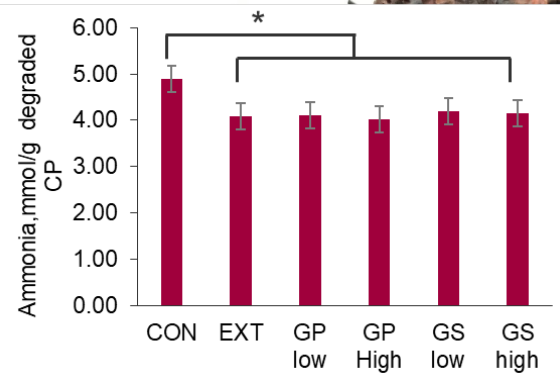
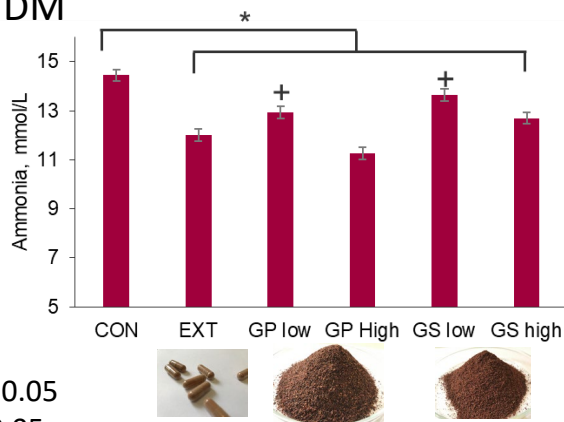
Seed meal



Seed extract

Wine by-products as functional feed

- Using grape tannins to decrease ruminal N losses from a high-fiber and high protein diet (56-70% **High quality hay**, dietary CP = 19-20%)
- 6 diets:
 - Neg Con
 - Pos Con (control diet + extract) (EXT)
 - Grape pomace (GP)- low (10%) and high (20%)
 - Grape seed (GS)-low (5%) and high (10%)
- EXT provided 3.4% total phenols in diet, winery by-products from 0.7 – 2.7% of diet DM



