



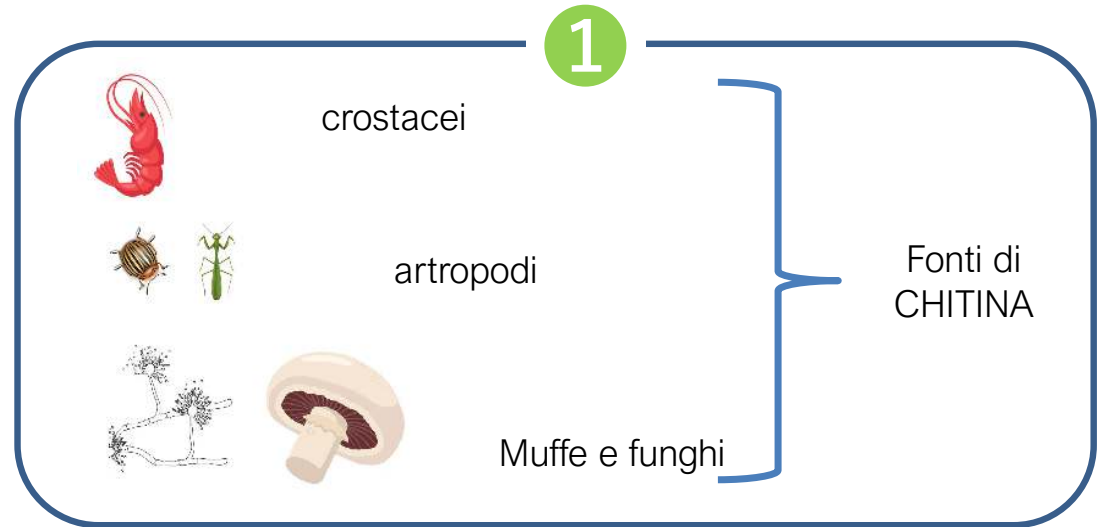
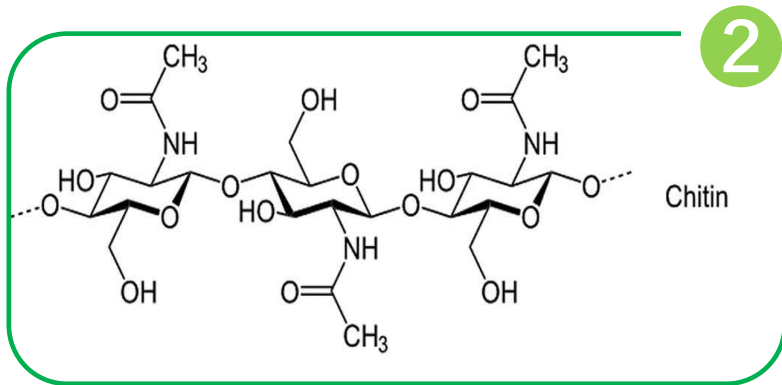
ALMA MATER STUDIORUM
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Il chitosano come possibile strumento per la riduzione dell'utilizzo di anidride solforosa nei vini: un focus sulle sue caratteristiche antiossidanti

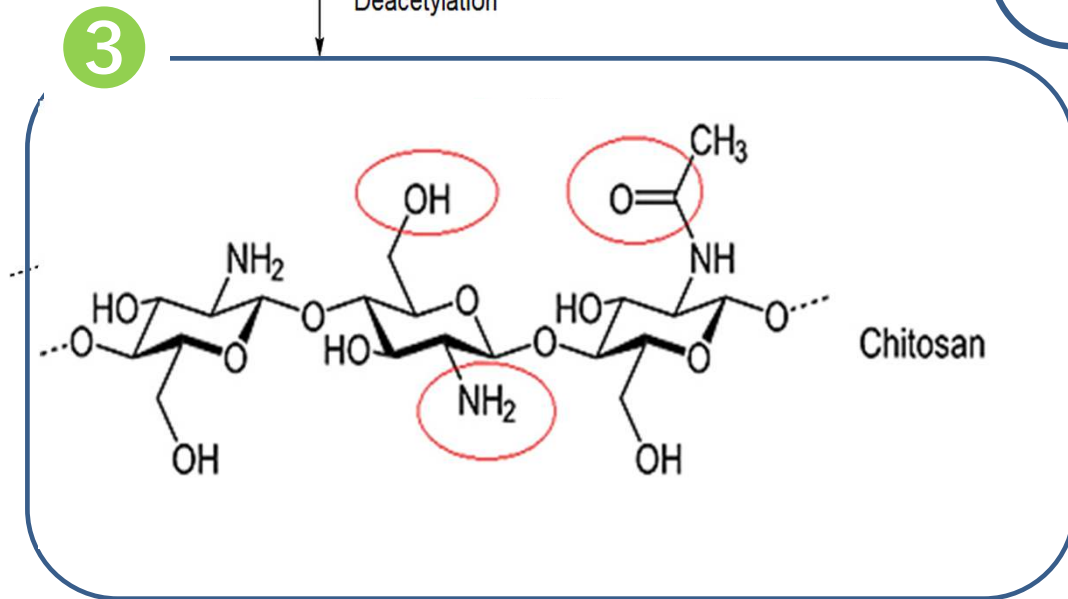
Fabio Chinnici

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COSA E' IL CHITOSANO



Deacetylation



Chitosano come antimicrobico

FIELD OF USE (OIV code sheets 336-7-8 A/2009)	TARGET	MATRIX	TESTED DOSE (mg/L)	EFFECT
Reduction of undesirable microorganisms	Yeasts: <i>Brettanomyces spp.</i>	Red wine	150–1500	MIC and MLC varying from 150 to 500 mg/L depending on <i>Brettanomyces/Dekkera</i> strain and KT molecular weight.
OIV maximum recommended dose: 100 mg/L		Red wine in barrique	40–100	At the tested doses, KT demonstrated a fungistatic activity.
			40	Fungistatic activity (prevention of the yeast development during elevelage). Batonnage promoted yeast cell recovery.
		Glucose, salts, and yeast extracts	100–6000	Lag-phase lengths >80 h. No biocidal activity against <i>B. bruxellensis</i> . Complete inhibition of <i>B. intermedius</i> at 2 g/L
		YPG culture medium	0.12–250	MIC ₉₀ = 62 mg/L and MBC ₉₀ > 250 mg/L for 16 strains
		Inoculum <i>in vitro</i>	40–400	Fungistatic activity (physical and biological effects on <i>Brettanomyces</i> cells).
	<i>H. uvarum</i> and <i>Z. bailii</i> <i>S. cerevisiae</i>	YPG culture medium	100–400	Growth inhibition at 0.3 or 0.4 g/L.
		Inoculum <i>in vitro</i>	1000–6000	Increase of the lag phase from 0 to 4 h depending on the concentration of KT.
		Apple juice	8–1000 100 and 400	Minimum biocidal concentration >250 mg/L Lag-phase extended by 2–3 days depending on the strains. Then, growth was recovered at levels similar to untreated samples
		YPG culture medium	0.12–250	MIC ₅₀ > 250 mg/L for 15 different strains.
			600–2000	Ethanol enhance yeast sensitivity Lag phase increase from 2 to 4 days depending on the concentration.
		Apple- elderflower juice	300	Better growth inhibition in juice (pH 3.3) than saline solution (pH 6.4)
	Lactic acid Bacteria Unspecified LAB	Apple-elderflower juice	300	Initial quick reduction in viable cells followed by restored growth after 8 days
	<i>O. oeni</i> ; <i>L. hilgardii</i>	YPG culture medium	200–2000	Complete inactivation at 200 mg/L for at least 6 days.
	<i>Pediococcus sp.</i>	Peptone water/Hopped malt extract	10–1000	In peptone water, growth was completely inhibited at 100 mg/L. In malt extract, the activity was only bacteriostatic.
	<i>L. plantarum</i>	YPG culture medium MRS agar	200–2000 30–1000	Complete inactivation at 1200 mg/L for at least 6 days. <i>L. plantarum</i> was the most resistant LAB. MIC = 500–800 mg/L depending on KT deacetylation degree
		Peptone water/Hopped malt extract	10–1000	Only 1000 g/L prevented the development of bacteria. The effect of the pH and the matrix were also evaluated.
	Acetic acid Bacteria: <i>A. malorum</i> and <i>A. pasteurianus</i>	Synthetic vinegar	200	Growth inhibition (reduction of <i>Acetobacter spp.</i> activity; effects comparable to 60 mg/L of SO ₂).

