



Master Brewers Conference

Free thiol release from cysteinylated & glutathionylated sulfanylalkyl alcohols in beer: modulation of sulfanylalkyl acetate/ alcohol ratio by selecting yeast strain & fermentation conditions

P-010

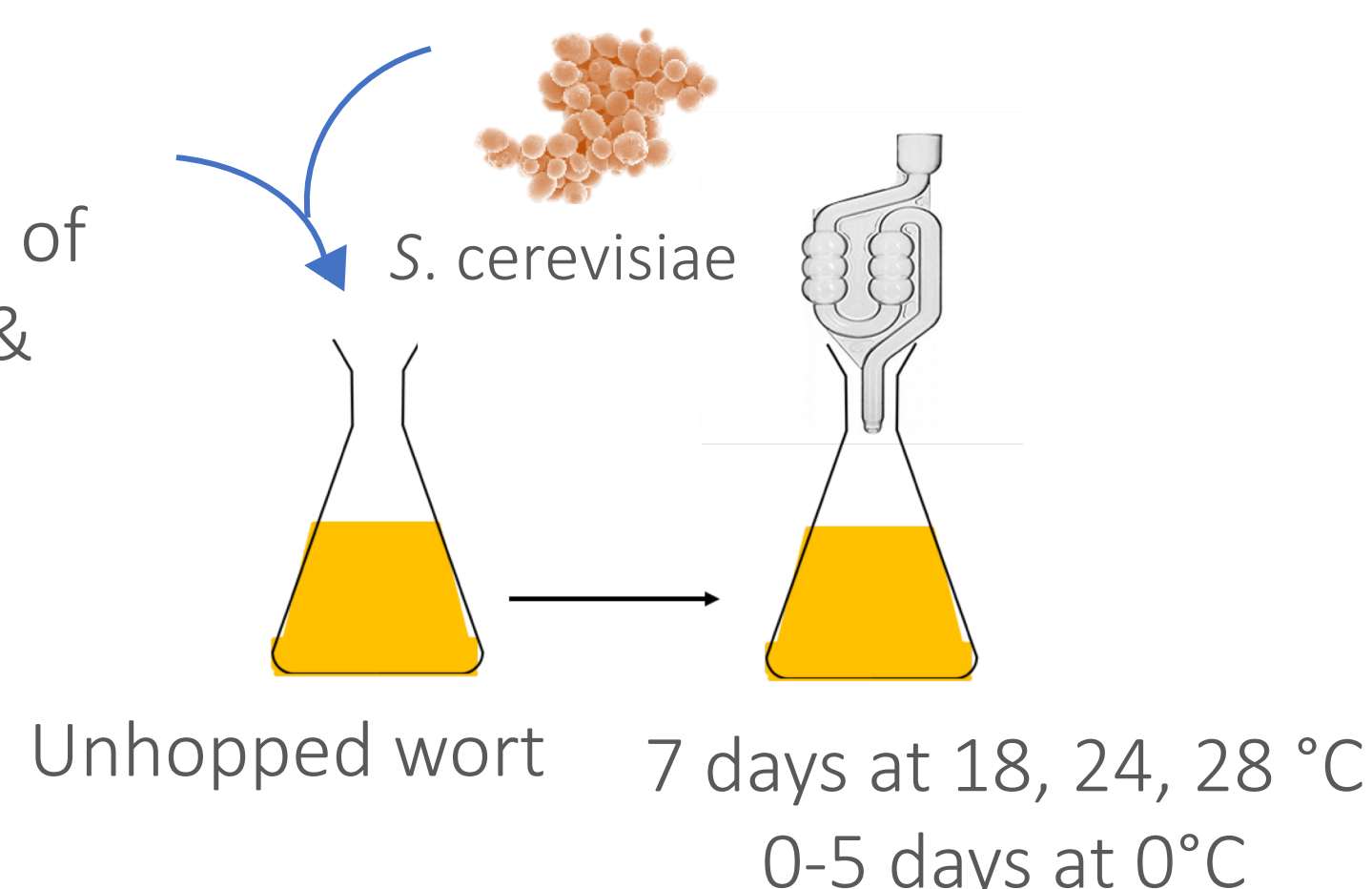
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Introduction : The occurrence of a substantial pool of cysteinylated (Cys-) and glutathionylated (G-) forms of polyfunctional thiols has been evidenced for several dual-purpose hop varieties (Cys- and G- 3-sulfanyl-hexanol (3SHol) and 3-sulfanyl-pentanol (3SPol) ubiquitous to all studied varieties, Cys- and G- 3-sulfanyl-4-methylpentanol (3S4MPol) peculiar to some varieties). In the brewing field, the ability of yeast to hydrolyze cysteinylated adducts was first confirmed after bottle refermentation. Very recently, the ability of brewing yeast to release free PFTs from both Cys- and G-adducts, using synthesized adducts, was confirmed, in primary fermentation. The present work aimed to investigate the effect of temperature, wort density, maturation time, and strain on the efficiency of sulfanylalkyl alcohols release and sulfanylalkyl acetates formation from Cys- or G- adducts by *S. cerevisiae* yeasts.