*Different from CON, $P < 0.05$

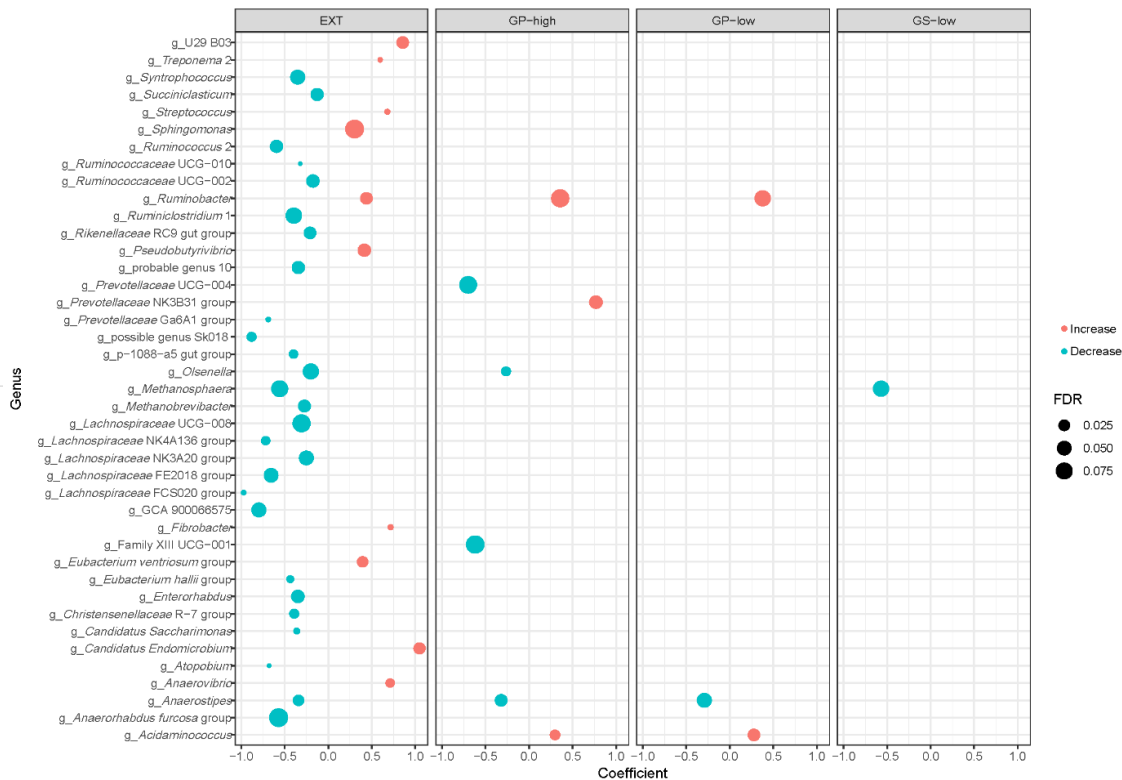
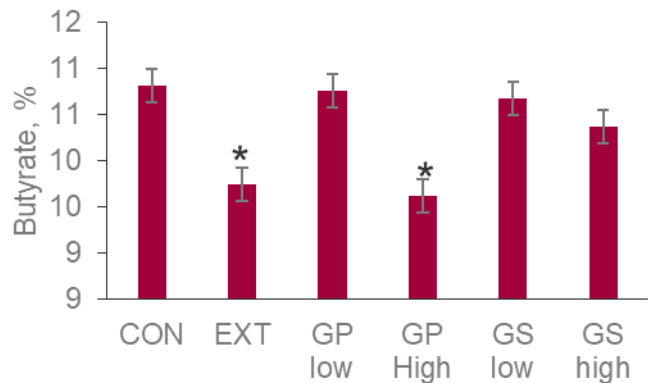
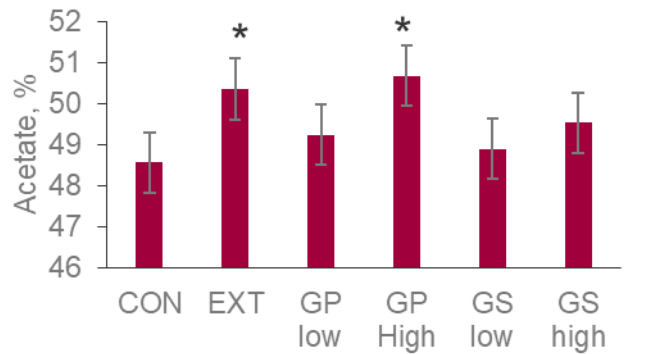
+Different from EXT, $P < 0.05$



Photo: R. Khiaosa-ard & Eduard Taufratzhofer



Bacteria & Archaea



Summary

- Possible to obtain similar effects of bioactive compounds in the plant matrix
- The more natural state of the product the friendlier the effect on rumen microbiota
- Modulation of a few genera might be sufficient to facilitate the community's functions
- Understanding the biochemistry and the fate of a compound under your test rumen conditions (which are affected by basal diets)
- Importance of positive controls

