

## **SUPPLEMENTARY MATERIALS**

### **The In Vitro Inhibitory Effect of Selected Asteraceae Plants on Pancreatic Lipase Followed by Phenolic Content Identification through Liquid Chromatography High Resolution Mass Spectrometry (LC-HRMS)**

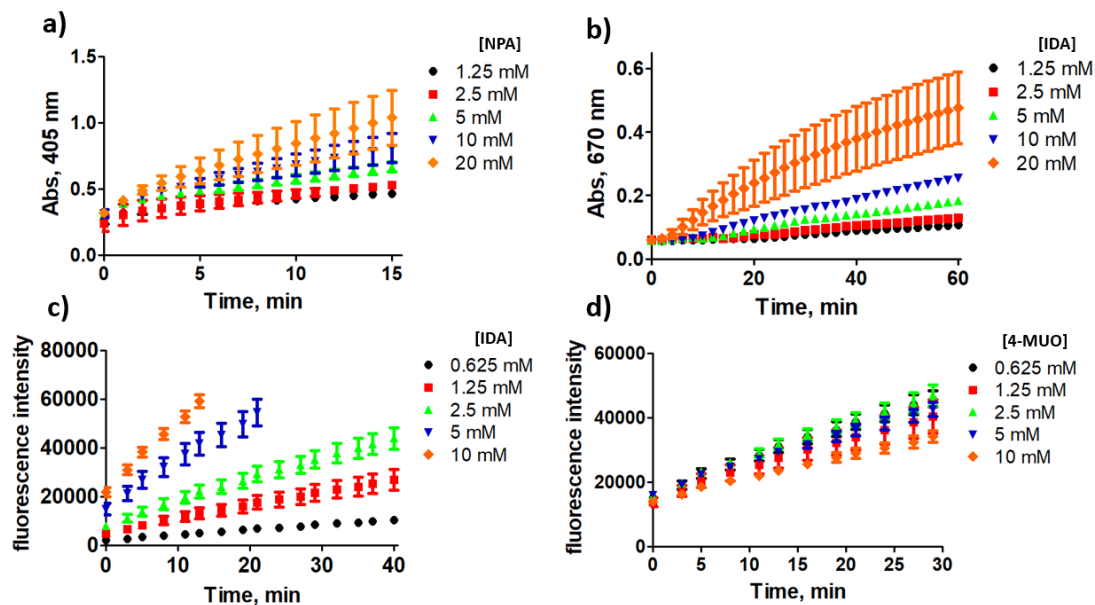
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## S1. Supporting information on substrate selection