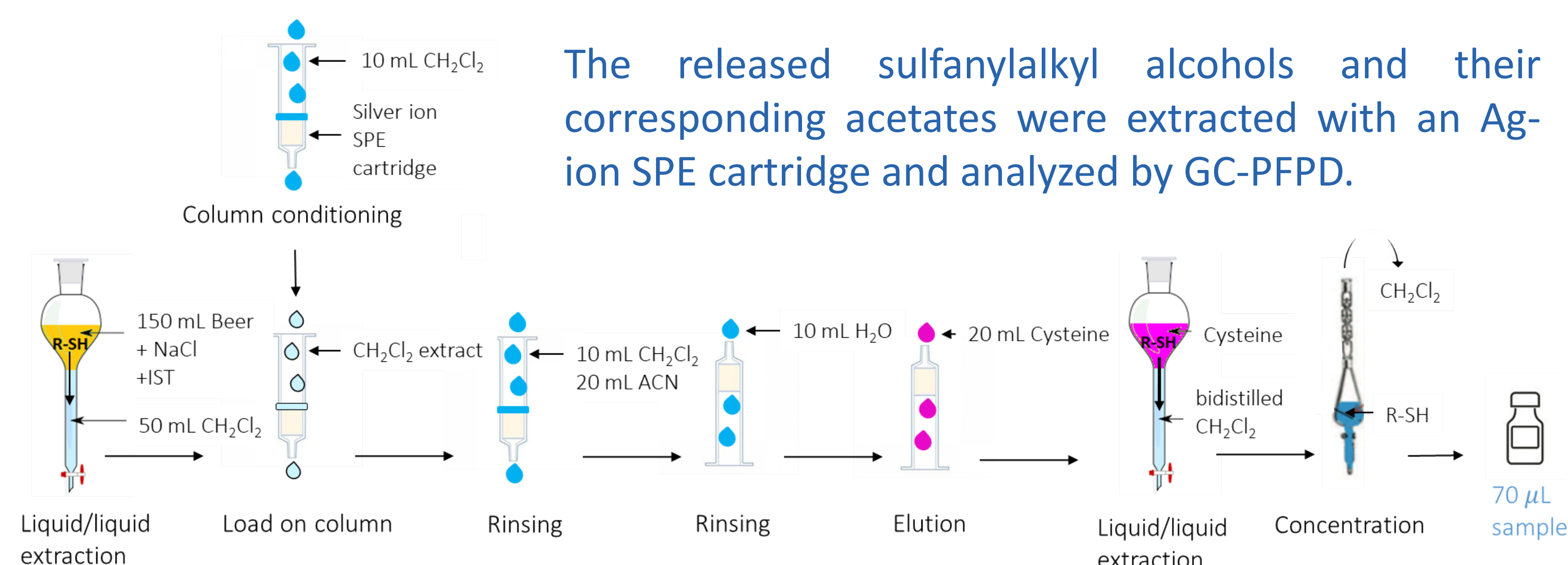
Materials & methods :

Unhopped worts (12-15-17 °P) were spiked with Cys- (5 ppm) or G- (10 ppm) 3SPol, 3S4MPol and 3SHol, fermented for 7 days (at 18-24-28 °C), and kept at 4 °C (for 0-3-5 days).

Spiking:
Cys-/G-adducts of
3SPol, 3SHol &
3S4MPol
(10-15 ppm)