PART 3

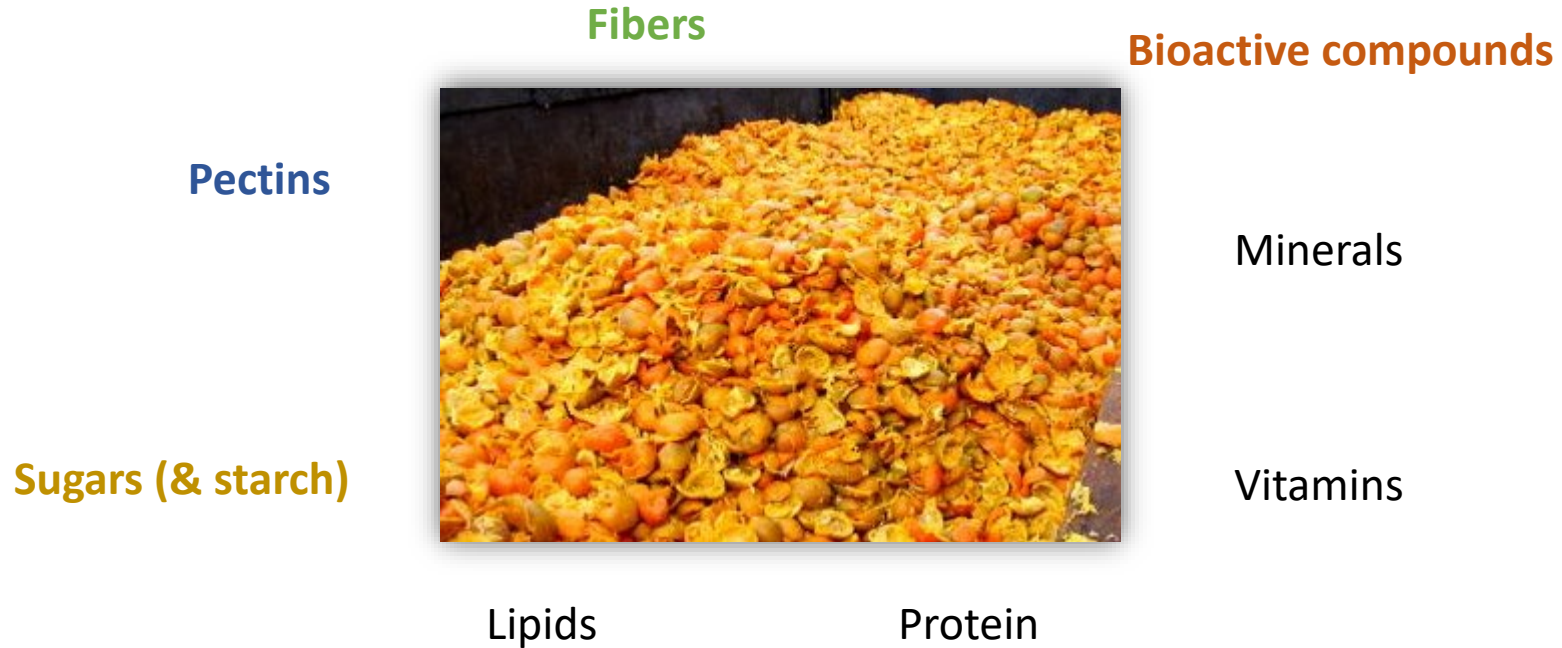
Future research



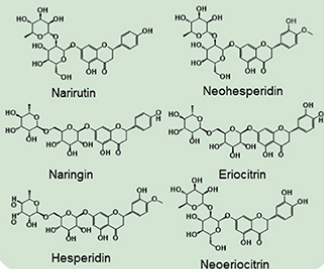
Future of bioactive compound research: Which direction?

- Use biomasses having low or no competition with human consumption
- Ask deeper questions about the biomass
- Effects beyond the animal level: we all are interconnected.

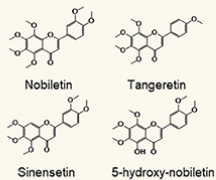
Fruit juice industry by-products



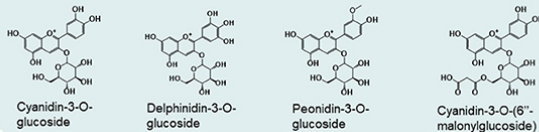
Flavanones



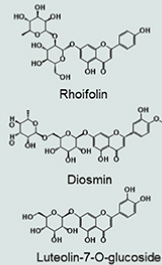
polymethoxylated flavones (PMFs)



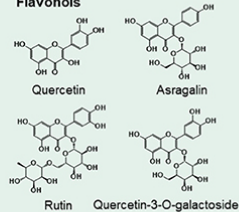
Anthocyanins



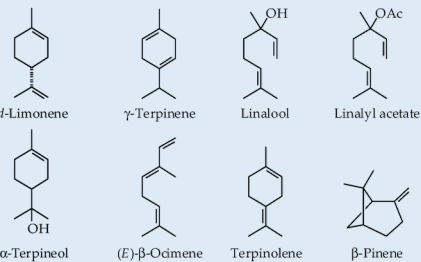
Flavones



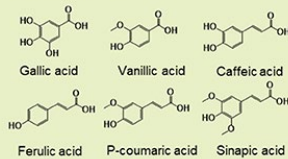
Flavonols



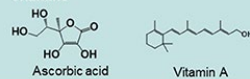
Terpenes (Key volatile components)



