

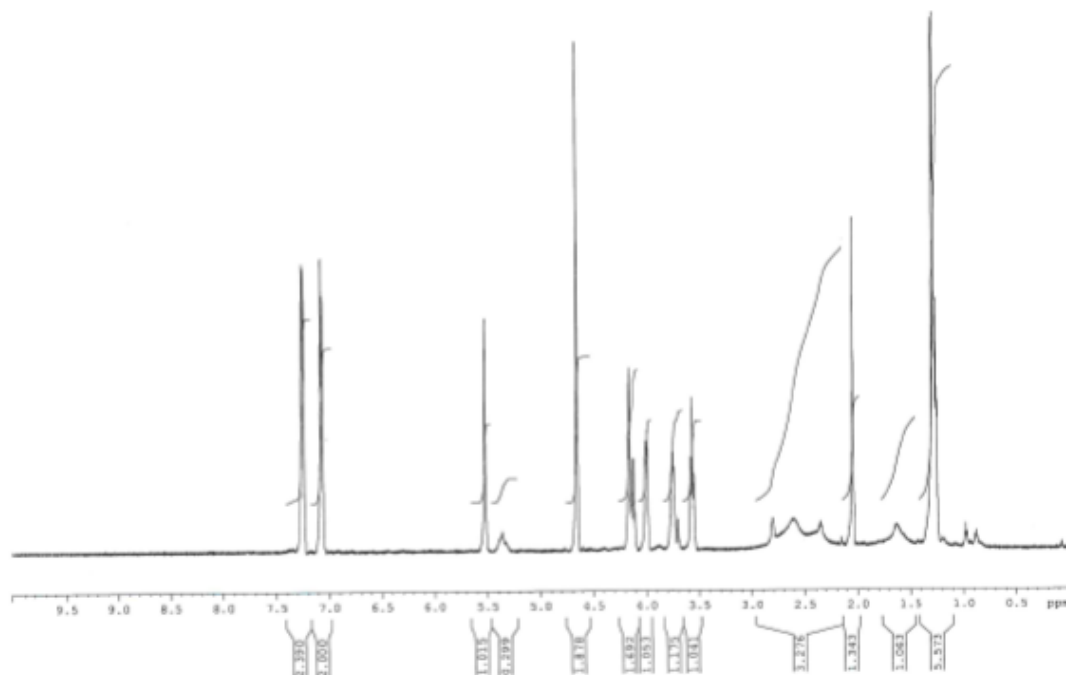
**Moringin, a stable isothiocyanate from *Moringa oleifera*, activates the somatosensory and pain receptor TRPA1 channel in vitro**, by *Gigliola Borgonovo, Luciano De Petrocellis, Aniello Schiano Moriello, Simona Bertoli, Alessandro Leone, Alberto Battezzati, Stefania Mazzini, Angela Bassoli*.

S1- supplementary material- Analytical data for isolated moringin and in vitro assays.

#### <sup>1</sup>H-NMR

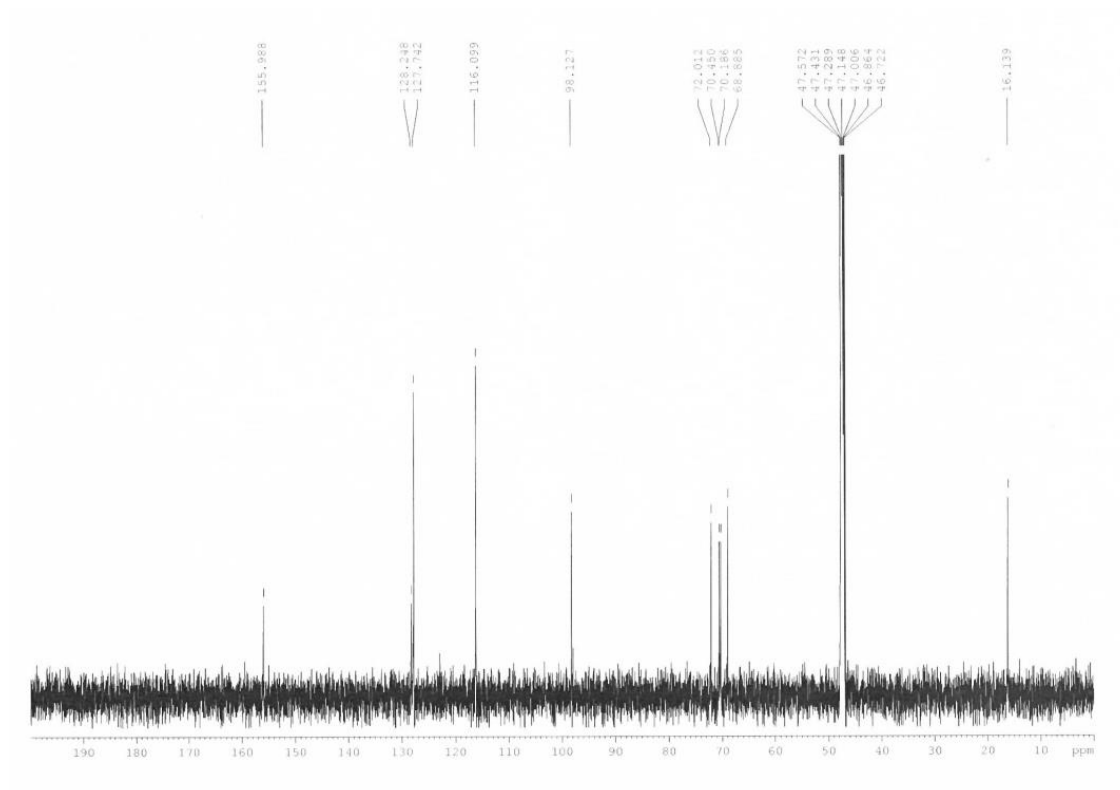
<sup>1</sup>H NMR (MeOH-d<sub>4</sub>) δ: 1.22 (3H, d, J= 6.1 Hz, CH<sub>3</sub>), 3.46 (1H, t, J= 9.5, H-4'), 3.63 (1H, m, H-5'), 3.84 (1H, dd, J= 3.5 and 9.5 Hz, H-3'), 4.00 (1H, bs, H-2'), 4.69 (2H, s, CH<sub>2</sub>), 5.44 (1H, bs, H-1'), 7.09 and 7.29 (4H, 2d, J= 8.4 Hz, arom.).

