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Antibacterial and antioxidant activities of extracts of plants growing in Latvia and their potential use in veterinary medicine.

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Introduction

Bovine mastitis is most common and costly disease of dairy farming, that requires antibiotics use. The use of antibiotics in animal farming is a major contributor to antimicrobial resistance, and it is expected to grow by 8% between by 2030. Research shows that good husbandry practices and alternative treatment such as herbal medicine could be used instead. Plant secondary metabolites such as phenolic compounds have shown antibacterial and antioxidant activities, which contribute to battle infections.

Quercus robur bark (QB)

Calluna vulgaris herb (CH)

Tanacetum vulgare flower (TF) and leaf (TL)

Figure No 1. Plants used in study.

Research Objective

The aim of our projects is to develop and put into practice new, innovative phytopreparations, which contain plant extracts typical of Latvian flora, which have anti-parasitic, antibacterial and antioxidant activity in vitro.
This part of the project focused on antibacterial potential of plant extracts against bacteria that induce bovine mastitis.

Research Methods

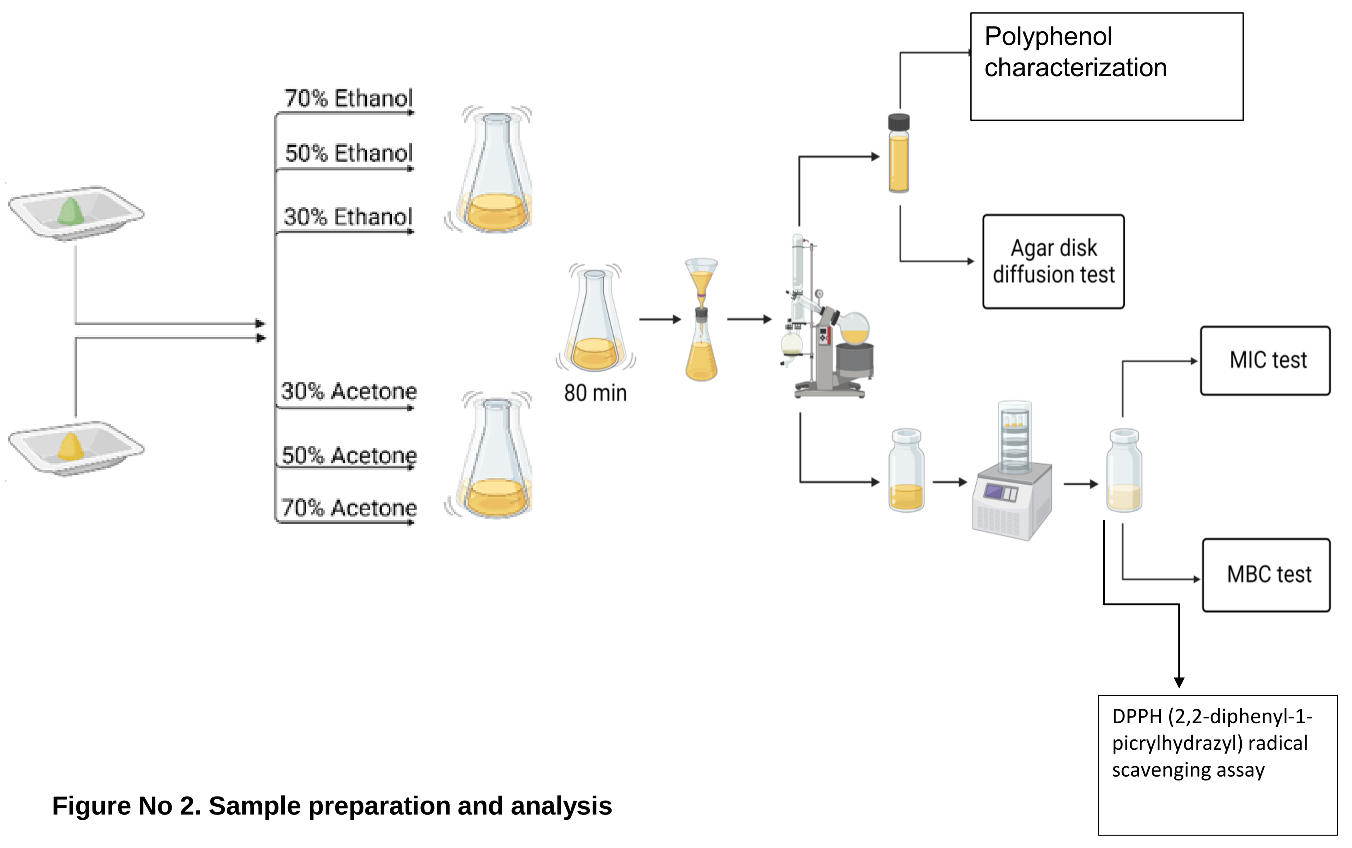


Figure No 2. Sample preparation and analysis

Results & Discussion

Plant Sample	TPC (mg GAE / g of lyophilised extract wt), ±SD	TFC (mg QE / g of lyophilised extract wt), ±SD	IC_{50} value of DPPH radical scavenging activity (μg/ml), ±SD
Ascorbic acid	-	-	43.92 ± 1.15
Ethanol extracts			
Calluna vulgaris herb	294.88 ± 14.20	51.13 ± 0.29	127.06 ± 1.07
Quercus robur bark	301.39 ± 10.17	5.11 ± 0.32	96.16 ± 1.03
Tanacetum vulgare flower	154.11 ± 7.95	25.12 ± 2.53	193.64 ± 1.10
Tanacetum vulgare leaf	158.48 ± 15.57	46.15 ± 0.29	185.35 ± 1.12
Acetone extracts			
Calluna vulgaris herb	285.61 ± 5.41	55.08 ± 2.23	104.71 ± 1.07
Quercus robur bark	316.02 ± 21.54	6.20 ± 0.22	83.95 ± 1.04
Tanacetum vulgare flower	155.38 ± 3.17	29.69 ±0.02	181.97 ± 1.07
Tanacetum vulgare leaf	225.99 ± 3.69	52.75 ± 2.37	146.55 ± 1.05

Table 1. Antioxidant activity (by DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay) of plant samples, Total phenolic (TPC) and Total flavonoid (TFC) content. Samples prepared with 50% ethanol or 50% acetone ratio 1:10

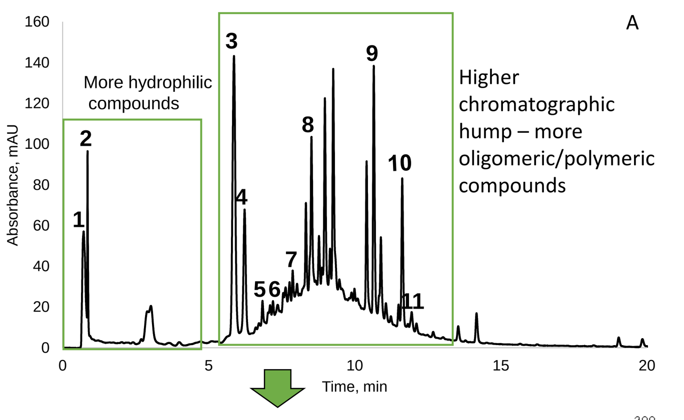
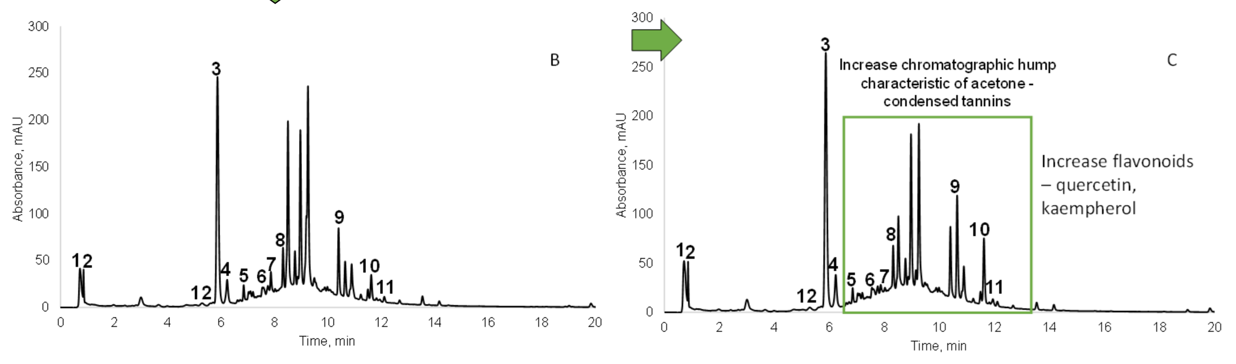