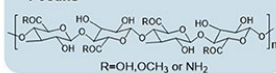
Phenolic acids



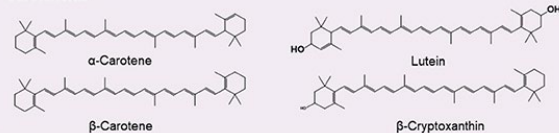
Vitamins



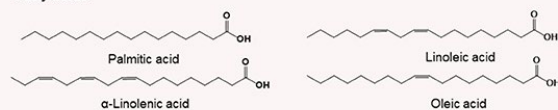
Pectins



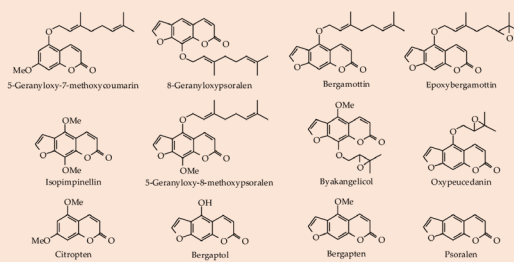
Carotenoids



Fatty acids



Coumarins (phenolic compounds)





The impact of citrus pulp inclusion on milk performance of dairy cows: A meta-analysis

Thomas Hartinger^a, Mubarik Mahmood^b, Ratchaneewan Khiaosa-ard^{a,*}

^a Centre for Animal Nutrition and Welfare, University of Veterinary Medicine Vienna, Veterinärplatz 1, Vienna 1210, Austria

^b Department of Animal Sciences, University of Veterinary and Animal Sciences, Lahore, Subcampus Jhang, Jhang 35200, Pakistan

Highlights

- Citrus pulp is rich in fibers, pectins and sugars but low in starch.
- Including up to 10% of citrus pulp in dairy rations improved milk yield and fat yield.
- Higher inclusion levels could affect intake but still maintain milk energy output.
- Citrus pulp has lipogenic properties by increasing milk fat precursors in the rumen.
- In support of sustainable farming, citrus pulp could become a common dairy feedstuff.





A project with Pakistan

Kooperation Entwicklungsforschung
Cooperation Development Research

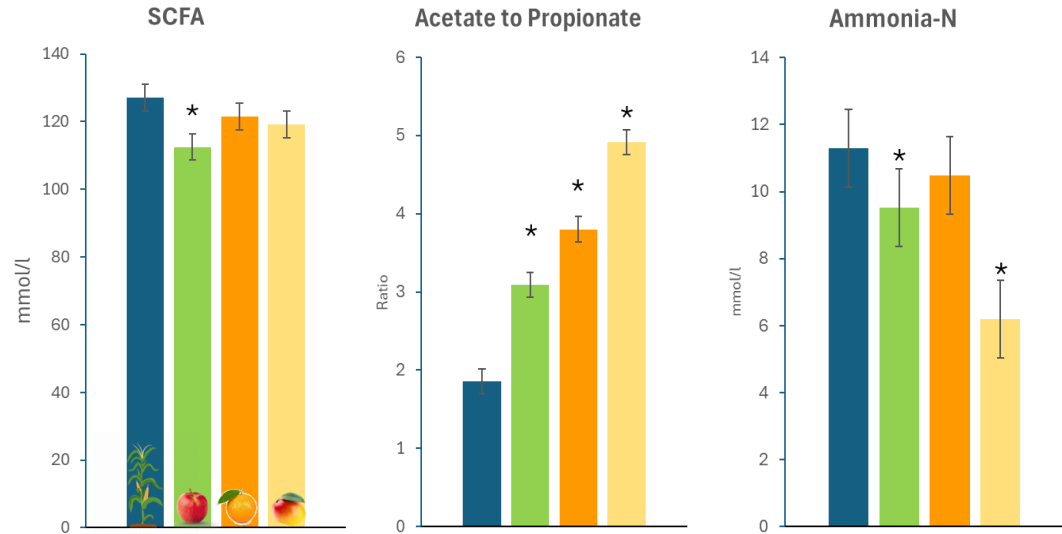
Federal Ministry
Republic of Austria
Education, Science
and Research

oead

Fruit and crop by-product silages



Apple pomace
Citrus pulp
Mango peels
+
Rice straw or
Sugarcane bagasse



See session 90 : Nutrition and feeding in the circular economy (16:30)

