Unveiling the Role of Chlorogenic Acid Esterase in Modulating Browning Index and Inhibition of Non-Fluorescent AGEs through Caffeic Acid Formation in Sunflower Muffins

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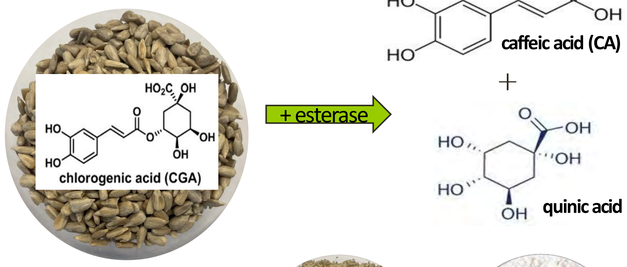
Citrus College STEM
SCIENCE TECHNOLOGY ENGINEERING MATHEMATICS

Introduction

- Sunflower meal (**SFM**) is a by-product of the sunflower oil industry traditionally used as animal feed that is rich in protein, fiber, essential amino acids, phenolic compounds, and minerals[1].
- The predominant antioxidant phenolic compound is chlorogenic acid (**CGA**)[2].
- Chlorogenic acid esterase (**CGA esterase**), cleaves CGA into caffeic acid (**CA**) and quinic acid and can influence the up-cycling of SFM in commercial applications[3].
- CGA can influence both color and formation of advanced glycation end products (**AGEs**)[4].



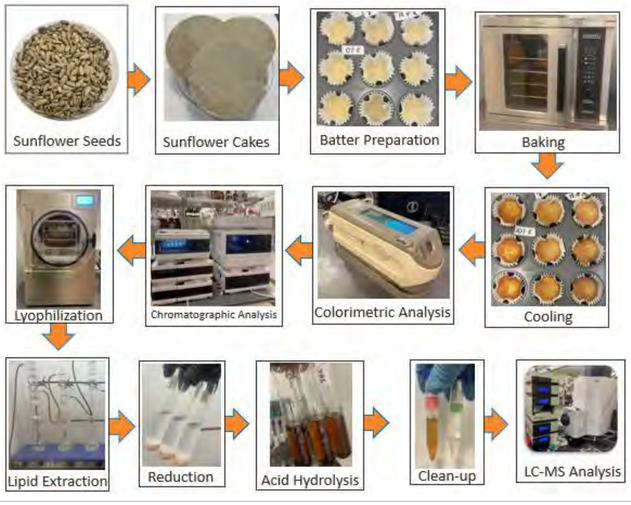
	Sunflower Meal	Wheat Flour
Proximates (per 100g)[5]		
Protein (g)	20.78	9.89
Carbohydrates (g)	20.0	0.97
Lipid (g)	51.46	74.22
Allergen potential	Non-Allergenic	Allergenic
Cost ($/ton)[6,7]	160	368

Goal and Objectives

Upcycling sunflower oil byproduct (SFM) using CGA esterase by

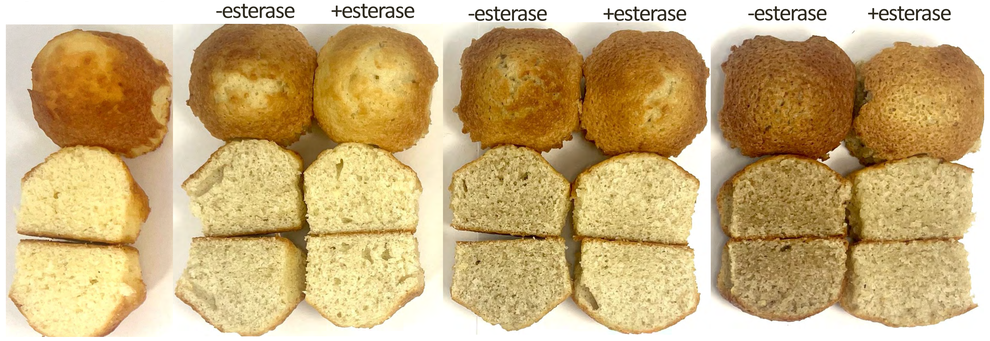
1. Creating a more visually appealing muffin
2. Creating a more nutritious muffin by lowering non-fluorescent AGEs

Workflow



Browning Index and Hydroxycinnamic Acids (HCAs)

Table 1: Lightness and browning index (BI) of muffins as a function of Chlorogenic acid (CGA) esterase and % SFM.