Figure No 3.
Effect of solvent concentration increase on extracted types of phenolic compounds.
A – acetone 30%
B – acetone 50%
C – acetone 70%



Minimal inhibitory concentration (MIC) and Minimal bactericidal concentration (MBC) of extracts by Broth microdilution method, mg/mL

	Q. robur bark Ethanol extract		*Q. robur* bark Acetone extract		*C.vulgaris* herb Ethanol extract		*T. vulgare* flower Ethanol extract		*T. vulgare* leaf Ethanol extract	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
E.coli ATCC	6.16	6.1	5.99	5.99	18.27	18.27	53.9	53.9	-	-
E. Coli V252	12.34	12.34	11.98	26.96	18.27	18.27	53.9	107.8	-	-
E.Coli V4	12.34	12.34	11.98	11.98	18.27	18.27	53.9	107.8	-	-
S.aureus ATCC	3.08	3.08	1.49	2.99	1.14	4.56	3.4	6.8	7.8	15.7
S.aureus V256	1.54	3.08	0.78	1.49	1.14	2.28	3.4	3.4	15.7	125.9
Strep. V171	24.68	24.68	23.96	23.96	18.27	36.53	53.9	53.9	31.4	62.9
Strep. V243	24.68	49.4	47.92	35.93	36.50	73.06	-	-	62.9	125.9
Serratia V251	-	-	47.92	47.92	73.06	73.06	-	-	125.9	125.9

Table 2. Antibacterial activity of plant samples

Bovine mastitis pathogens and reference cultures: *Escherichia coli (ID. V-2019-4), E. coli (ID. V-2019-252), E. coli (ATCC 25922), Streptococcus agalactiae (ID. V-2019-171), S. uberis (ID. V-2019-243), Serratia liquefaciens (ID. V-2019-251), Staphylococcus au-reus (ID. V-2019-256), S. aureus (ATCC 25923).*

- All six types of extracts for each plant were screened by the agar disc diffusion test for antibacterial activity. The best samples were selected for MIC and MBC determination.
- All *Q. robur* bark acetone extracts had antibacterial activity, but ethanol extract did not affect *Serratia liquefaciens.* The 30% ethanol extract of *Q. robur* bark measured the highest bacterial inhibition zones, whereas 70% acetone extracts - the broadest spectrum (all tested bacteria). The lowest MIC/MBC was observed against *S. aureus*.
- Lower concentration extracts of *C. vulgaris* were effective against all tested bacteria. The 30% ethanol extract of *C. vulgaris* herb had the broadest spectrum, although it had better activity against Gram-positive bacteria.
- MIC and MBC values of the 70% ethanol extract of *Tanacetum vulgare* flower were lower than for the 70% ethanol extract of *T. vulgare* leaf.
- All analyzed plants are good source of phenolic compounds. These compounds contribute to antibacterial and antioxidant activity, which can be helpful in healing process.
- Antioxidant activity of 50% extracts: TF ethanol< TL ethanol< TF acetone< TL acetone< CH extracts< QB extracts.
- Various factors could affect the antibacterial effectivity of extract like the type of solvent, concertation, extraction process, and plant material. Solvent polarity and the plant part used could affect extracted phenolic compound amounts and effects.
- Type of bacterial wall (Gram positive or Gram negative) also affects activity since most of the extracts had better effects against Gram positive bacteria.

Conclusions

Tested extracts can potentially be used in the development of antibacterial phytopreparations, which would contribute to a more ecologically friendly medicine and reduce resistant pathogens contamination in the environment.

Acknowledgements

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