Test your local by-products



Pomegranate seed pulp, pistachio hulls, and tomato pomace as replacement of wheat bran increased milk conjugated linoleic acid concentrations without adverse effects on ruminal fermentation and performance of Saanen dairy goats

A. Razzaghi¹, A.A. Naserian^{2*}, R. Valizadeh³, S.H. Ebrahimi³, B. Khorrami³, M. Malekkhahi³, R. Khiaosa-ard³



Punic acid
(C9,t11,c13-18:3)
+ phenols



α -linolenic acid +
Protein



Oleic acid +
Phenols (esp.
Tannins)

Journal of Dairy Research

cambridge.org/dar

Effect of inclusion of bakery by-products in the dairy cow's diet on milk fatty acid composition

Ratchaneewan Khiaosa-ard, Anna Kaltenecker, Elke Humer and Qendrim Zebeli



Lipids (esp. oleic acid)

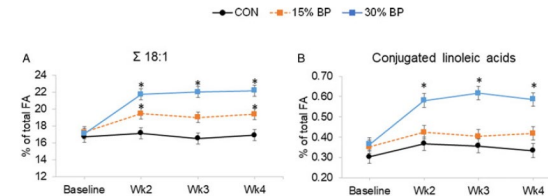
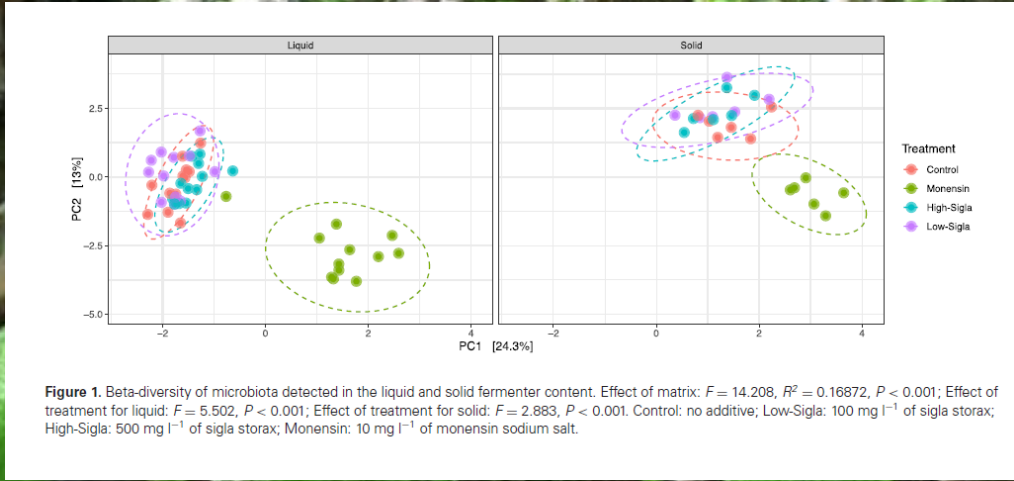


Table 1. Essential oil composition, antioxidant activity, and total phenolics of the tested sigla storax.

Composition of the essential oil (% of total identified components)		
Compounds	RI	Sigla storax
4-Ethyl-Phenol	1172	0.5
Benzenepropanol	1234	0.2
E-Cinnamyl alcohol	1307	
α -Cubebene	1353	1.0
Hydrocinnamyl acetate	1373	
β -Cubebene	1392	0.4
E-Caryophyllene	1421	0.3
β -Copaene	1432	1.4
Cinnamyl acetate	1449	
Ethyl-Cinnamate	1468	0.7
trans-4,10-Epoxy-Amorphane	1479	1.3
Germacrene D	1483	0.5
epi-Eubebol	1496	0.5
α -Muurolene	1502	1.0
Cubebol	1518	0.5
δ -Cadinene	1527	1.4
Oxygenated Sesquiterpene	1629	1.9
δ -Cadinol = Torreol =	1650	4.9
α -Muurolol		
Benzyl cinnamate	2093	1.0
3-Phenylpropanyl cinnamate— isomer	2174	0.7
Z-Cinnamyl cinamate	2256	0.7
3-Phenylpropanyl cinnamate	2320	38.1
E-Cinnamyl cinnamate	2424	38.8
Sum		95.9
Antioxidant activity and total phenolics		
Test system	Activity (mg g ⁻¹)	Sigla storax
DPPH	Trolox equivalents	14.8
	Gallic acid equivalents	6.0
FRAP	FeSO ₄ ·7H ₂ O equivalents	93.7
	Gallic acid equivalents	88.2
Total phenolics	Catechin equivalents	79.2
	Gallic acid equivalents	57.9