	0% SFM (100% Wheat)	12.5% SFM (87.5% Wheat) -esterase	12.5% SFM (87.5% Wheat) +esterase	25% SFM (75% Wheat) -esterase	25% SFM (75% Wheat) +esterase	50% SFM (50% Wheat) -esterase	50% SFM (50% Wheat) +esterase
Lightness (Hunter L*) Over Time (hrs)							
0.5	84.69±1.35	75.83±1.05	79.34±0.60 ⇑	71.51±1.07	75.47±6.13 ⇑	56.45±1.81	69.47±3.16 ⇑
3	84.58±1.75	74.15±0.59	78.52±0.70 ⇑	67.32±3.82	74.38±0.83 ⇑	59.73±1.73	73.60±2.38 ⇑
6	84.05±0.47	76.0±1.19	77.14±1.60 ⇑	70.63±0.40	73.34±0.44 ⇑	58.59±3.42	68.72±2.28 ⇑
Browning Index							
0.5	28.55±3.16	37.20±4.50	31.20±0.07 ⇓	39.14±4.17	33.17±9.75 ⇓	62.40±0.30	41.09±0.41 ⇓
3	28.82±0.93	39.04±2.28	32.55±0.77 ⇓	41.91±3.86	33.071±2.72 ⇓	57.11±1.54	35.20±2.77 ⇓
6	28.78±2.71	33.37±2.07	32.54±4.41 ⇓	40.63±0.57	37.53±0.21 ⇓	55.98±0.32	42.23±0.20 ⇓
Hydroxycinnamic Acids (µg/mL)							
CGA	5.03±0.60	20.16±3.09	8.65±0.84 ⇓	33.60±5.30	11.44±2.57 ⇓	70.61±1.75	27.61±0.37 ⇓
CA	1.26±0.48	1.27±0.08	12.27±0.11 ⇑	1.48±0.11	17.23±4.30 ⇑	2.14±0.08	39.40±1.72 ⇑

⇑ Represents an increase in Hunter L*, BI, and HCA concentration while ⇓ represents a decrease as a function of %SFM and CGA esterase.

Key Findings

CGA esterase increased CA by 864% in 12.5% SFM and 1742% in 50.0% SFM (Figure 1)

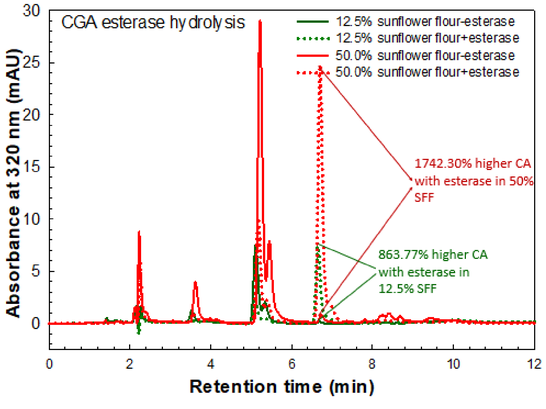


Figure 1: Chromatogram of muffin samples containing 12.5% and 50% SFM (n=2). Control samples (=) did not contain the CGA esterase enzyme while (⁝⁝) contained CGA esterase.

Table 2: Pearson Correlation coefficients between % SFM, protein, phenolic acids with browning index.

	Caffeic	CGA	BI
% SFM (-esterase)	0.994	0.997	0.965
%SFM (+esterase)	0.987	0.981	0.990
BI (-esterase)	0.988	0.982	
BI (+esterase)	0.999	0.999	
Soluble Protein	0.246	0.916	0.942

- Browning was highly correlated with % SFM which had higher caffeic acid and CGA content
- Soluble protein was highly correlated with CGA but not caffeic acid (Table 2)

