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Study into the polyphenol content and antioxidant activity of rapeseed pomace extracts

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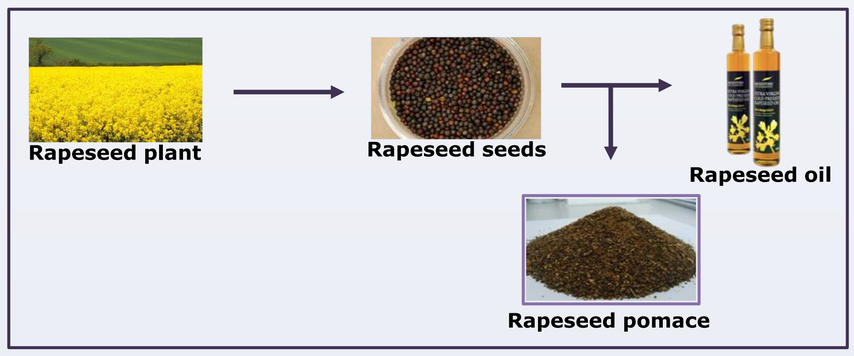
Introduction

Rapeseed (*Brassica napus)*, is a plant used for oil production and naturally grown in Scotland. During the oil production it provides a by-product called **rapeseed pomace** or rapeseed meal/cake.



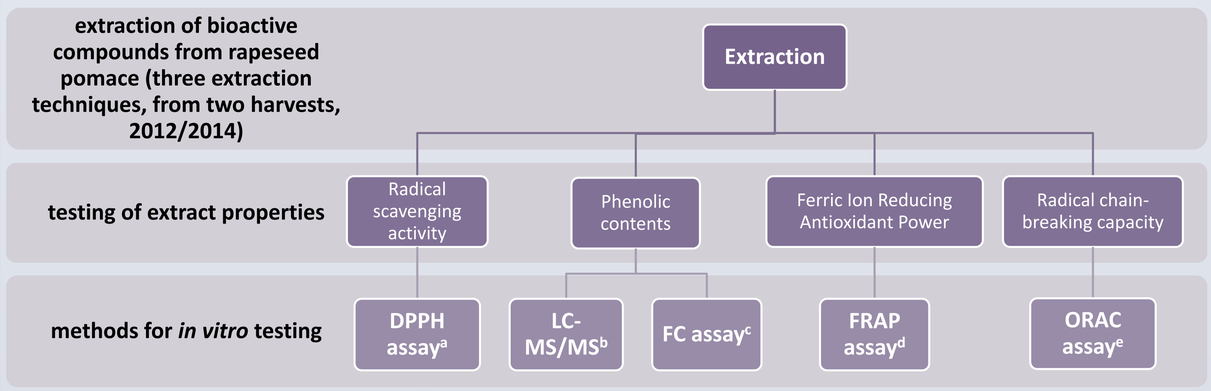
Several bioactive compounds have been found in rapeseed, such as tocols, phytosterols, phospholipids and **phenolic compounds**[1]. Most abundant phenolics in rapeseed are **ferulic acid, salicylic acid, cinnamic acid, p-hydroxybenzoic acid and sinapic acid**[2]. Several phenolics have been observed to act as antioxidants and radical scavengers, potentially counteracting oxidative stress[3]. Due to these properties they could play an important role in the **prevention/treatment of oxidative stress related disease**.

Aims

The ***aims*** of this study was it to determine the polyphenol content, radical scavenging activity, ferric iron reducing antioxidant power and the oxygen-radical absorbance capacity assay, of RSP extracts, with regard to their potential application in the treatment and/or prevention of neurodegenerative diseases.

Methods

Rapeseed pomace obtained from Mackintosh of Glendaveny (Peterhead, Aberdeenshire), was subjected to the following procedures:



[a] 2,2-diphenyl-1-picrylhydrazyl assay, [b] liquid chromatography-mass spectrometry, [c] Folin-Ciocalteu assay, [d] Ferric Ion Reducing Antioxidant Power assay, [e] Oxygen Radical Absorbance Capacity assay

Statically significant differences were analysed via two-way ANOVA and Bonferroni's multiple comparisons test between the two years for each extraction technique ($p \leq 0.05$*, $p \leq 0.01$**, $p \leq 0.001$***, $p \leq 0.0001$****, ns-not significant).

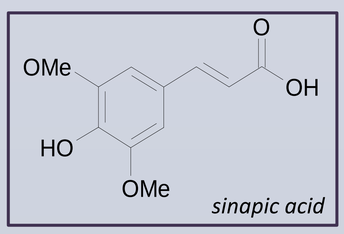
Results

- The extraction yield (Table 1) show accelerated solvent extraction (ASE) to be the most efficient, followed by ultrasonic assisted (UAE) and soxhlet extraction (SOX)

Table 1: Extraction yields obtained from rapeseed pomace, for three different extraction techniques and two variant harvests (2012, 2014)

Extraction	Year	Yield [mg/1g pomace]	Yield (%)
SOX	2012	79.55	7.96
	2014	75.59	7.56
UAE	2012	158.59	15.86
	2014	175.46	17.55
ASE	2012	362.31	**36.23**
	2014	344.67	**34.47**