REVIEW

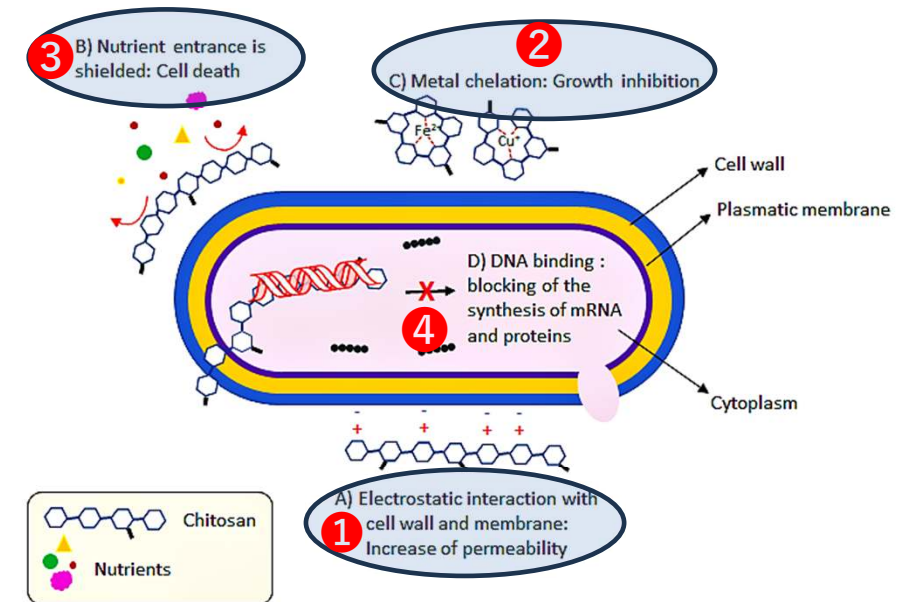


Relevance and perspectives of the use of chitosan in winemaking: a review

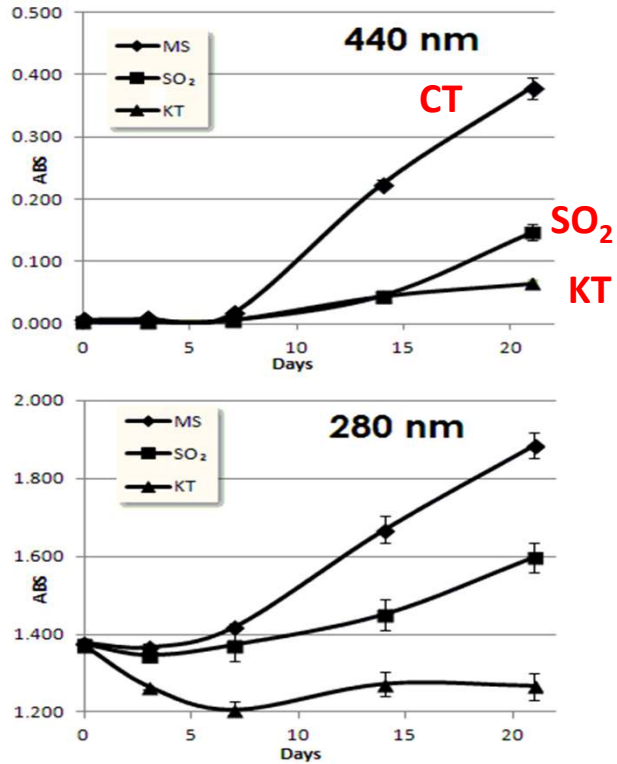
Antonio Castro Marin^a, Donato Colangelo^b, Milena Lambri^b, Claudio Riponi^a, and Fabio Chinnici^b

^aDepartment of Agricultural and Food Sciences, University of Bologna, Bologna, Italy; ^bDipartimento di Scienze e Tecnologie Alimentari per una filiera agro-alimentare Sostenibile – DISTAS, Università Cattolica del Sacro Cuore, Piacenza, Italy

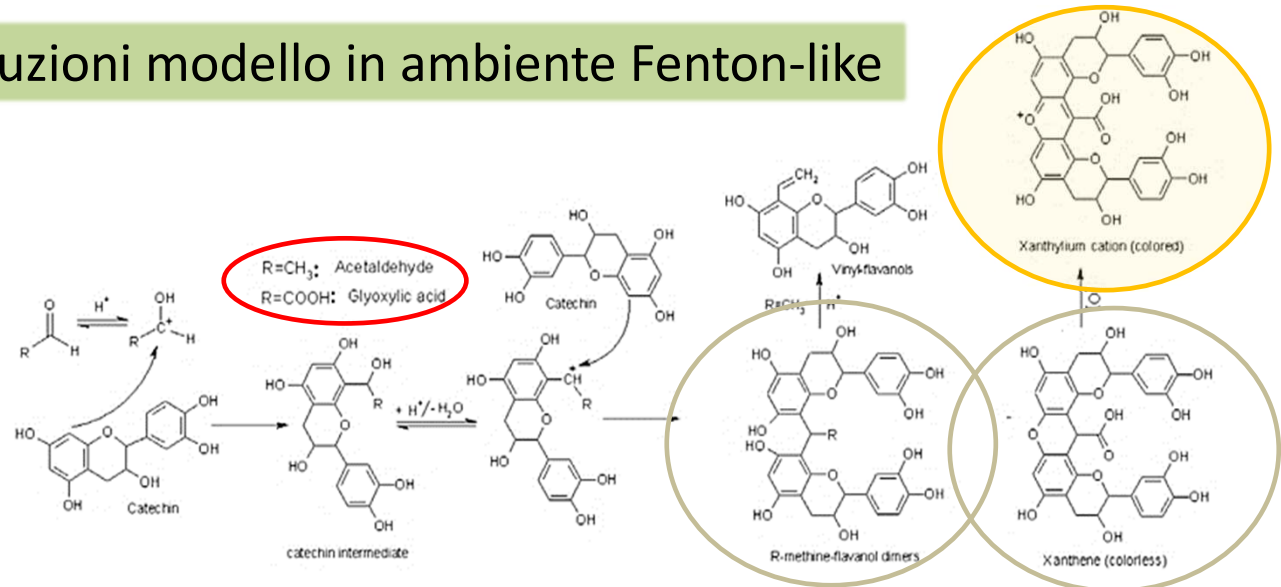
Meccanismi di azione proposti



Attività anti-imbrunimento del chitosano



Soluzioni modello in ambiente Fenton-like



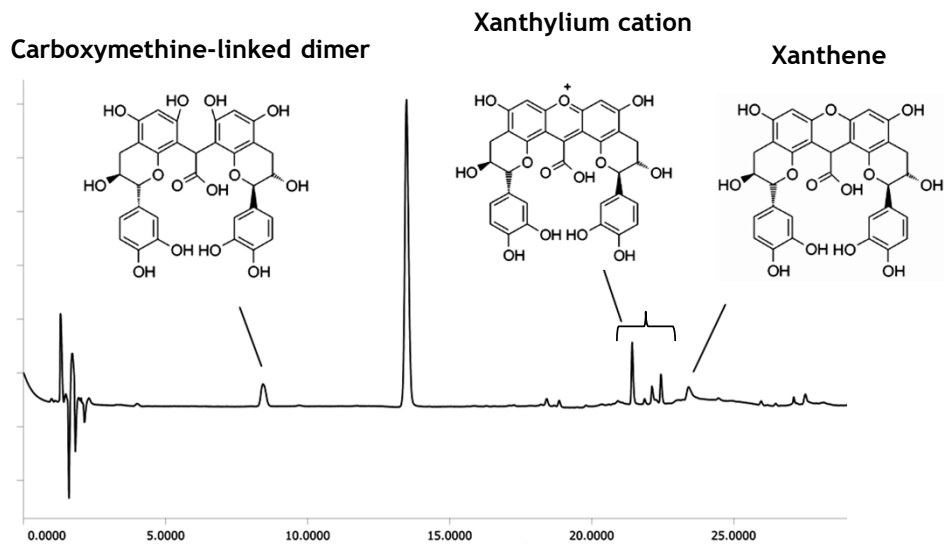
Formazione di composti bruni a partire dalla (+)-catechina



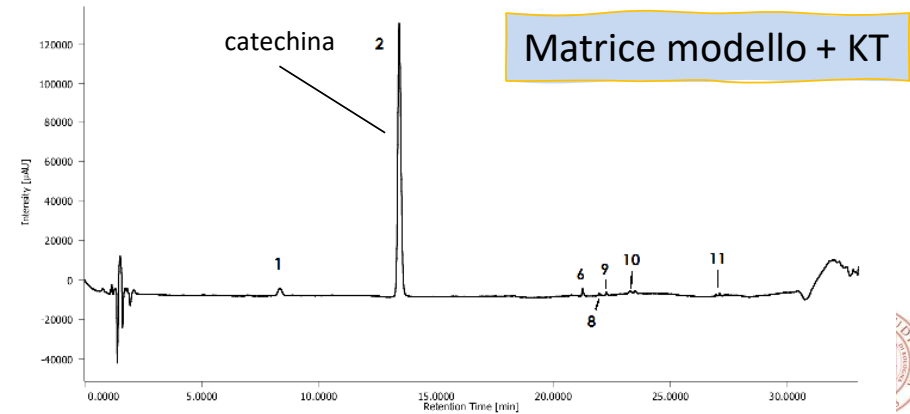
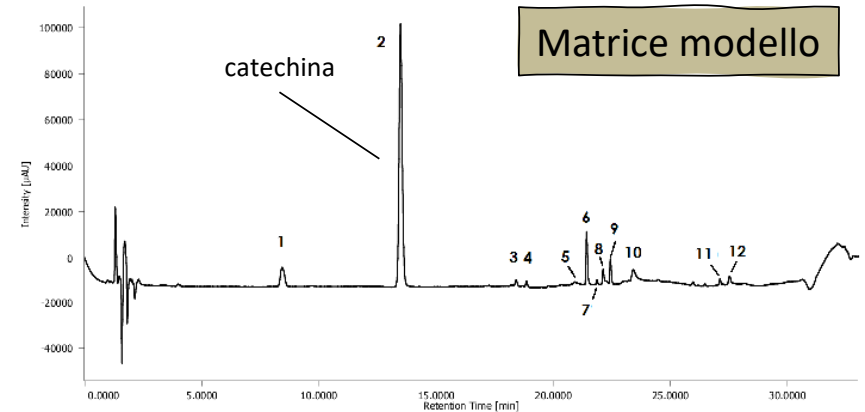
Imbrunimento delle soluzioni modello in presenza di SO₂ o chitosano