CGA esterase increased CML values in 50% SFM

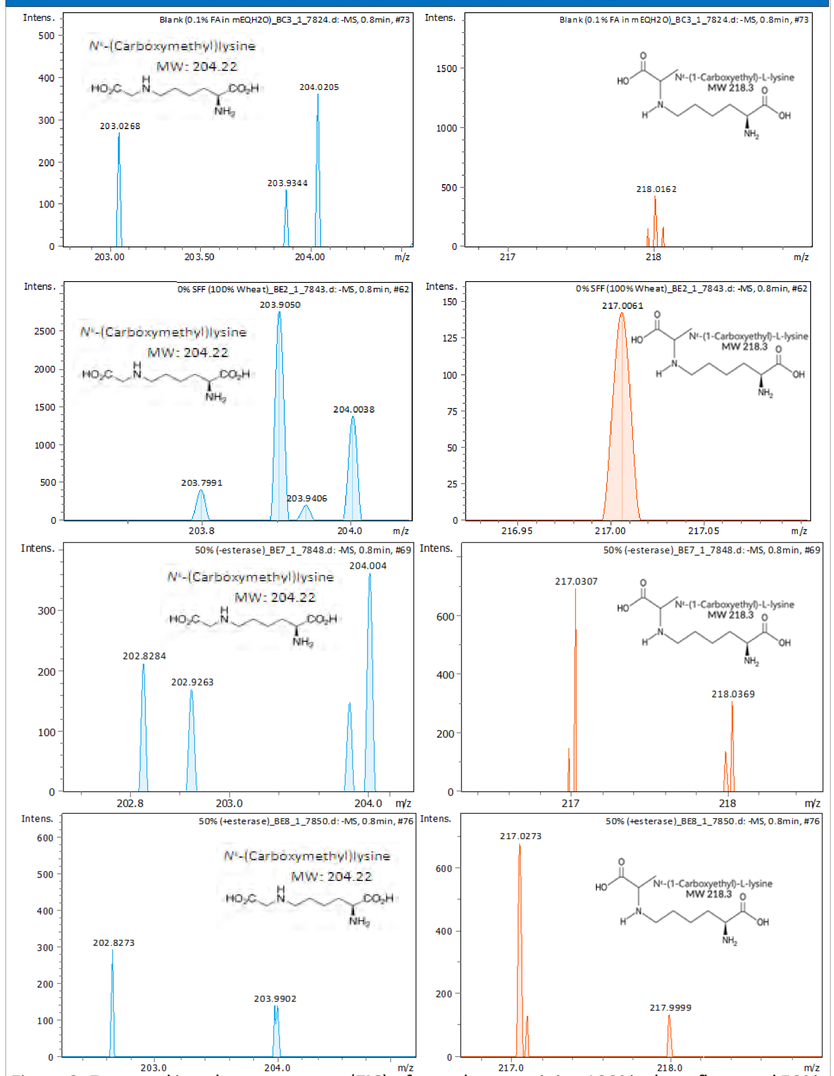


Figure 2: Extracted ion chromatograms (**EIC**) of samples containing 100% wheat flour and 50% SFM with and without CGA esterase. Samples in left column represent N^{ϵ}-(Carboxymethyl)lysine (**CML**) and right column represent N^{ϵ}-(Carboxyethyl)lysine (**CEL**).

Conclusion

- Chlorogenic acid esterase decreased the browning index in all treatments of muffins
- Modifying the LC-MS method gradient may allow CML and CEL to elute later and limit possible ionization from any salts present in samples

References

1. Yegorov B., Turpurova T., Sharabaeva E., Bondar Y. Prospects of using by-products of sunflower oil production in compound feed industry. *J. Food Sci. Technol. Ukr* 2019;13:106–113. doi: 10.15673/fstv13i1.1337.
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Alternate Text

Alexander Gomez

Chapman University

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•The predominant antioxidant phenolic compound is chlorogenic acid (**CGA**)2.
•Chlorogenic acid esterase (**CGA esterase**), cleaves CGA into caffeic acid (**CA**) and quinicacid and can influence the up-cycling of SFM in commercial applications3.
•CGA can influence both color and formation of advanced glycation end products (**AGEs**)4.

Visual aids titled: 'Chlorogenic Acid', '+ eterase' 'caffeic acid' 'quinic acid', 'sunflower meal', and 'wheat flour'.

Goal and Objectives:

Upcycling sunflower oil byproduct (SFM) using CGA esterase by

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Workflow:

Sunflower Seeds----Sunflower Cakes---Batter Preparation---Baking---Cooling---Colorimetric Analysis---Chromatographic Analysis---Lyophilization---Liquid Extraction---Reduction---Acid Hydrolysis---Clean-up---LC-MS Analysis.

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