A typical 60% concentrate diet
4 treatments:

- Neg control (no additive)
- Pos control (monensin)
- Low Sigla oil
- High Sigla oil



Sigla storax (*Liquidambar orientalis*) mitigates *in vitro* methane production without disturbances in rumen microbiota and nutrient fermentation in comparison to monensin

Ahu Demirtas¹, Cátia Pacifico^{2,3}, Theresa Gruber⁴, Remigius Chizzola⁴, Qendrim Zebeli⁴, Ratchaneewan Khiaosa-ard^{4,5}

Methane %

Treatment	Methane %
neg Control	~16.5
Low-S	~15.5
High-S	~13.5
Monensin	~7.5

SCFA

Treatment	SCFA
neg Control	~100
Low-S	~98
High-S	~102
Monensin	~85

Form and properties of compounds in the biomass

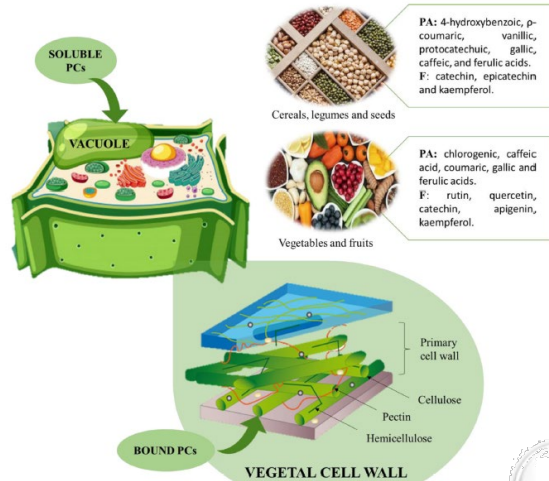
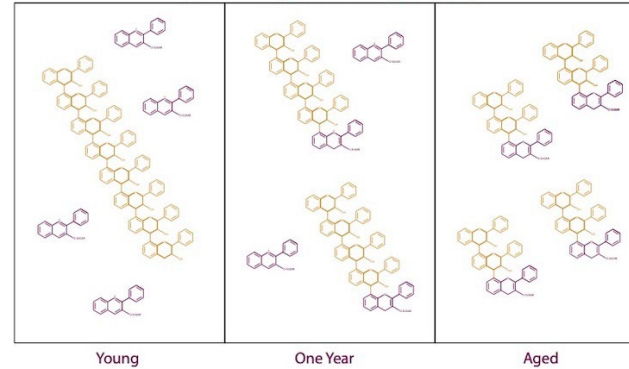