Attività anti-imbrunimento del chitosano

Composti di ossidazione della catechina nelle tesi dopo 20 giorni



Cromatogramma relativo ai composti di ossidazione della catechina



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Article
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Efficacy of Chitosan in Inhibiting the Oxidation of (+)-Catechin in White Wine Model Solutions

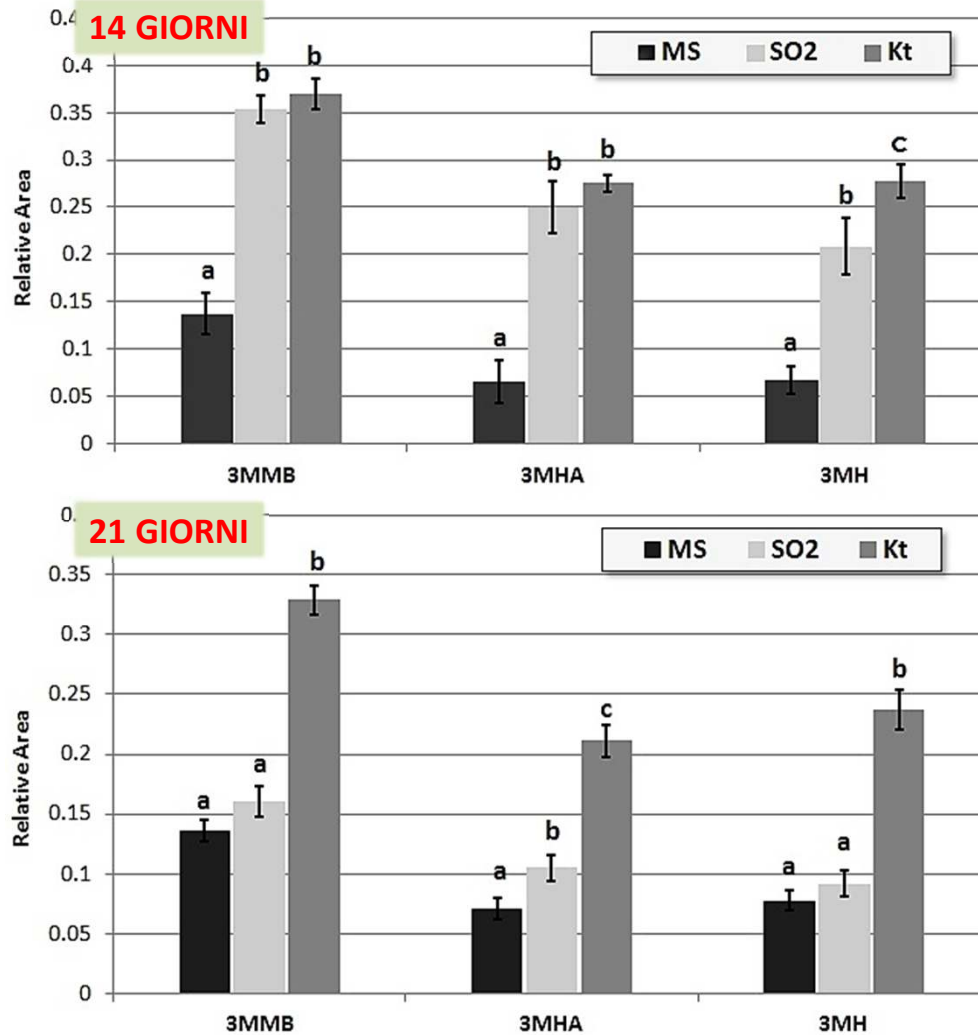
Fabio Chinnici,* Nadia Natali, and Claudio Riponi

Department of Agricultural and Food Sciences, Alma Mater Studiorum, University of Bologna, Piazza Goianich, 60, 47521 Cesena, Forlì-Cesena, Italy

AIVV – Tornata sul Lambrusco, Modena 15-16 Dicembre 2023

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Attività del chitosano nei confronti di molecole tioliche

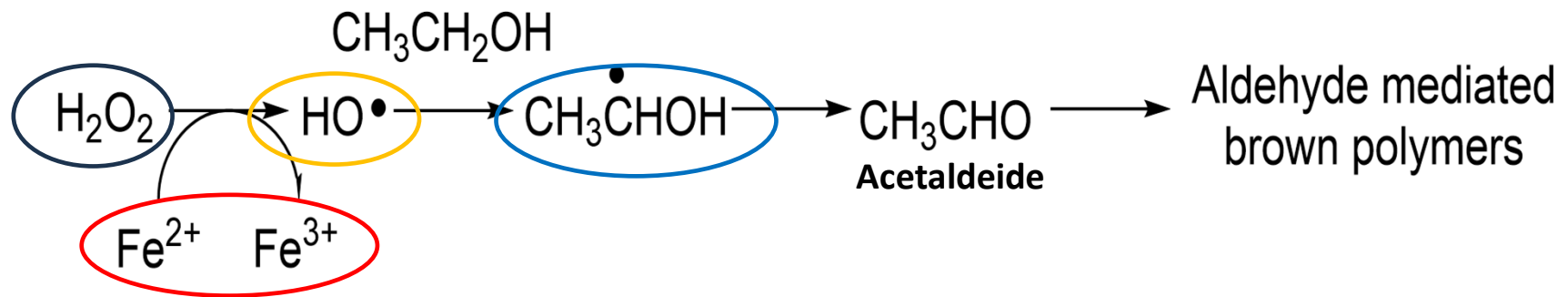


Quantità residua di tioli (3-mercaptopetilbutanolo, 3-mercaptopetilacetato e 3-mercaptopetilacetato) nelle soluzioni dopo 14 e 21 giorni di ossidazione



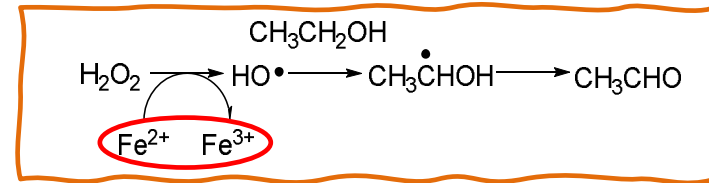
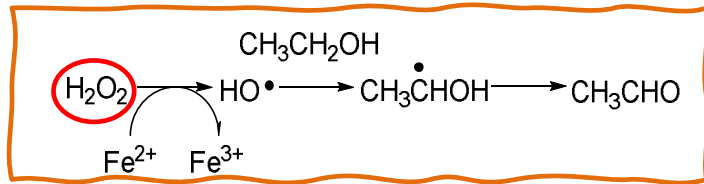
Studio dei meccanismi legati all'attività antiossidante del chitosano:

La reazione di Fenton nella matrice vino



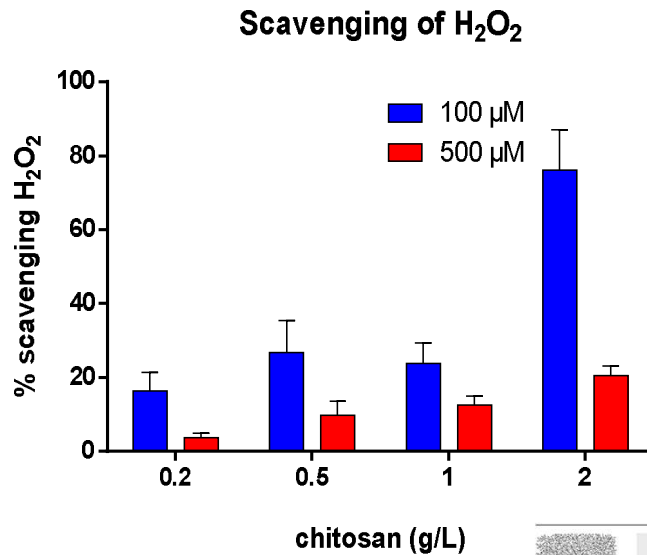
**Molecole coinvolte nelle reazioni di ossidazione chimica dei vini
mediata dalla cinetica di Fenton**