- Most abundant phenolics found in the extracts analysed by LC-MS/MS[4] profiling are listed in Table 2, with **sinapic acid** (Figure 1) being at the most abundant in all the extracts



Sinapic acid has previously been shown to have anti-inflammatory properties[5] and attenuated memory impairments in amyloid-ß $(Aß)_{1-42}$ protein induced mouse model.[6]

*Figure 1: Most abundant phenolic sinapic acid (*4-hydroxy-3,5-dimethoxycinnamic acid)

Table 2: Most abundant phenolics found in the rapeseed pomace extracts (mean ± std)

extraction	Year	Most common phenolics (conc. g/kg)		
SOX	2012	**sinapic acid (7.497 ± 0.199)**	**ferulic acid** (0.227±0.008)	**4-hydroxyphenylpyruvic acid** (0.172±0.043)
	2014	**sinapic acid (4.897±0.282)**	**syringic acid** (0.224±0.017)	**ferulic acid** (0.183±0.010)
UAE	2012	**sinapic acid (4.610±0.062)**	**4-hydroxyphenylpyruvic acid** (0.160±0.015)	**ferulic acid** (0.126±0.007)
	2014	**sinapic acid (2.769±0.080)**	**4-hydroxyphenylpyruvic acid** (0.129±0.005)	**syringic acid** (0.113±0.009)
ASE	2012	**sinapic acid (2.682±0.058)**	**4-hydroxyphenylpyruvic acid** (0.1118±0.008)	**ferulic acid** (0.071±0.002)
	2014	**sinapic acid (1.923±0.018)**	**4-hydroxyphenylpyruvic acid** (0.161±0.014)	**cinnamic acid** (0.085±0.003)

- Results obtained for the **antioxidant properties** of the extracts are shown below:

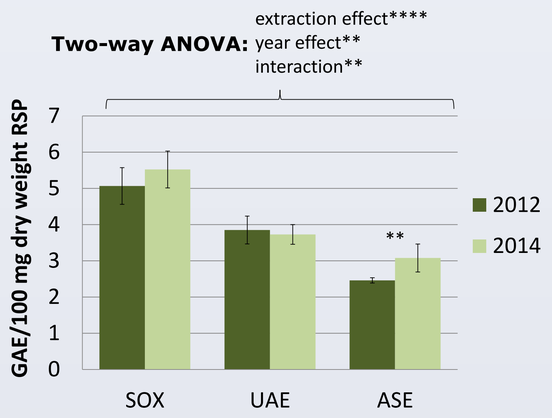


Figure 2: FC assay

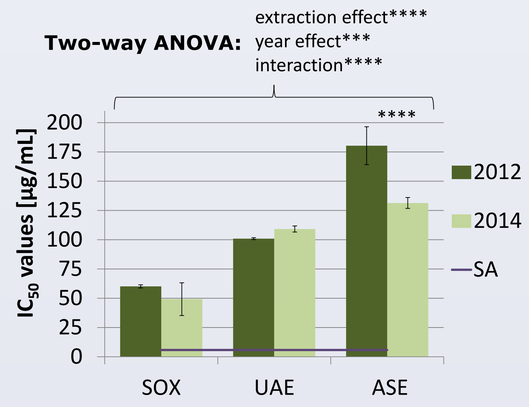


Figure 3: DPPH assay

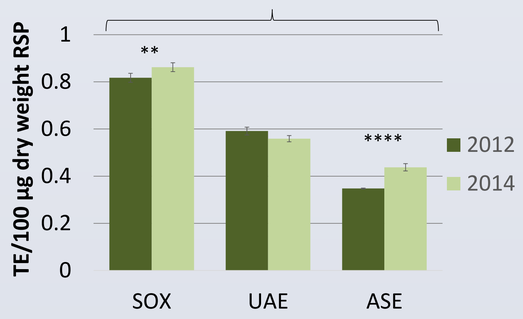


Figure 4: FRAP assay

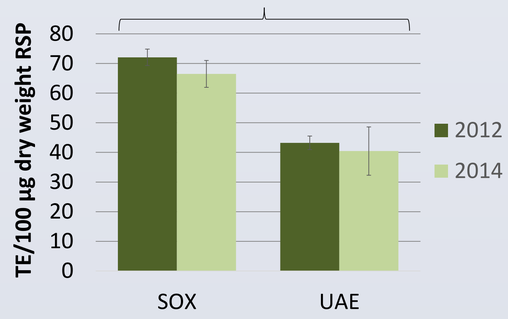


Figure 5: ORAC assay

Conclusions

- Investigation into the phenolic content and antioxidant activity showed:
  - **Phenolic content in all three extracts is qualitatively similar**
  - **Sinapic acid is the most abundant phenolic measured in all extracts**
  - **All extracts show *in vitro* antioxidant activity (FRAP, ORAC and DPPH)**
- Extraction technique has a higher significant difference than harvest year
- **SOX** extracts contained **the highest amount of phenolics** and **the greatest antioxidant capacity for all the *in vitro* analysis**
- Next steps to investigate the potential use of rapeseed pomace extracts for the prevention/treatment of oxidative stress related disease are:
  - **Antioxidant activity/protective effect in neuroblastoma cell line SH SY-5Y**
  - **Bioactivity and bioavailability analysis in neurodegeneration related disease models of *C.elegans***

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Acknowledgements

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