Photo: Rochetti et al., 2022

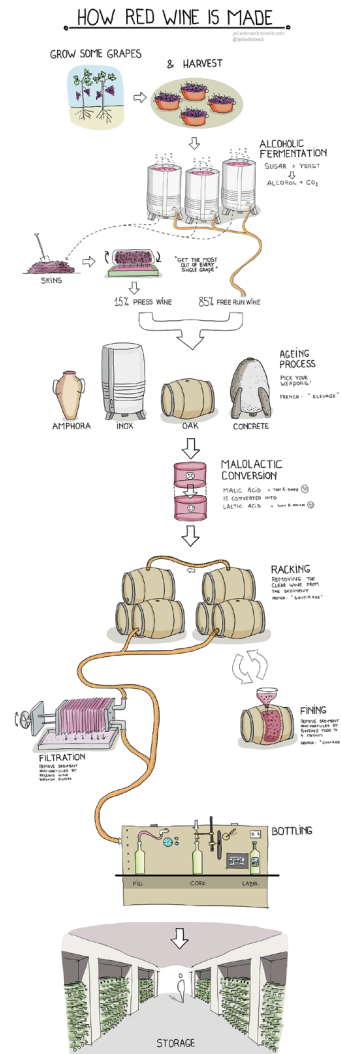


Photo: Quirós-Sauceda et al.
Rochetti et al., 2014

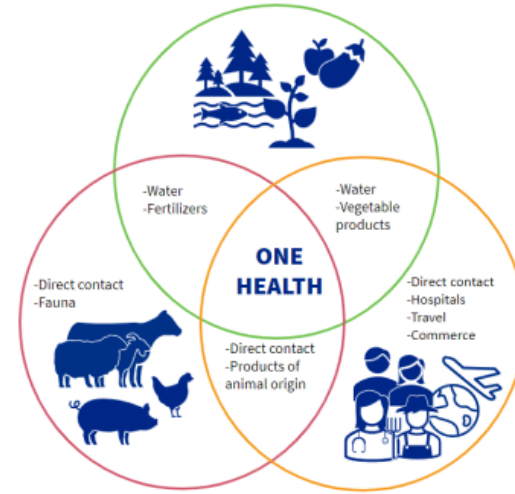
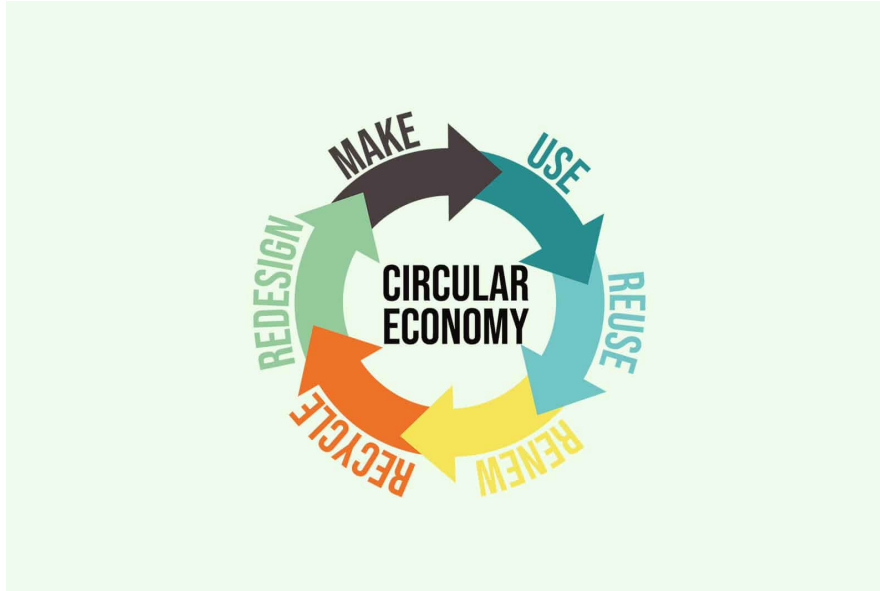
Behavior of Key Phenols During Wine Aging



Photos: Angelosante, <https://www.gildsomm.com/>
<https://winefolly.com/>



Beyond animal health & nutrition



Feed safety issue: microbial contaminants

Highlights

- Rumen bacteria and fungi were sensitive to grape phenols in an extract form.
- Winery by-products introduced non-*Saccharomyces* yeasts into the rumen community.
- Affected genera were not major contributors to the nutrient degradation.
- Donor cows' microbiota data are essential when interpreting the *in vitro* effects.



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Volume 89, October 2024, 102893



Anaerobes in the microbiome

Changes in the solid-associated bacterial and fungal communities following ruminal *in vitro* fermentation of winery by-products: aspects of the bioactive compounds and feed safety