Studio dei meccanismi legati all'attività antiossidante del chitosano: Reattività del chitosano nei confronti delle specie chimiche coinvolte nelle reazione di Fenton

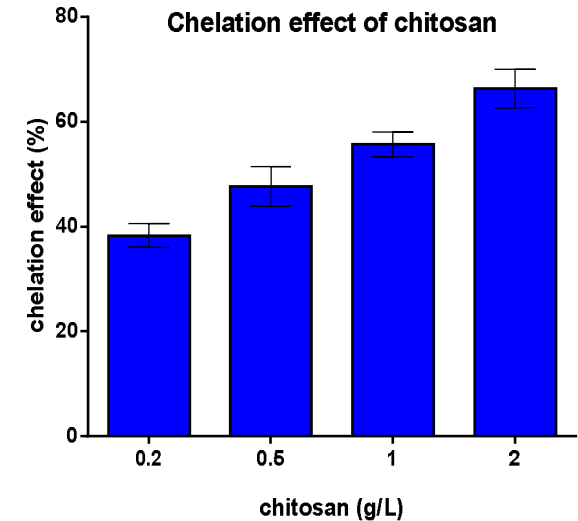


L'esperienza:
20mL di vino modello
+
100/500 μM H_2O_2
+
Chitosano (0.2-2g/L)
+
30 min of incubation

 H_2O_2 quantificazione
fluorimetrica
(Lucigenina)



L'esperienza:
20mL di vino
+
2,5 mg/L Fe (II)
+
Chitosano (0,2-2g/L)
+
48 h of incubation
+
Spettrometria ad
assorbimento di fiamma



- Idrolisi del chitosano dovuta a H_2O_2



Chitosan as an antioxidant alternative to sulphites in oenology: EPR investigation of inhibitory mechanisms

Antonio Castro Marín^{a,b}, Marcel Culcasi^{a*}, Mathieu Cassien^a, Pierre Stocker^a, Sophie Thétiot-Laurent^a, Bertrand Robillard^c, Fabio Chinnici^b, Sylvia Pietri^a



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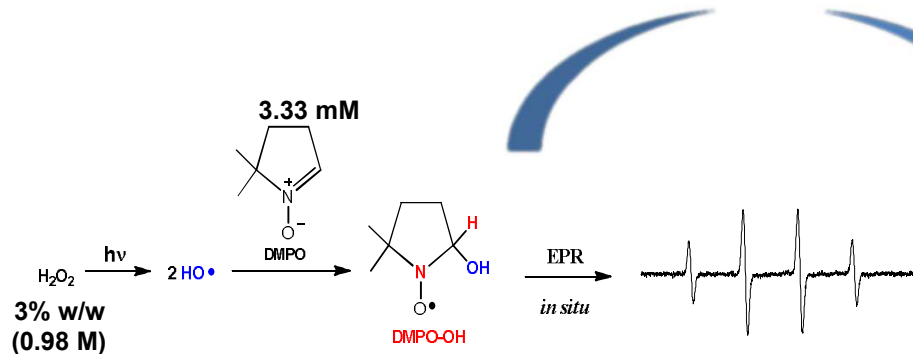
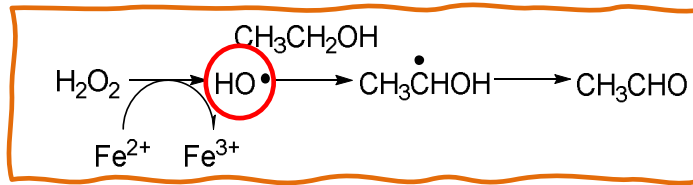
Studio dei meccanismi legati all'attività antiossidante del chitosano:

Reattività del chitosano nei confronti delle specie chimiche coinvolte nelle reazioni di Fenton

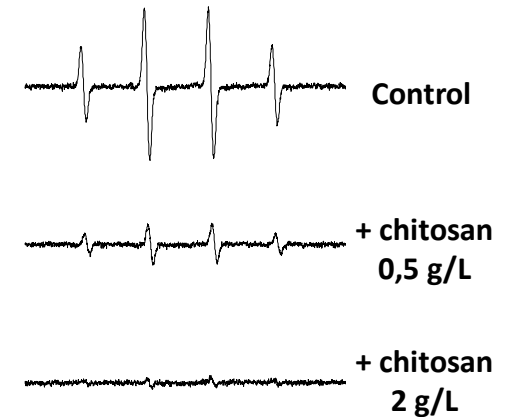
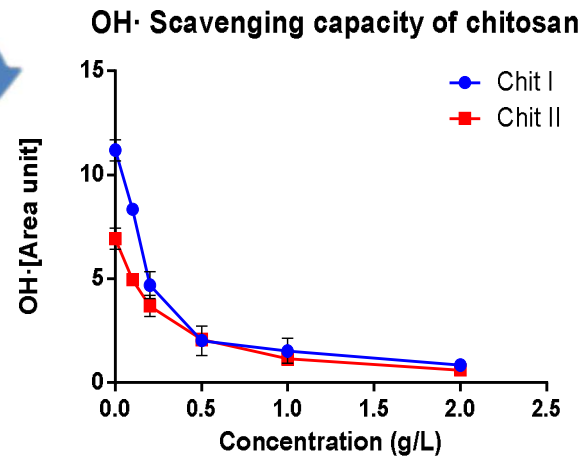


Chitosan as an antioxidant alternative to sulphites in oenology: EPR investigation of inhibitory mechanisms

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Generazione fotolitica di OH^\bullet e detezone mediante EPR

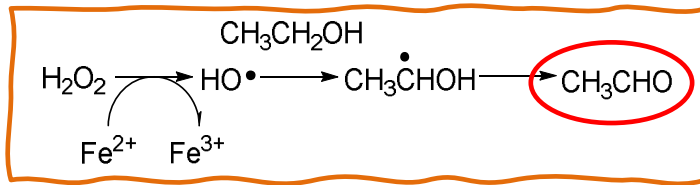


- Quenching diretto di HO^\bullet
- Dose-dipendente



Studio dei meccanismi legati all'attività antiossidante del chitosano:

Reattività del chitosano nei confronti delle specie chimiche coinvolte nelle reazioni di Fenton

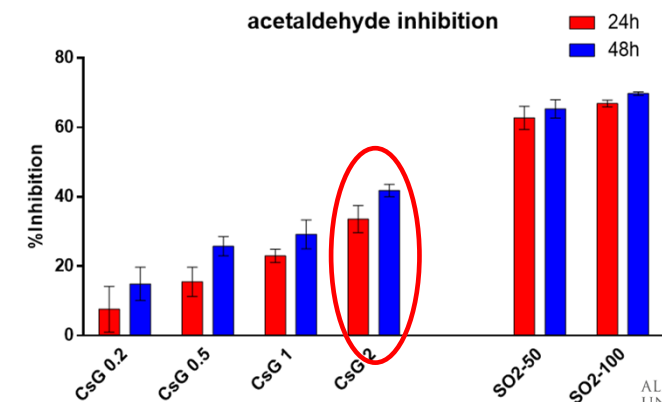
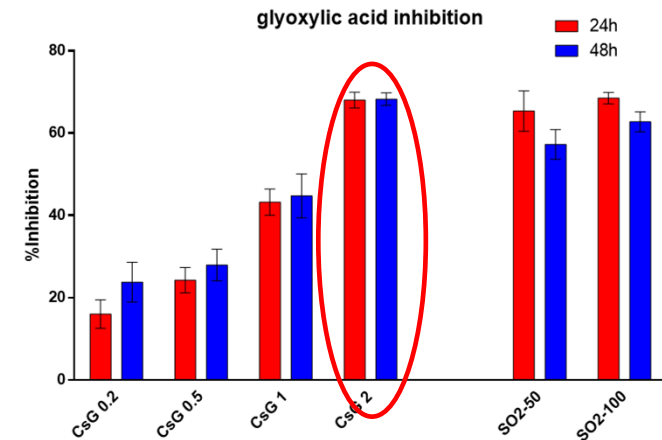


Inhibitory effect of fungoid chitosan in the generation of aldehydes relevant to photooxidative decay in a sulphite-free white wine

Antonio Castro Marin^{a,b}, Pierre Stocker^a, Fabio Chinnici^b, Mathieu Casjen^{b,c}, Sophie Th  t  t-Laurent^a, Nicolas Vidal^{b,c}, Claudio Riponi^b, Bertrand Robillard^d, Marcel Culcasi^{b,c}, Sylvia Pietri^a