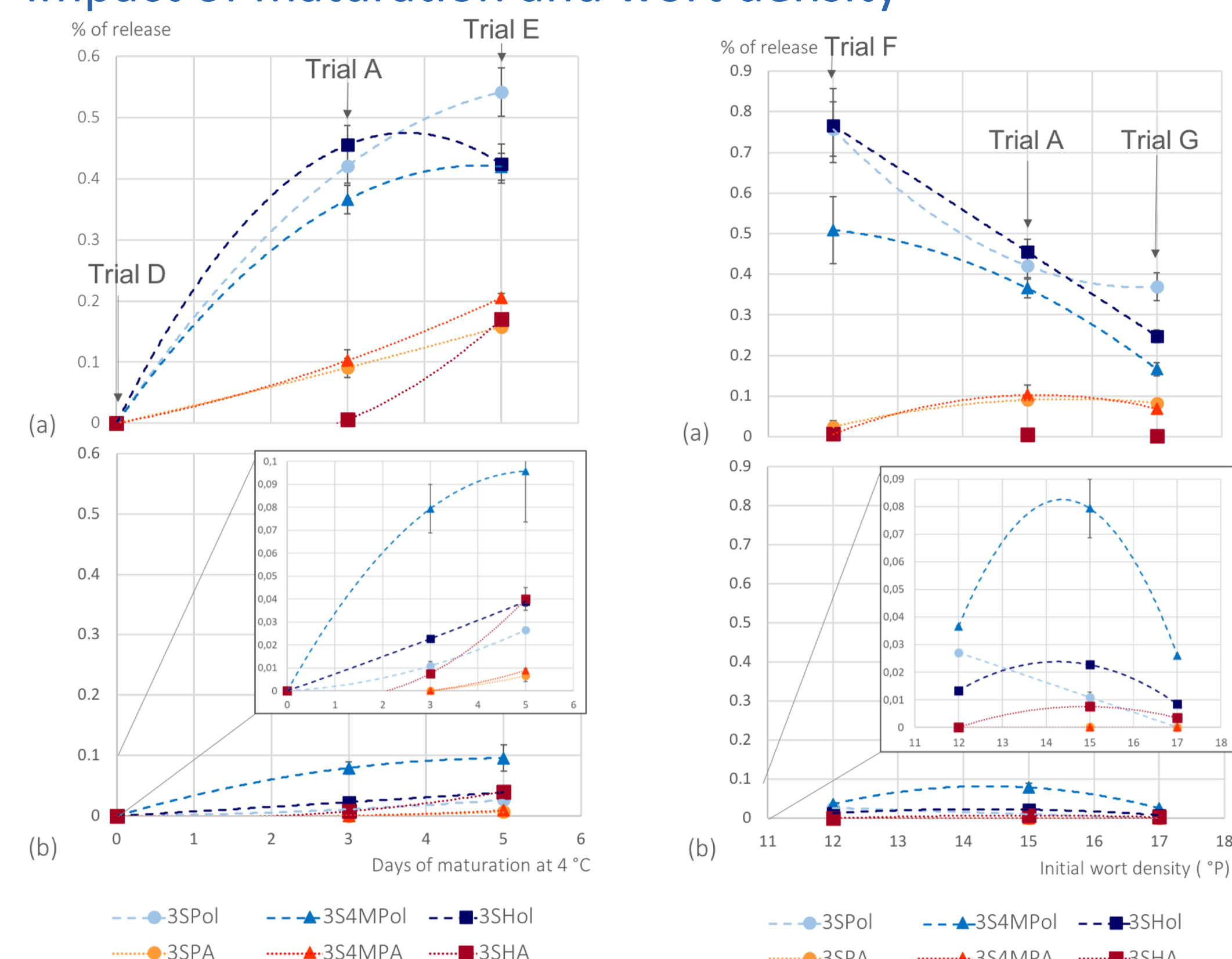


The released sulfanylalkyl alcohols and their corresponding acetates were extracted with an Ag-ion SPE cartridge and analyzed by GC-PFPD.