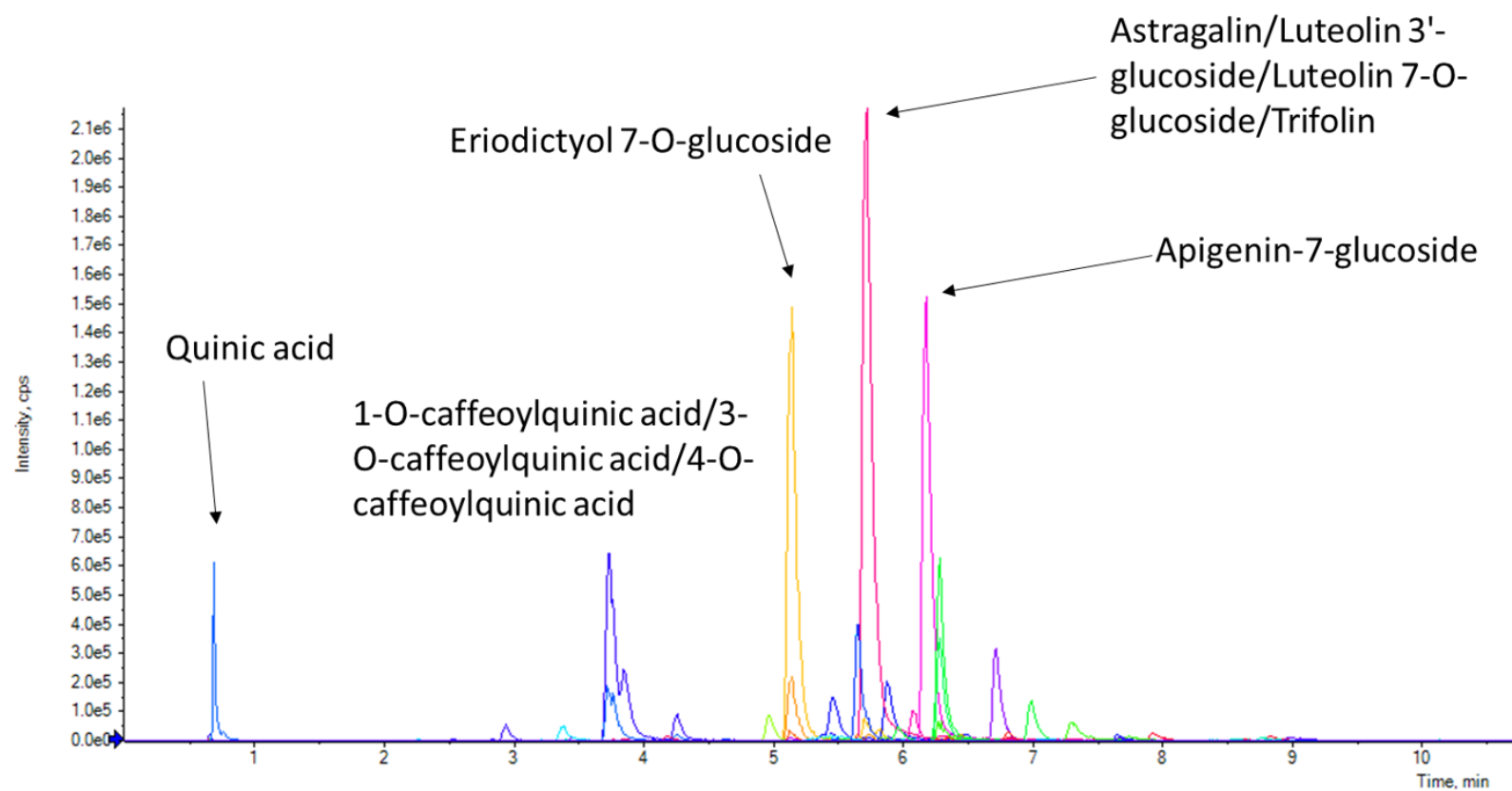
**Figure S1.** PNLIP assay signal development using a) NPA, b,c) IDA and d) 4-MUO as the substrate. PNLIP concentration was 1250  $\mu\text{g mL}^{-1}$  in every case and each point represents the mean value ( $n=9$ ) and the error bars represent the standard deviation in each case.

