EVALUATION OF THE ANTIMICROBIAL EFFICACY OF *SATUREJA MONTANA* ESSENTIAL OIL NANOEMULSIONS

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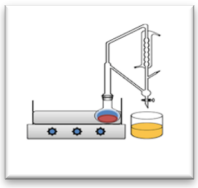
Introduction

Poultry meat exhibits the highest overall levels of *E. coli* contamination and its ability to form biofilm contributes to the bacterial persistence on surfaces of poultry products[1]. Recently, several Authors reported that essential oils (EOs) from different species of *Satureja*, belonging to the botanical family of *Lamiaceae*, possess remarkable antibacterial activity[2]. Oil in Water nanoemulsions (NEs) composed by *Satureja* essential oil (SEO), Hepes buffer and nonionic surfactant as NEs stabilizer were prepared, deeply characterized then assayed on the planktonic growth and biofilm formation of *E. coli* strains from fecal samples of healthy chickens.

Materials and Methods

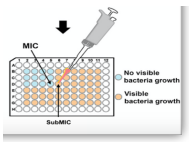
PLANT HARVESTING

OIL EXTRACTION AND CHARACTERIZATION

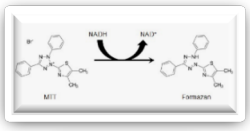


NANOEMULSION DESIGN

ANTIMICROBIAL ANALYSIS



MTT ASSAY



Results

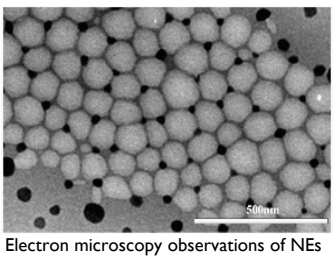
CHARACTERIZATION OF SEO AND NEs

Class	Compound	SEO (%)
	α-thujene	-
Terpene	α-pinene	0.4
	camphene	0.2
	β-pinene	0.2
	β-myrcene	0.5
	3-carene	-
	α-terpinene	-
	p-cymene	12.3
	limonene	1.8
Terpenoid	eucalyptol	6.8
Terpene	ocimene	0.2
	γ-terpinene	15.0
Terpenoid	linaloxide (cis)	0.2
Terpene	terpinolene	-
Terpenoid	linaloxide (trans)	0.1
	linalool	16.4
	camphor	0.3
	borneol	0.5
	terpinen-4-ol	-
	γ-terpineol	0.2
	α-terpineol	-
	thymolmethylether	-
	D-carvone	1.6
	thymol	14.5
	carvacrol	23.9
	eugenol	0.4
Sesquiterpene	β-caryophyllene	0.4
	α-humulene	0.1
	caryophylleneoxie	-
	unidentified	4.0
Terpene		30.6
Terpenoid		64.9
Sesquiterpene		0.5

Percentage of volatile components of SEO

HD (nm) ± SD	95.05 ± 1.55
ζ-Pot (mV) ± SD	-13.02 ± 0.36
PDI ± SD	0.23 ± 0.01

Hydrodynamic diameter (HD), ζ -potential and polydispersity index (PDI) values of NEs

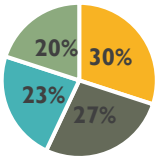


Electron microscopy observations of NEs

ANTIMICROBIAL ACTIVITY OF SEO AND NEs

MIC/MBC values of SEO and NEs ranged from 0.78 to 3.12 mg/mL. Interestingly, while SEO was bactericidal for 43% of the strains, SEO in NEs was for 100% at the concentration of 1.56 mg/mL.

BACTERIAL BIOFILM PRODUCTION

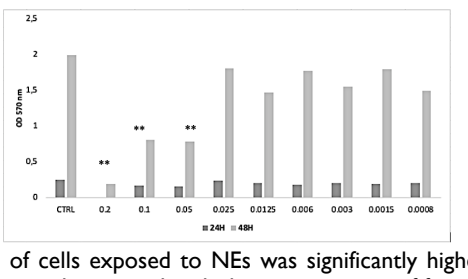


ANTIBIOFILM ACTIVITY

E. coli	Biofilm	SEO inhibition (%)	NEs inhibition (%)	SEO eradication (%)	NEs eradication (%)
AV1	Strong	43	53	47	144
AV2	Strong	53	95	45	212
AV3	Strong	147	6	239	518
AV12	Strong	116	65	170	157
AV18	Strong	26	28	133	155
AV25	Strong	138	63	104	386
AV27	Strong	43	86	44	252
AV38	Strong	44	61	6	160
AV40	Strong	79	80	119	422

Sub-MIC concentrations of SEO, free or in NEs, significantly reduced biofilm production of the strong producer strains, whereas they were unable to efficiently eradicate the preformed biofilm.

CYTOTOXICITY



The survival of cells exposed to NEs was significantly higher than that found in samples treated with the same amount of free oil ($p \leq 0.05$ = * $p \leq 0.01$ = **)

Conclusions

Newly designed *S. montana* essential oil based nanoemulsions was able to preserve and improve antimicrobial and antibiofilm activity compared to essential oil alone. SEO nanoemulsions may represent a bright antibacterial solution due to the enhanced contact-rate with the external structure. Further studies are needed to recognize the contribution of components of essential oils to the antimicrobial effect.

References

[1] Yi Sun, Xueyan Hu, Du Guo, Chao Shi, Chunling Zhang, Xiaoli Peng, Hua Yang, and Xiaodong Xia. Disinfectant Resistance Profiles and Biofilm Formation Capacity of Escherichia coli Isolated from Retail Chicken. MICROBIAL DRUG RESISTANCE Volume 25, Number 5, 2019.

[2] Vitanza, L., Maccelli A., Marazzato M., Scazzocchio F., Comanducci A., Fornarini S.; Crestoni M.E.; Filippi A.; Fraschetti C.; Rinaldi F. Satureja montana L. essential oil and its antimicrobial activity alone or in combination with gentamicin. Microb. Pathog., 126, 323–331, 2019.