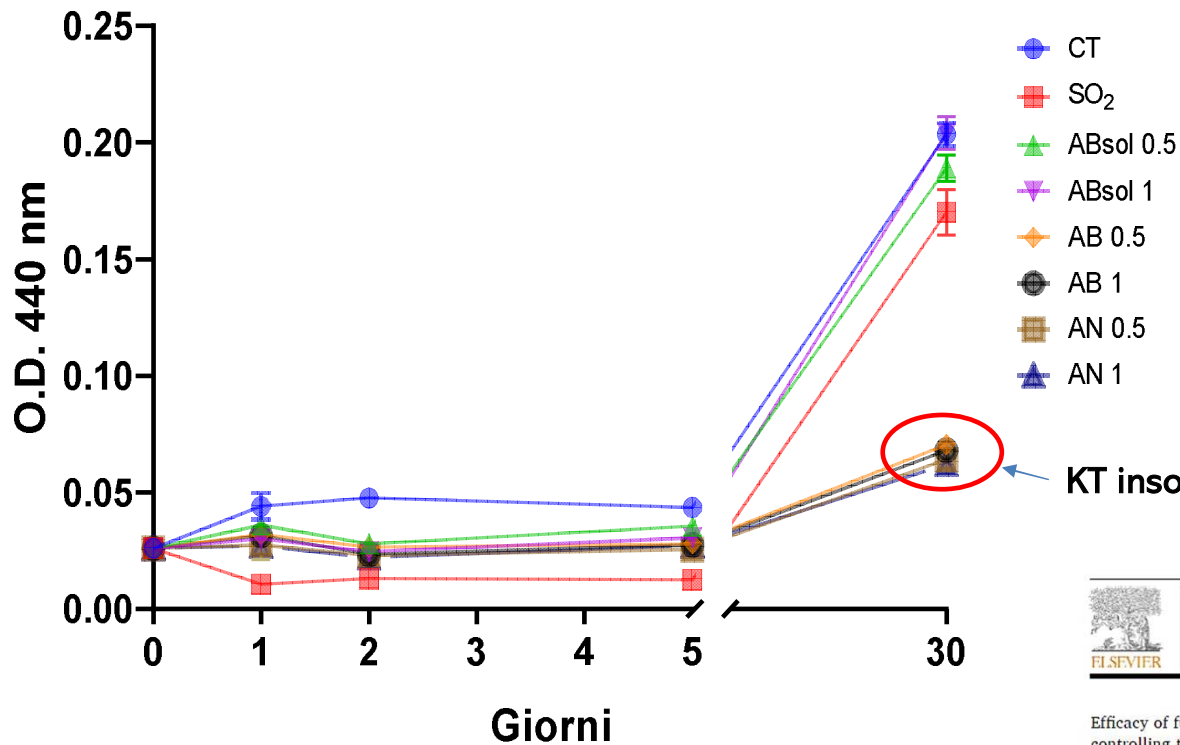
Incubazione:
 20 ml vino/vino modello + 2,5 mg/L Fe(II)
 +
 Chitosano (0.2-2g/L)
 SO₂ (50-100mg/L)



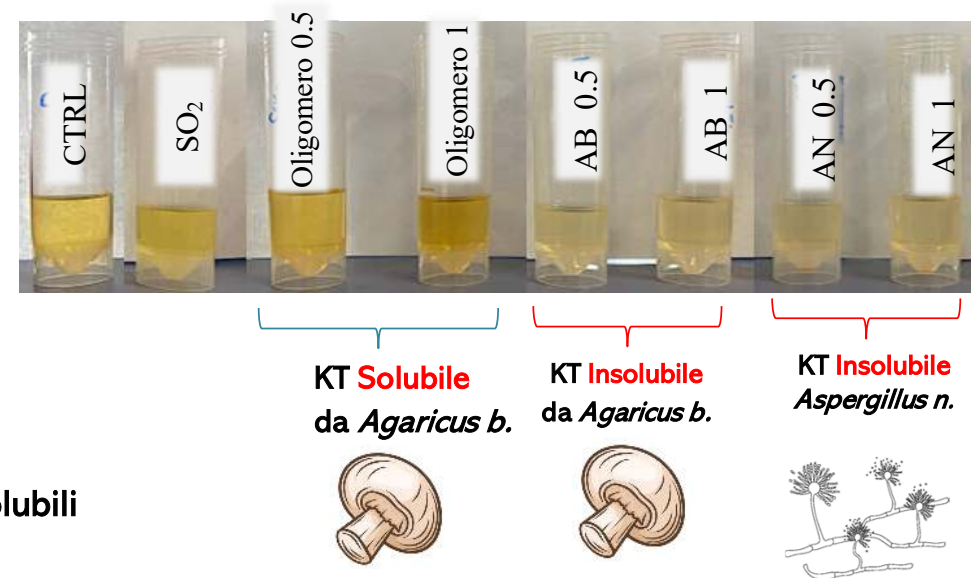
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Comparazione dell'efficacia anti-imbrunimento di chitosani da *Aspergillus niger* e *Agaricus Bisporus*:

Evoluzione dell'imbrunimento nelle soluzioni durante 30 giorni



Imbrunimento delle soluzioni modello dopo 30 giorni di ossidazione



KT insolubili



Efficacy of fungoid chitosans from *Aspergillus niger* and *Agaricus bisporus* in controlling the oxidative browning of model white wines

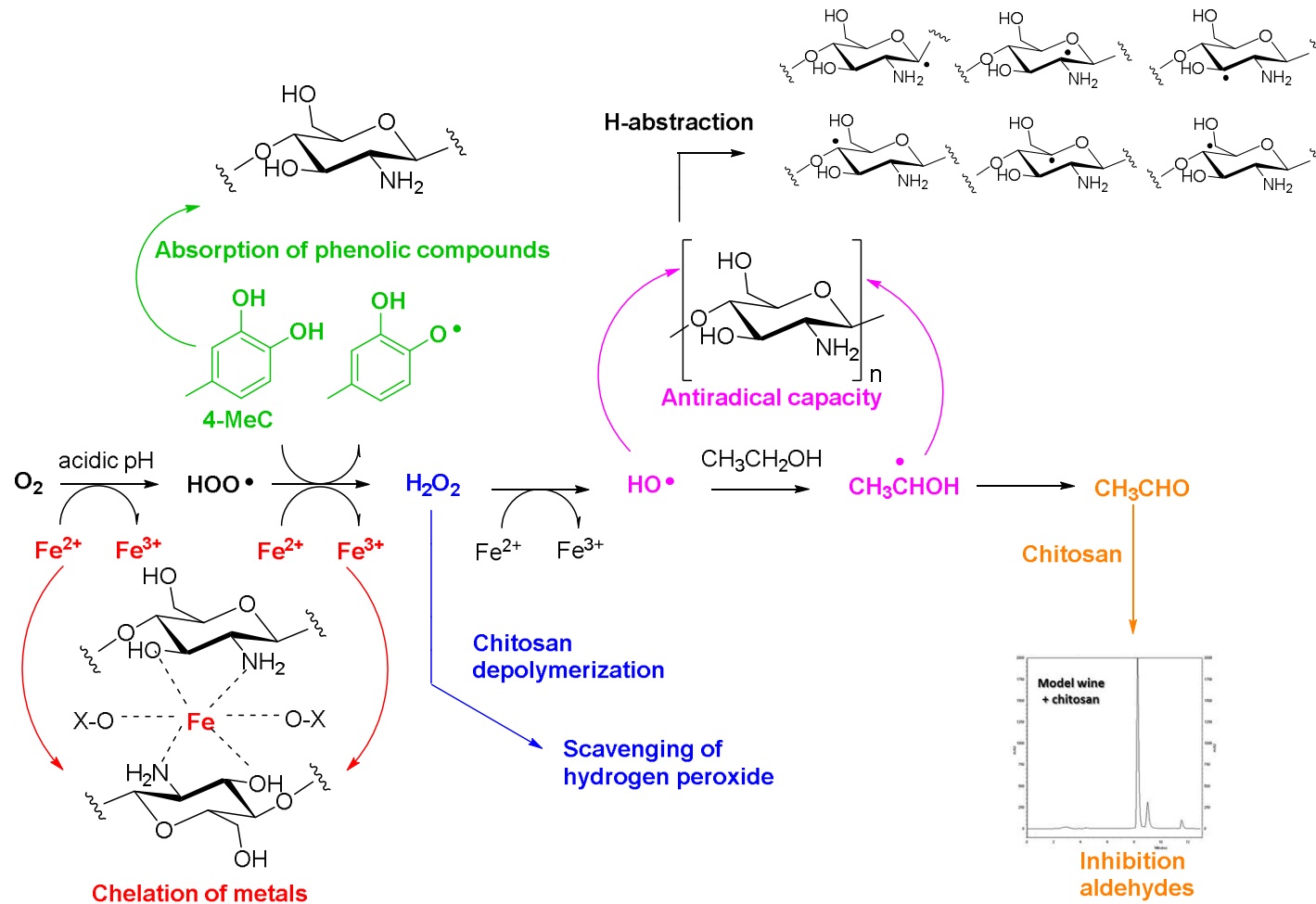
Federico Baris, Antonio Castro Marín, Ana Cristina De Aguiar Saldanha Pinheiro, Silvia Tappi, Fabio Chinnici



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Studio dei meccanismi legati all'attività antiossidante del chitosano:

Schema riassuntivo dei meccanismi antiossidanti dimostrati per il chitosano

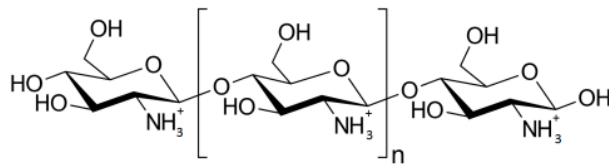


Prove tecnologiche: Vini Bianchi

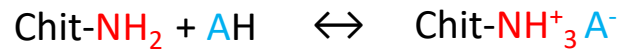


Impatto del chitosano sul quadro acido di mosti e vini bianchi

		Tartaric	Piruvic	Malic	Shikimic	Citric	Lactic	Acetic	Succinic
Stabilization of must (Objective 1)	KT	3.52 ^b	n.d.	1.56 ^b	4.38 ^a	0.27 ^b	n.d.	n.d.	n.d.
	SO ₂	3.80 ^a	n.d.	1.74 ^a	5.00 ^a	0.36 ^a	n.d.	n.d.	n.d.
Stabilization of wine (Objective 2)	KT	2.45 ^b	190.80 ^b	1.25 ^b	5.95 ^b	0.23 ^b	0.22 ^b	0.15 ^a	0.40 ^a
	SO ₂	2.80 ^a	306.90 ^a	1.47 ^a	8.49 ^a	0.30 ^a	0.31 ^a	0.17 ^a	0.47 ^a



pKa Chitosan: 6.2



CONSEGUENZA: aumento del pH pari a 0.08-0.12 unità, nei vini aggiunti di KT



Volatile and fixed composition of sulphite-free white wines obtained after fermentation in the presence of chitosan

Antonio Castro-Marín, Ana Gabriela Buglia, Claudio Riponi, Fabio Chinnici*

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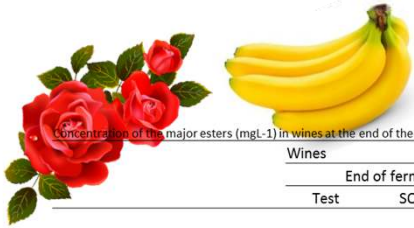


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Prove tecnologiche: Vini Bianchi



Impatto del chitosano sulla componente volatile di vini bianchi



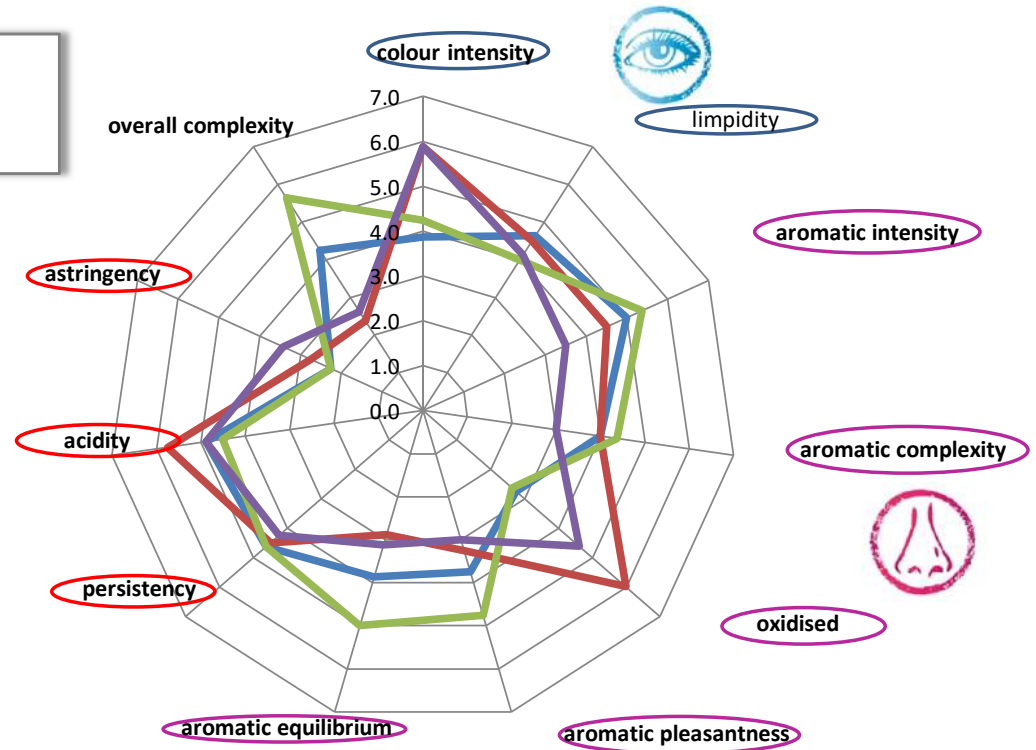
Concentration of the major esters (mgL-1) in wines at the end of the alcoholic fermentation and after 1 year of storage.

Wines	End of fermentation			1 year of storage		
	Test	SO2	KT	Test	SO2	KT
Total esters	17.31 ^a	18.25 ^a	14.21 ^b	72.58 ^a	86.16 ^a	81.88 ^a
isoamyl acetate	0.77 ^b	0.69 ^b	1.11 ^a	0.21 ^a	0.22 ^a	0.20 ^a
ethyl hexanoate	0.23 ^b	0.21 ^b	0.52 ^a	0.34 ^b	0.31 ^b	0.60 ^a
ethyl pyruvate	0.04 ^b	0.06 ^a	0.05 ^b	0.11 ^b	0.17 ^a	0.08 ^b
methyl lactate	0.02 ^b	0.03 ^b	0.05 ^a	n.d.	n.d.	n.d.
ethyl lactate	0.51 ^b	0.53 ^a	0.42 ^c	1.65 ^a	1.44 ^b	1.46 ^b
ethyl octanoate	0.15 ^b	0.16 ^b	0.43 ^a	0.64 ^b	0.50 ^b	1.15 ^a
ethyl-3-hydroxybutyrate	0.05 ^b	0.06 ^b	0.10 ^a	0.05 ^b	0.09 ^a	0.09 ^a
ethyl decanoate	0.03 ^b	0.04 ^b	0.15 ^a	0.11 ^b	0.09 ^b	0.34 ^a
diethyl succinate	0.34 ^a	0.39 ^a	0.27 ^b	13.33 ^{a,b}	15.56 ^a	9.35 ^b
methyl salicylate	0.01 ^a	0.01 ^a	0.01 ^a	n.d.	n.d.	n.d.
ethyl 4-hydroxybutanoate	2.93 ^b	3.47 ^a	1.31 ^c	0.23 ^{a,b}	0.30 ^a	0.19 ^b
B-phenylethyl acetate	0.33 ^b	0.32 ^b	0.75 ^a	0.07 ^b	0.08 ^b	0.15 ^a
diethyl hydroxybutanedioate	0.26 ^a	0.31 ^a	0.17 ^b	0.04 ^a	0.01 ^a	0.07 ^a
diethyl malate	n.d.	n.d.	n.d.	6.26 ^b	9.75 ^a	6.45 ^b
diethyl tartrate	n.d.	n.d.	n.d.	0.58 ^b	1.00 ^a	0.35 ^b
ethyl hydrogen succinate	11.65 ^a	11.96 ^a	8.87 ^b	48.97 ^a	56.65 ^a	61.40 ^a

Maggior contenuto in acidi grassi a media catena

Concentrations of the major acids (mgL-1) in wines at the end of the alcoholic fermentation and after 1 year of storage.

Wines	End of fermentation			1 year of storage		
	Test	SO2	KT	Test	SO2	KT
Total acids	8.90 ^a	8.83 ^b	13.04 ^a	7.72 ^b	7.70 ^b	11.69 ^a
isobutyric acid	1.12 ^a	1.02 ^a	0.53 ^b	0.95 ^a	0.81 ^a	0.41 ^b
n-butyric acid	0.31 ^b	0.34 ^b	0.39 ^a	0.21 ^c	0.29 ^b	0.33 ^a
pentanoic acid	1.91 ^a	1.90 ^a	1.07 ^c	1.86 ^a	1.85 ^a	0.87 ^b
hexanoic acid	1.42 ^b	1.46 ^b	2.43 ^a	1.39 ^b	1.43 ^b	2.57 ^a
octanoic acid	3.11 ^b	3.11 ^b	5.67 ^a	2.65 ^b	2.69 ^b	5.45 ^a
decanoic acid	0.75 ^c	0.63 ^c	2.74 ^a	0.58 ^b	0.51 ^b	1.93 ^a
dodecanoic acid	0.16 ^b	0.17 ^b	0.14 ^b	0.03 ^b	0.04 ^b	0.08 ^a
benzenoic acid	0.12 ^b	0.20 ^a	0.07 ^c	0.05 ^b	0.10 ^a	0.06 ^b



Volatile and fixed composition of sulphite-free white wines obtained after fermentation in the presence of chitosan

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Prove tecnologiche: Vini Rossi