## S2. Supporting information on assay optimisation

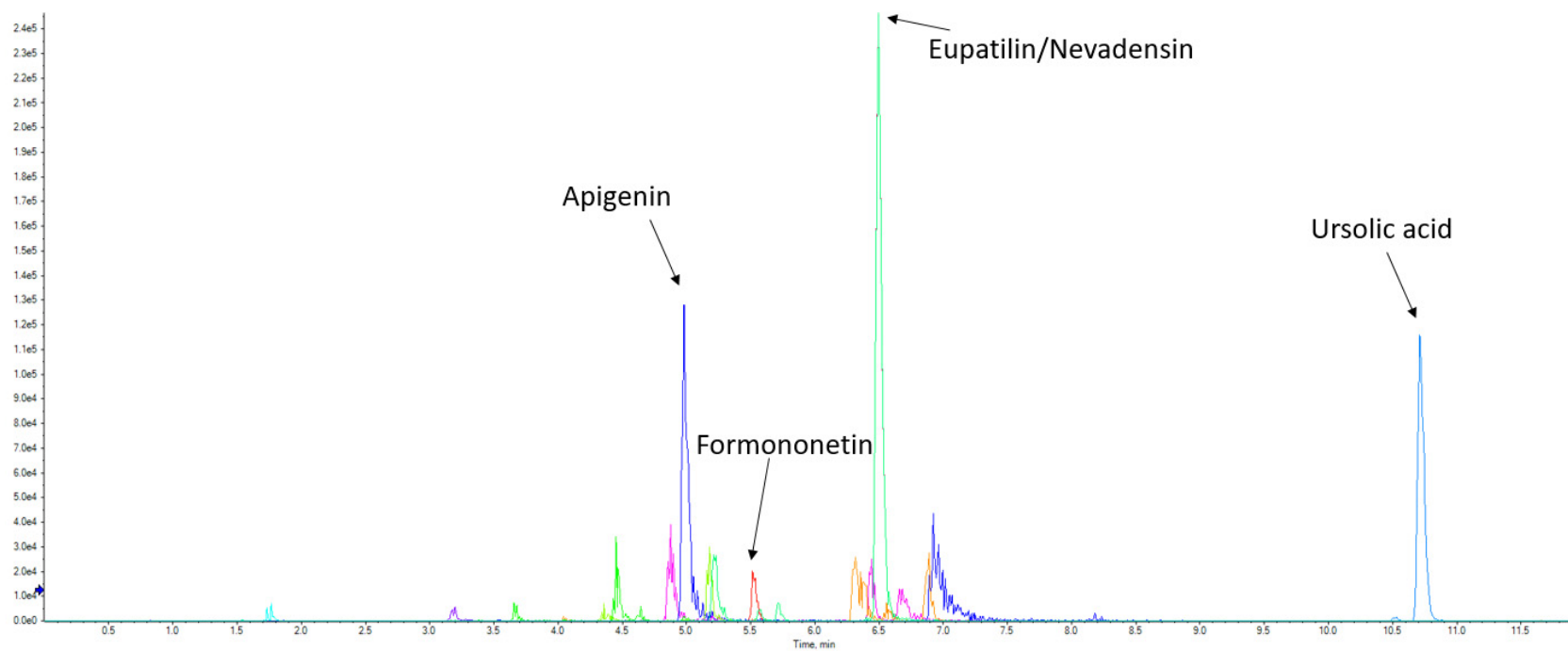
**Table S1.** Calculated  $K_m$  and  $V_{max}$  at different end-points using  $1250 \mu\text{g mL}^{-1}$  PNLIP. The results are expressed as “mean (95% intervals)” (n=6 replicates per level).

end-point (min)	$K_m$ (mM)	$V_{max}$ (dAbs/min)
5	16 (9.6-22)	0.066 (0.054-0.078)
10	14 (6.2-23)	0.046 (0.035-0.058)
20	17 (4.6-29)	0.043 (0.029-0.058)
40	14 (8.1-19)	0.035 (0.029-0.041)
60	16 (10-22)	0.035 (0.029-0.041)

### S3. Indicative XIC chromatograms



**Figure S2.** XIC chromatogram of *Chrysanthemum morifolium* flower aqueous extract, ESI-.



**Figure S3.** XIC chromatogram of *Achillea millefolium* leaves dichloromethane extract, ESI-.

#### S4. Enzyme assays protocols

##### *PNLIP assay using NPA as the substrate*

Eighty  $\mu\text{L}$  PNLIP ( $1250 \mu\text{g mL}^{-1}$ ) were incubated with 10  $\mu\text{L}$  of a sample for 15 min. Next, 10  $\mu\text{L}$  of a NPA solution (in the range 1.25-20 mM) in DMSO were added and the absorbance was measured at 405 nm for 15 min.

##### *PNLIP assay using IDA as the substrate*

Eighty  $\mu\text{L}$  PNLIP ( $1250 \mu\text{g mL}^{-1}$ ) were incubated with 10  $\mu\text{L}$  of a sample for 15 min. Next, 10  $\mu\text{L}$  of an IDA solution (in the range 1.25-20 mM) in DMSO were added and the absorbance was measured at 670 nm for 60 min. In the case of fluorescence monitoring, 10  $\mu\text{L}$  of an IDA solution (in the range 0.625-10 mM) in DMSO were added and the fluorescent signal was monitored using the following wavelengths  $\lambda_{\text{exc}} = 395 \text{ nm}$  and  $\lambda_{\text{em}} = 470 \text{ nm}$ .