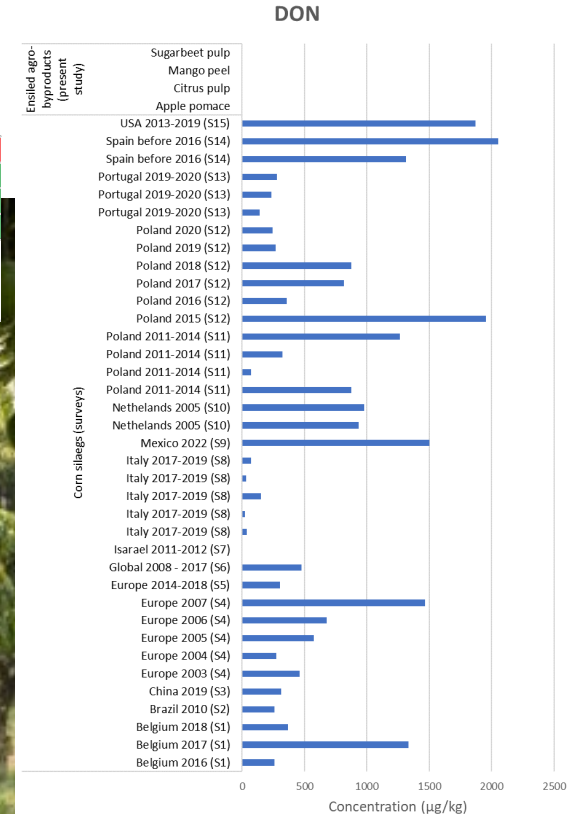
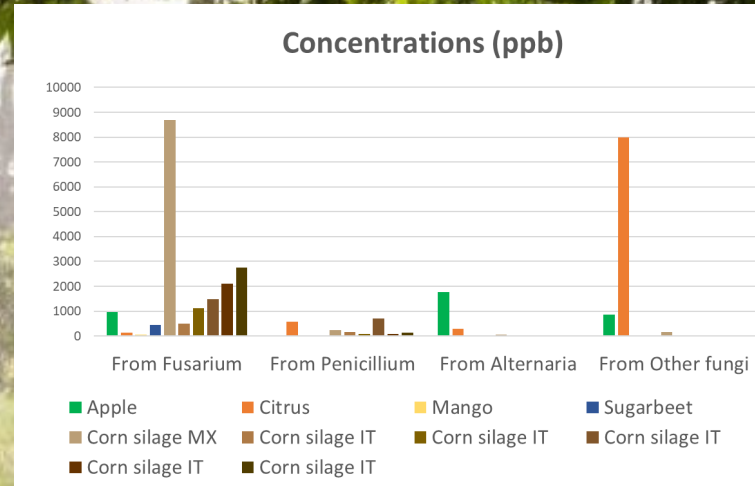
Ratchaneewan Khiaosa-ard ^{a,2}, Cátia Pacifico ^{a,2,1}, Mubarik Mahmood ^b,
Elsayed Mickdam ^c, Julia Meixner ^d, Laura-Sophie Trautinger ^d, Qendrim Zebeli ^a



Photo: R. Khiaosa-ard

Feed and food safety: chemical contaminants

Pesticide residues in samples (µg/kg)	Imidacloprid	Lufenuron	Metaxyl	Methoxyfenozid	Phorate-sulfoxide	Phoxim	Profenofos	Propargite	Pyridaben
Apple pomace silage	0.0	29.8	0.0	0.7	3.0	8.1	10.3	4937.3	27.0
Citrus pulp silage	0.0	0.0	2.5	0.0	0.0	0.0	91.7	62.7	0.0
Mango peel silage	2.6	0.0	0.0	0.0	0.0	14.5	0.0	108.3	0.0
Sugarbeet pulp silage	0.0	2.5	0.0	0.7	0.0	0.0	0.0	19.2	0.0
Applications	insecticide	veterinary flea control	fungicide	insecticide	insecticide metabolite	Insecticide	insecticide/miticide	insecticide	insecticide



Conclusions

Bioactive compounds play important roles in the food system

Rethinking about the sources

- researching your locally available resources
- Commonly abundant biomassess
- low to no competition with the sources for human needs

Understanding your target biomass

- Seeking more information and collaborations with other fields (e.g., botany, biochemistry, pharmacology, microbiology, food science and biotechnology, environmental science)

Considering the effects beyond the animal level

Special thanks to ...

University of Veterinary Medicine, Vienna

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Mubarik Mahmood
Ijaz Muawuz
Abdul Rahman

Egypt

Elsayed Mickdam

Turkey

Ahu Demirtas

Iran

Ali Razzaghi

Austria

Frederike Lerch
Julia Meixner
Laura-Sophie Traintinger
Franz-Puis Traintinger
Cátia Pacífico
Thomas Hartinger
Eduard Taufratzhofer
Elke Humer
Anna Kaltennegger
Qendrim Zebeli



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Kooperation Entwicklungsforschung
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PHYTOBIOTICS