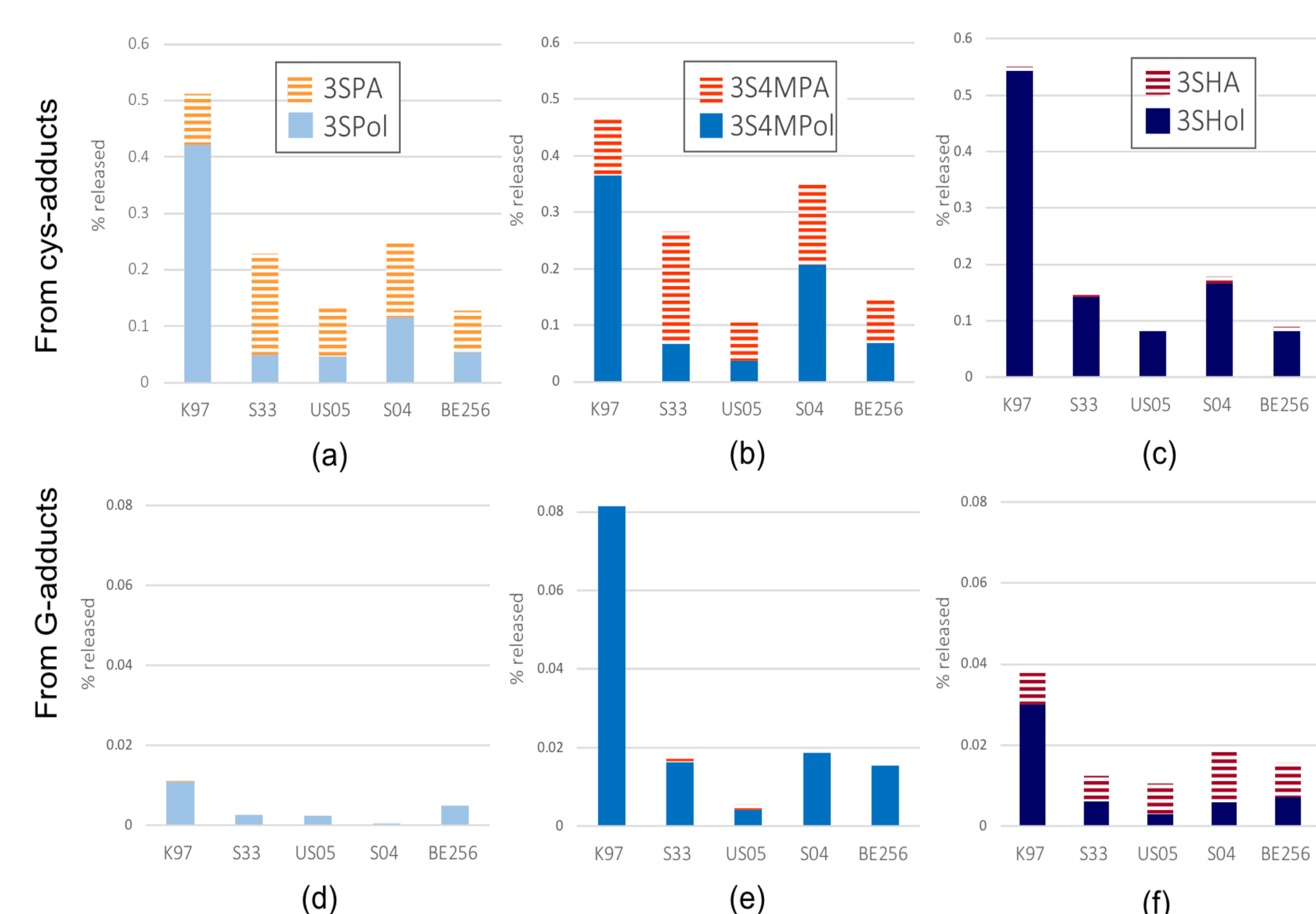
Results :

Impact of maturation and wort density



- ⇒ No thiol release observed if no maturation
- ⇒ 3 days of maturation = good compromise to observe release while avoiding oxidation risk
- ⇒ Higher release of sulfanylalkyl alcohols at 12 °P but higher esterification at 17 °P; 15 °P = good compromise

Yeast strains comparison



Ratio of acetate (released acetate/released alcohol+acetate, %) from Cys- and G-adducts for different yeast strains

Yeast ->	Spiking with									
	Cys-adducts					G-adducts				
	K-97	S-33	US-05	S-04	BE-256	K-97	S-33	US-05	S-04	BE-256
3SPA	18	79	66	53	58	1	0	0	0	0
3S4MPA	22	75	64	40	53	0	6	23	0	0
3SHA	1	6	4	7	8	20	50	71	68	54

- ⇒ K-97 = the highest thiol release efficiencies from both Cys- & G- adducts
- ⇒ S-33 = greater ability to esterify released sulfanylalkyl alcohols
- ⇒ Same selectivity of release according to thiol nature for the five yeasts:
 - Stronger esterification of 3SPol & 3S4MPol when released from Cys-adducts resulting in higher total (alcohol+acetate) release (except for K-97)
 - Stronger esterification of 3SHol when released from G-adducts but higher release of 3S4MPol

Conclusion : Regardless the strain used, these new results confirm the ability of ale-brewing yeasts to release PFTs from both cysteinylated and glutathionylated adducts. Our evidence further shows stronger-than-expected effects of wort density and maturation time. The ratio of acetate also varies greatly according to the fermentation conditions and yeast strain. SafAle™ K-97 remains the best candidate for its ability to release sulfanylalkyl alcohols (release efficiency up to 0.45% and 0.08% from Cys- and G-adducts, respectively), while SafAle™ S-33 and SafAle™ S-04 emerge as good challengers, for their better ester-producing efficiency. To enhance the citrusy notes most probably conferred by sulfanylalkyl alcohols, fermenting a 12°P wort with K-97 can be recommended. A higher-gravity wort is more likely to produce more delicate, and flowery notes characteristic of esters.