##### *PNLIP assay using 4-MUO as the substrate*

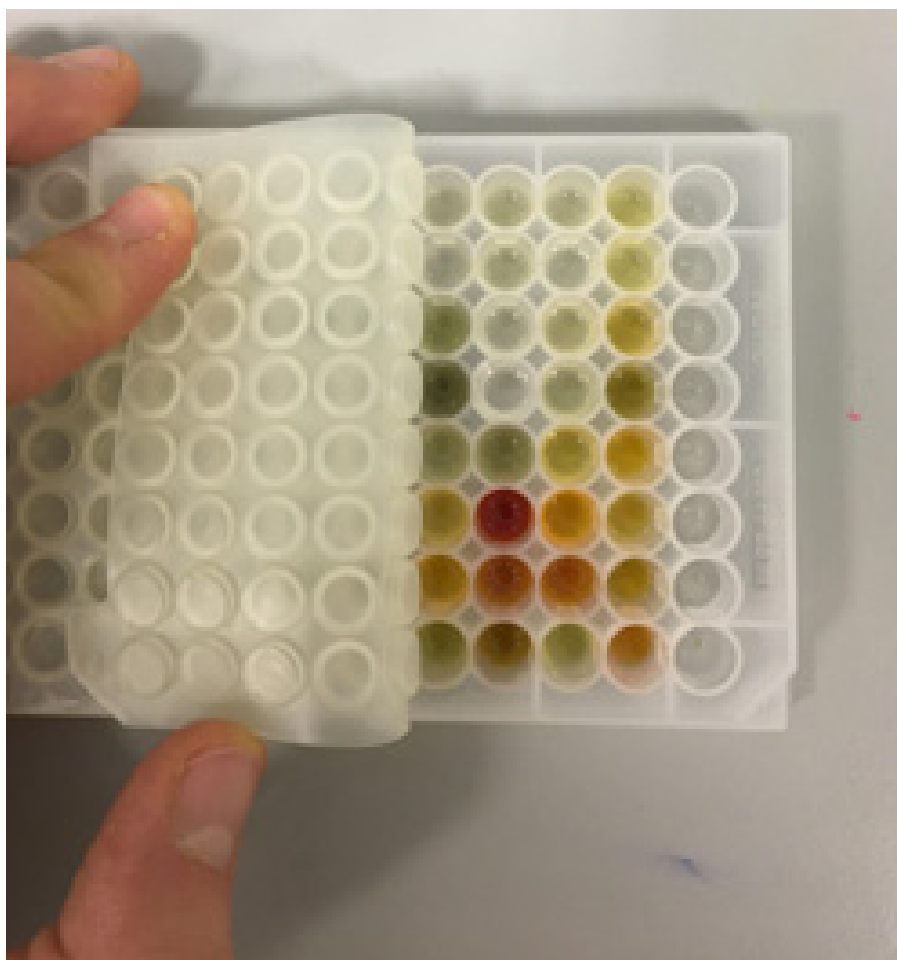
Eighty  $\mu\text{L}$  PNLIP ( $1250 \mu\text{g mL}^{-1}$ ) were incubated with 10  $\mu\text{L}$  of a sample for 15 min. Next, 10  $\mu\text{L}$  of a 4-MUO solution (in the range 0.625-10 mM) in DMSO were added and the fluorescent signal was monitored using the following wavelengths  $\lambda_{\text{exc}} = 355 \text{ nm}$  and  $\lambda_{\text{em}} = 460 \text{ nm}$ .