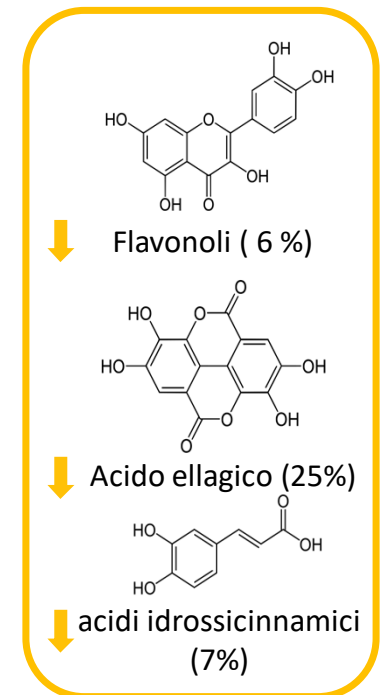
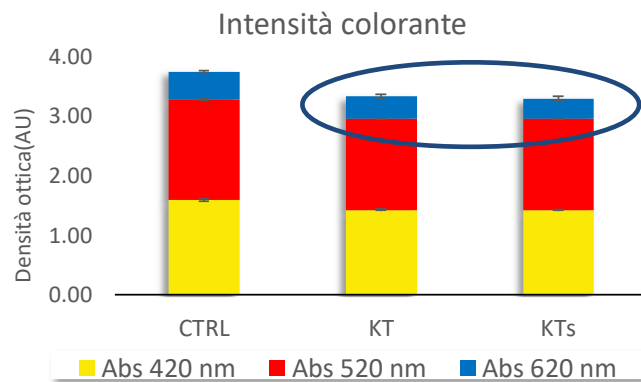
Impatto del chitosano sulla componente fenolica di vini rossi

Dotazione acidica e polifenolica dei vini aggiunti di KT (0.5 g/L)

	C			KT		
pH	3.65	± 0.01	b	3.70	± 0.01	a
Titrateable acidity (g/L)	5.89	± 0.13	a	5.61	± 0.09	b
TPI (GAE) (mg/L)	1286	± 17.43	a	1179	± 19.23	b

Molecole interessate dalla diminuzione dopo aggiunta di KT (0.5 g/L)

Intensità colorante dei vini aggiunti di KT (0.5 g/L)



Article
Physico-Chemical Features of Sangiovese Wine as Affected by a Post-Fermentative Treatment with Chitosan

Antonio Castro Marin * and Fabio Chinnici

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Prove tecnologiche: Vini Rossi



Impatto del chitosano sulla componente fenolica di vini rossi

Meccanismo d'azione di chitosano fungino



Co-pigmenti



Adsorbimento idrossicinnamici, acido ellagico, flavonoli



Leggera riduzione del colore

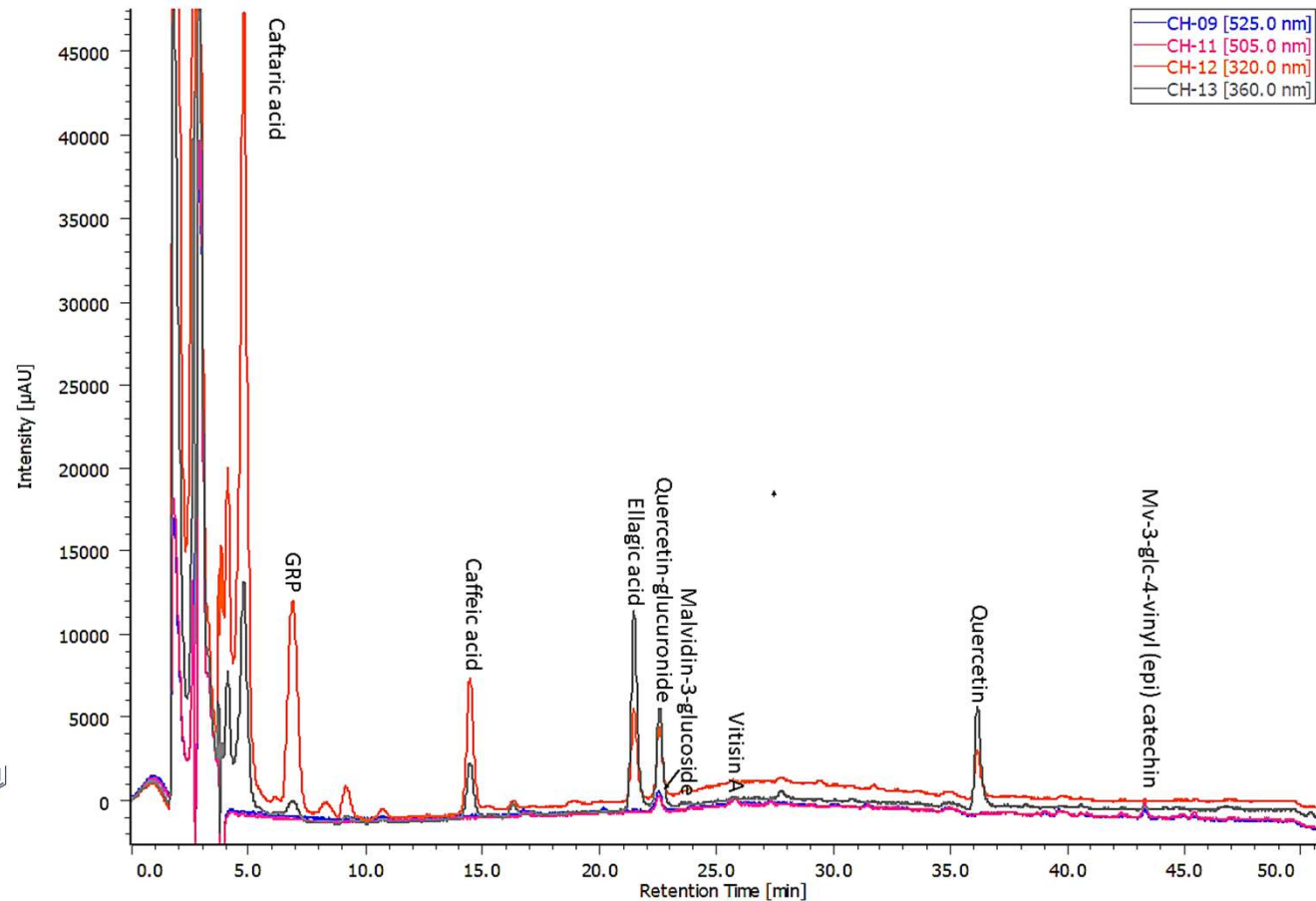


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Cromatogramma dei fenoli adsorbiti dal chitosano

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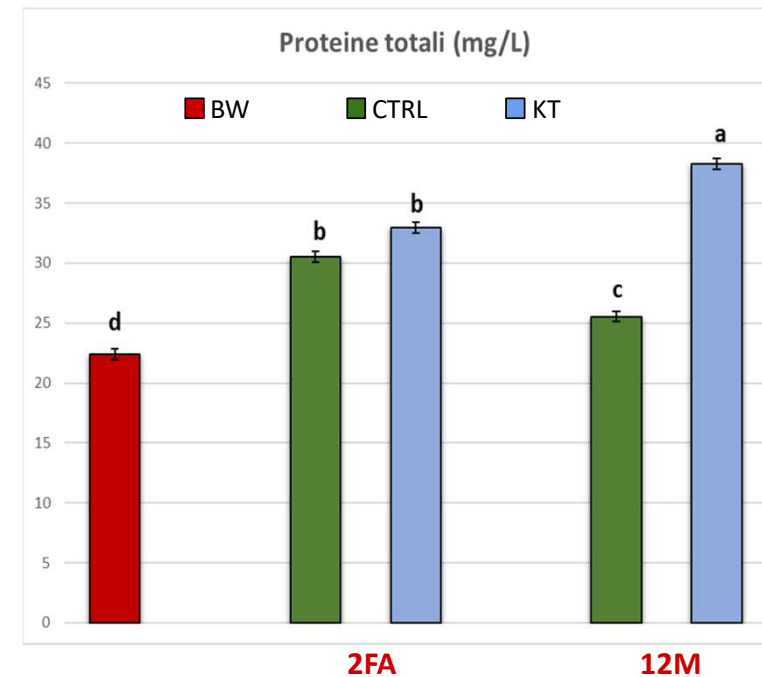
Prove tecnologiche: Vini Spumanti



Impatto del chitosano aggiunto durante la presa di spuma in metodo classico (12 mesi su feccia)

