Evaluation of antimicrobial activities of phytophenols against pathogens that cause liver abscesses in feedlot cattle

<u>Cierra Roubicek¹, R.G.Amachawadi¹, Harith Salih¹, Jinrong Wang³, Yonghui Li³, and T.G. Nagaraja²</u> Departments of ¹Clinical Science and ²Diagnostic Medicine/Pathobiology, College of Veterinary Medicine, Kansas State University, Manhattan Kansas.

Department of ³Grain Science and Industry, College of Agriculture, Kansas State University, Manhattan, Kansas.

Introduction

Liver abscesses

- Liver abscesses occur in finishing cattle fed high-grain, low-roughage diets.
- The prevalence of liver abscesses is highly variable, but the average ranges from 10 to 20%.
- Liver abscesses in cattle continue to be of significant economic concern to the feedlot industry.
 Fusobacterium necrophorum, particularly of the subsp. necrophorum, which originates from the rumen, is the primary etiologic agent.

Broth Macrodilution Method

- Bacteria were cultured in Mueller-Hinton broth (Salmonella and Trueperella) or anaerobic brainheart infusion broth (Fusobacterium) with and without phytophenols.
- At 6, 12, 24, and 48 hours and bacterial concentrations were determined by reading optical density then doing serial dilution followed by spread plating to determine the viable bacterial count.

Minimum Inhibitory Concentrations (MIC) of phytophenols against liver abscess causing pathogens

Bacteria/ Compounds	Black sorghum (µg/ml)	Sumac sorghum (µg/ml)	Green tea (µg/ml)	Grape seed (µg/ml)	Rosemary (µg/ml)
Fusobacterium necrophorum	>100	>100	>100	>100	>100

- Trueperella (formerly Arcanobacterium) pyogenes is the secondary pathogen.
- A recent study has reported on isolation of Salmonella from liver abscesses of Holstein cattle.
- Tylosin is the most widely used feed additive to prevent liver abscesses.
- The future of tylosin use as a feed additive in feedlot cattle is uncertain do to it being added to the VFD in 2017.

Phytophenols

- Phytophenols are known to have antimicrobial/anti-inflammatory/antioxidant properties.
- High in phenolic compounds, thus, these plant extracts may have the potential to be used as natural antibiotic alternatives.
- The mechanism of antibacterial action of phenolic compounds are not yet fully understood.

Minimum Inhibitory Concentrations of phytophenols

• Microbroth dilution method was also used to quantify the inhibition.

Statistical analysis

- Data were analyzed using SAS (v. 9.4; Cary, NC).
- The PROCGLM procedure was used to fit the least squares in a linear model.The model included the fixed effect of tested phenolic extracts and replication as a random effect.
- Analysis of variance was performed on the log transformed MIC values.

Results

Total Phytophenolic Content (TPC) of phytophenols extracted from black sorghum bran, sumac sorghum bran, rosemary, green tea, and grapeseed

Plant extracts $TPC (m\sigma GAE/\sigma)$

Fusobacterium funduliforme	100	100	100	100	100
Salmonella Lubbock	>100	>100	>100	>100	>100
*Trueperella þyogenes	42.5	23.7	14.1	31.2	18.1
				*P	0.00



Microtiter plate showing inhibitory effects of phytophenols against Trueperella pyogenes

Objectives

• Our objectives were to investigate antimicrobial activities of plant based phenolic compounds on the liver abscess causing bacterial pathogens.

Materials and Methods

Extraction of Phytophenols

• Black sorghum, Sumac sorghum, Green tea extract, Grape seed extract, and Rosemary extracts were used for testing.



• The phytophenols were extracted using 75% aqueous acetone and total phenolic content was determined in a spectrophotometer.



Green tea extract	878.4		
Grape seed extract	614.1		
Sumac sorghum bran	374.9		
Black sorghum bran	258.3		
Rosemary extract	153.8		



Blood agar plates showing inhibitory effects of phenolic compounds from Black sorghum against Trueperella pyogenes (P < 0.001)



Summary

- Phytophenols from green tea, grape seed, rosemary, sumac sorghum and black sorghum inhibited *T. pyogenes*.
- Inhibitory effects of these extracts were directly correlated to their total phenolic content.
- Black sorghum phenolic compound had more inhibitory effect when compared to phenolic compound from Sumac sorghum.
- Phytophenols that inhibit the pathogens may have the potential to control liver abscesses.
- Further studies are ongoing to investigate different concentrations of phenolic compounds on the liver abscess pathogens.

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VWR-double beam spectrophotometer

Image: Control = Trueperella pyogenesImage: Treatment 24 h = Sumac sorghum + Trueperella pyogenesBlood agar plates showing inhibitory effects of
phenolic compounds from Sumac sorghum
against Trueperella pyogenes (P < 0.001)</td>

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