#### S5. Tested plants

**Table S2.** The list of samples investigated for PNLIP inhibition potency.

Botanical Family	Species	Common name	Part of plant
Asteraceae	<i>Achillea millefolium</i>	Yarrow	Leaves
	<i>Anthemis nobilis</i>	Chamomile Herb	Flower
	<i>Arctium lappa</i>	Burdock Root	Root
	<i>Artemisia abrotanum</i>	Southernwood Herb	Leaf
	<i>Artemisia absinthium</i>	Wormwood	Aerial parts
	<i>Artemisia annua</i>	Sweet wormwood (Qing Hao)	Stem

<i>Artemisia dracunculus</i>	Tarragon Herb	Leaf
<i>Artemisia vulgaris</i>	Mugwort Herb	Aerial parts
<i>Atractylodes macrocephala</i>	Atractylodes	Rhizome
<i>Calendula officinalis</i>	Marigold flowers	Flower
<i>Calendula officinalis</i>	Marigold Petals	Petal
<i>Cichorium intybus</i>	Chicory Root	Root
<i>Cnicus benedictus</i>	Holythistle	Aerial parts
<i>Cynara cardunculus</i>	Artichoke	Leaf of stem
<i>Eclipta alba</i>	Bhringaraj Root	Root
<i>Echinacea angustifolia</i>	Echinacea Angustifolia Root	Root
<i>Echinacea purpurea</i>	Echinacea Purpurea Root	Root
<i>Eupatorium perfoliatum</i>	Boneset	Leaf
<i>Eupatorium purpureum</i>	Gravel root	Root
<i>Grindelia camporum</i>	Grindelia Herb	Aerial parts
<i>Helianthus annuus</i>	Sunflower seed	Seed
<i>Hieracium pilosella</i>	Mouseear, hawkweed	Aerial parts
<i>Chrysanthemum morifolium</i>	Chrysanthemum flowers	Flower
<i>Inula helenium</i>	Elecampane Root	Root
<i>Lactuca sativa</i>	Lettuce	Leaf
<i>Lactuca virosa</i>	Wild Lettuce	Leaf
<i>Matricaria recutita</i>	Chamomile, German	Flower
<i>Silybum marianum</i>	Milkthistle Seed	Seed
<i>Solidago virgaurea</i>	Golden Rod	Aerial parts
<i>Stevia rebaudiana</i>	Stevia leaf	Leaf
<i>Tanacetum parthenium</i>	Feverfew Herb	Aerial parts
<i>Tanacetum vulgare</i>	Tansy Herb	Aerial parts
<i>Taraxacum officinale</i>	Dandelion Herb	Leaf
<i>Taraxacum officinale</i>	Dandelion Root	Root
<i>Tussilago farfara</i>	Coltsfoot	Aerial parts



**Figure S4.** The microplate format in which the purchased extracts were stored



## S6. Formulas for inhibition rate calculation

### *Raw data correction*

1. Test sample raw absorbance was corrected by subtracting the raw data of the test blank sample and the raw data of substrate blank (both not containing any PNLIP solution) from the test sample raw data.

$$\text{Corrected test sample Abs} = A_{\text{test sample}} - A_{\text{test sample blank}} - A_{\text{substrate blank}} \text{ (S1)}$$

2. Positive control (orlistat) raw absorbance was corrected by subtracting the raw data of the substrate blank (not containing any PNLIP solution) from the positive control raw data.

$$\text{Corrected positive control Abs} = A_{\text{positive control}} - A_{\text{substrate blank}} \text{ (S2)}$$

3. Negative control-buffer blank: raw data of buffer blank was corrected by calculating raw data of buffer blank minus substrate blank.

$$\text{Corrected negative control Abs} = A_{\text{test buffer blank}} - A_{\text{test substrate blank}} \text{ (S3)}$$

### *Inhibition rate% using the blank corrected data*

4. The inhibition rate% was based on the ratio of the corrected absorbance of a sample (test sample or positive control) divided by the corrected absorbance of the negative control.

$$\text{Inhibition rate\%} = 1 - (A_{\text{corrected sample}} / A_{\text{corrected negative control}}) \times 100 \text{ (S4)}$$