Parametri compositivi dei vini spumanti

	Base Wine	2nd AF		12 Months "Sur lie"	
		CTRL	KT	CTRL	KT
Titrate acidity (g/L)	5.75 ± 0.07 ^{ab}	5.90 ± 0.14 ^a	5.85 ± 0.07 ^a	5.59 ± 0.06 ^{ab}	5.51 ± 0.01 ^b
pH	3.10 ± 0.01 ^a	3.10 ± 0.01 ^a	3.10 ± 0.02 ^a	3.11 ± 0.01 ^a	3.12 ± 0.01 ^a
Volatile acidity (g/L)	0.29 ± 0.02 ^a	0.28 ± 0.01 ^a	0.30 ± 0.01 ^a	0.32 ± 0.01 ^a	0.28 ± 0.01 ^a
Alcohol (%v/v)	10.32 ± 0.71 ^b	11.42 ± 0.09 ^a	11.36 ± 0.19 ^a	11.40 ± 0.06 ^a	11.30 ± 0.03 ^a
Optical Density 420 nm	0.092 ± 0.001 ^a	0.093 ± 0.01 ^a	0.089 ± 0.012 ^a	0.101 ± 0.001 ^a	0.104 ± 0.001 ^a
TPI (GAE)	172.3 ± 0.02 ^{ab}	162.8 ± 0.07 ^b	180.4 ± 0.28 ^a	177.1 ± 0.03 ^{ab}	176.9 ± 0.03 ^{ab}
Total proteins (mg/L)	22.43 ± 0.45 ^d	30.53 ± 1.28 ^b	32.99 ± 1.37 ^b	25.55 ± 0.85 ^c	38.25 ± 1.01 ^a



Aumento del 35% delle proteine in soluzione, al termine della sosta su feccia

Prove tecnologiche: Vini Spumanti

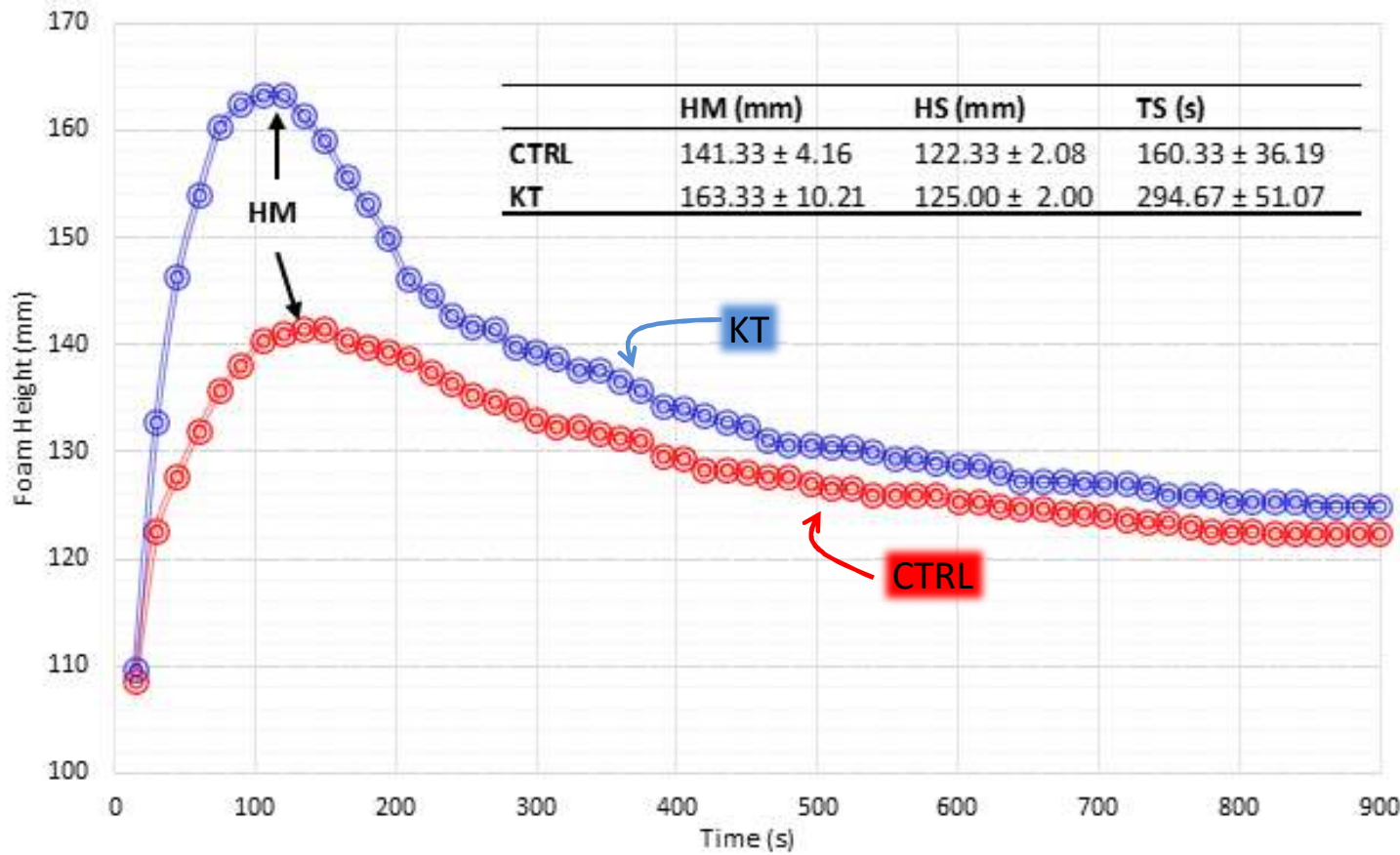
Impatto del chitosano aggiunto durante la presa di spuma in metodo classico



Article
Chitosan in Sparkling Wines Produced by the Traditional Method: Influence of Its Presence during the Secondary Fermentation

Antonio Castro Marin, Claudio Riponi and Fabio Chinnici*
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Caratteristiche della schiuma ottenute mediante tecnica MOSALUX

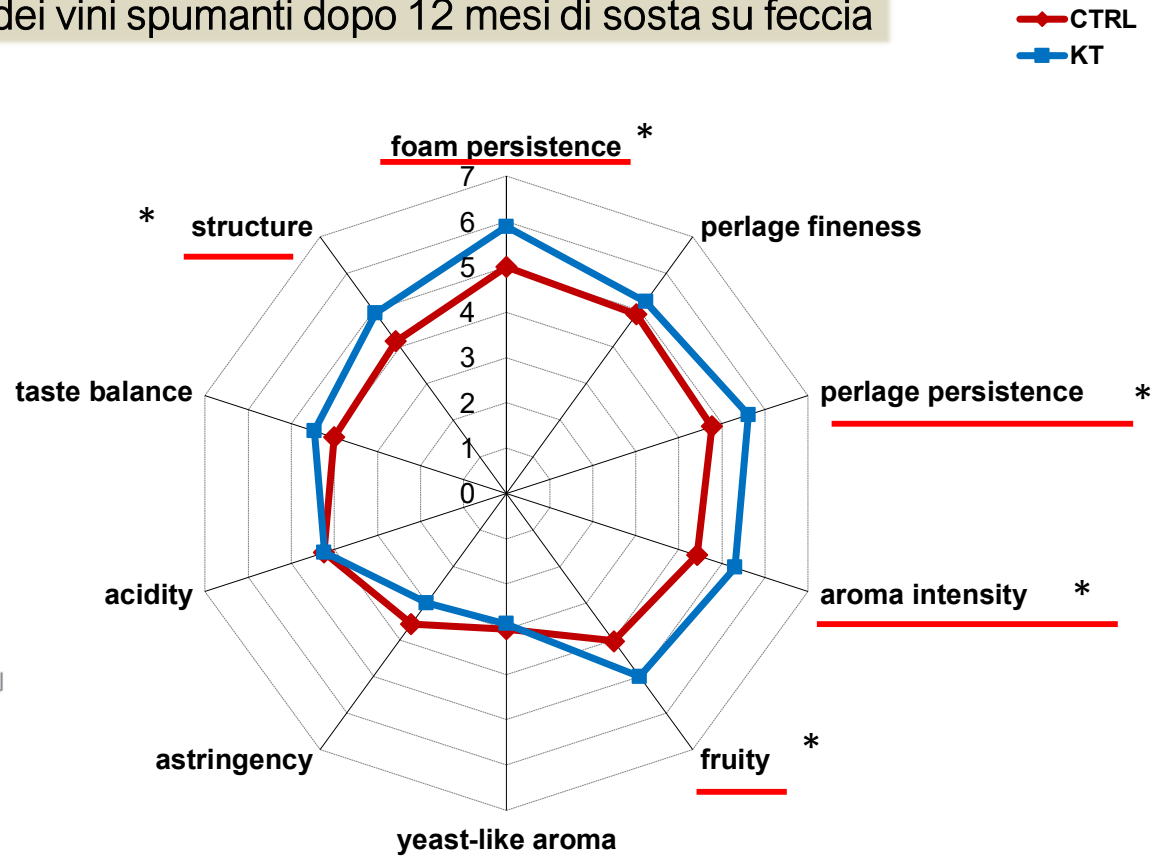


Prove tecnologiche: Vini Spumanti



Impatto del chitosano aggiunto durante la presa di spuma in metodo classico

Test sensoriale dei vini spumanti dopo 12 mesi di sosta su feccia



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Il chitosano come possibile strumento per la riduzione dell'utilizzo di anidride solforosa nei vini: un focus sulle sue caratteristiche antiossidanti

Grazie per l'attenzione

Dipartimento di Scienze e Tecnologie Agroalimentari



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Un ringraziamento particolare va ai coautori...

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