

Trajnostni razvoj kmetijstva in turizma na čezmejnem Krasu Sviluppo sostenibile dell'agricoltora e del turismo nel Carso transfrontaliero

AGROTUR II Simpozij Akademija o teranu/Accademia di Terrano Grad Štanjel/II castello Štanjel 8. 11. 2018

















Preliminarni rezultati študije mlečnokislinskih bakterij povezanih z vinom Teran

I resultati preliminari dello studio di batteri malico-latici associati ai vino Terrano

dr. Lorena Butinar in Jelena Topić Božič Univerza v Novi Gorici

















LACTIC ACID BACTERIA (LAB)

- Naturally present in grapes, musts and wines

 predominant genera Leuconostoc,

 Pediococcus, Lactobacillus, Oenococcus
- Malolactic fermentation (MLF) conversion of L-malic to L-lactic acid
 - a decrease in total wine acidity,
 - enhances organoleptic properties,
 - improves microbiological stability.
- Formation of biogenic amines (BA)
- Formation of volatile phenols





















LACTIC ACID BACTERIA (LAB)

- to isolate, characterize and identify autochthonous LAB present on grapes and wines
- to test autochthonous LAB for wine production

















Identification:

- Molecular methods

(rpoB, 16S rRNA genes)

- API CH50 test

Grape/ wine

Set up of autochthonous LAB collection

- 1. Plating on MRS + 2% TJ + cycloheximide
- 2. Isolation of pure LAB cultures
- 3. Storage of cryo-cultures in 30% glycerol on -80 °C

Characterization

BA-producing LAB

- decarboxylase screening medium (the plate/microtiter plate procedure)
- Chromatographic analysis (HPLC), multiplex PCR VP-producing LAB
- Medium + p-CA acid

NON-producing BA and VP LAB strains:

SO₂ and ethanol tolerance, qualitative determination of enzyme activities (glucosidase, esterase)

Micro-vinification experiment using LAB selection

















LACTIC ACID BACTERIA (LAB)

- tested 227 LAB strains:
 - 30 strains thyramin-producers
 - 6 strains putrescine-producers
 - 9 strains cadaverine-producers
 - 6 strains histamine-producers
 - 13 strains ethyl phenol-producers

















ANALYSIS OF BIOGENIC AMINES

Analysis was focused on the detection of **five biogenic amines**:





Putrescine

Cadaverine



Ethanolamine











HC



Tyramine





SAMPLE PREPARATION

- 12 wine samples analyzed (T1 T12)
- Analysis was performed in duplicates



Sample was prepared directly before the injection due to instability of derivates

















ANALYSIS OF BIOGENIC AMINES

HIGH PERFORMANCE LIQUID CHROMATOGRAPHY COUPLED TO FLUORESCENCE DETECTOR (adapted method from OIV - Resolution OIV-Oeno 346/2009)



- TIME OF ANALYSIS PER SAMPLE: 75 min
- CALCULATION OF CONCENTRATIONS: external standards of biogenic amines
- Retention times
 - Putrescine (PUT): 16.7 min
 - Cadaverine (CAD): 22.0 min
 - o Ethanolamine (ETH): 23.6 min
 - Histamine (HIS): 24.8 min
 - Tyramine (TYR): 49.7 min



















Presence of biogenic amines in tested wines.

BIOGENIC AMINE									
SAMPLE	PUT	CAD	HIS	TYR	ETH				
T1	+	-	-	+	+				
Т2	+	-	+	+	+				
Т3	+	-	+	+	+				
Т4	+	-	-	+	+				
Т5	+	-	-	+	+				
Т6	+	-	-	+	+				
Т7	-	-	-	+	+				
Т8	+	-	-	+	+				
Т9	+	-	-	+	+				
T10	+	-	-	+	+				
T11	+	-	-	+	+				
T12	+	-	-	+	+				

















Concentration range [mg/L] of the tested biogenic amines.

SAMPLES	PUT	CAD	HIST	TYR	ETH
T1 – T12	14,0 - 114,5	n.d.	n.d – 10,3	< LOD – 5,7	7,7 – 16,7

* n.d. = not detected

- * <LOD = under limit of detection
- The highest concentration of biogenic amine was found in sample T10 (putrescine 115 mg/L).
- In general putrescine was present in the highest concentrations in the samples
- In two of the samples (T2 and T3) histamine was presented in concentration 2 and 10 mg/L.
- Average concentration of putrescine was 47.5 mg/L, while ethanolamine had the average concentration of 12.9 mg/L

















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Hvala za pozornost. Grazie per l'attenzione.