13/03/19 T-298K



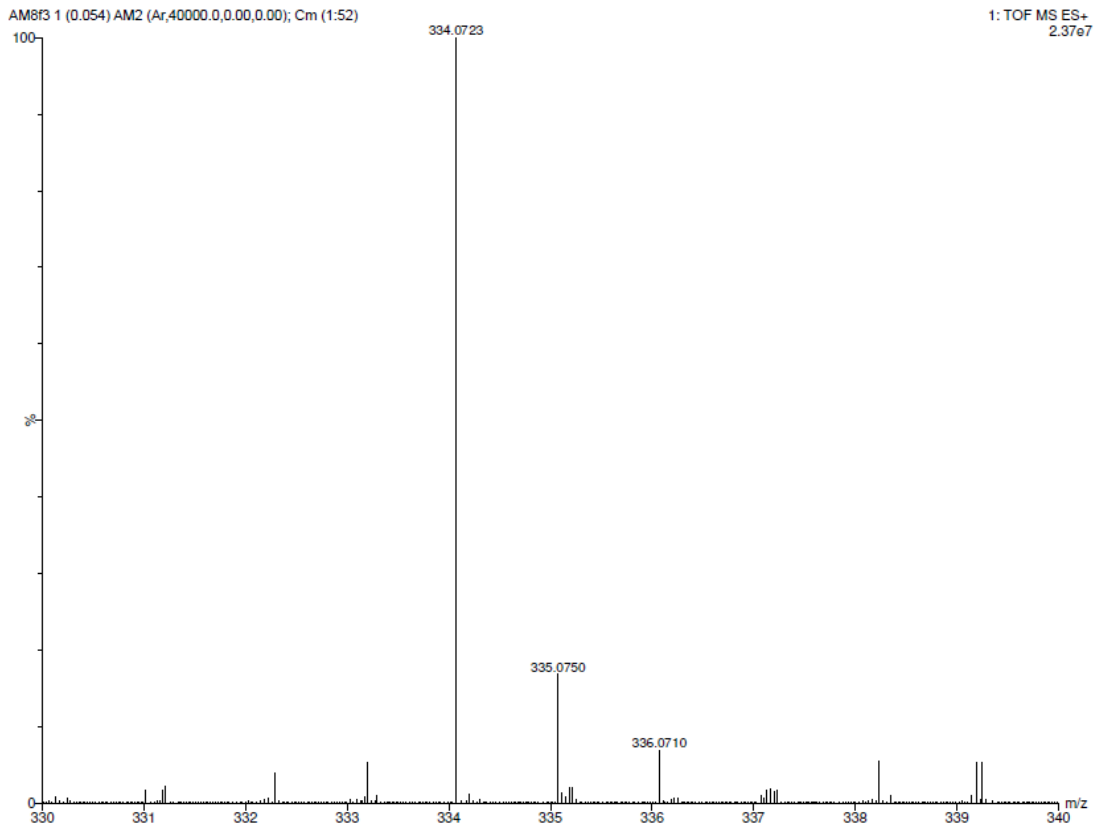
<sup>13</sup>C-NMR spectrum

$^{13}\text{C}$  NMR (MeOH- $d_4$ )  $\delta$ : 13.1, 46.7, 68.9, 70.2, 70.5, 72.0, 98.1, 116.1, 127.8, 128.2, 127.9, 155.9.



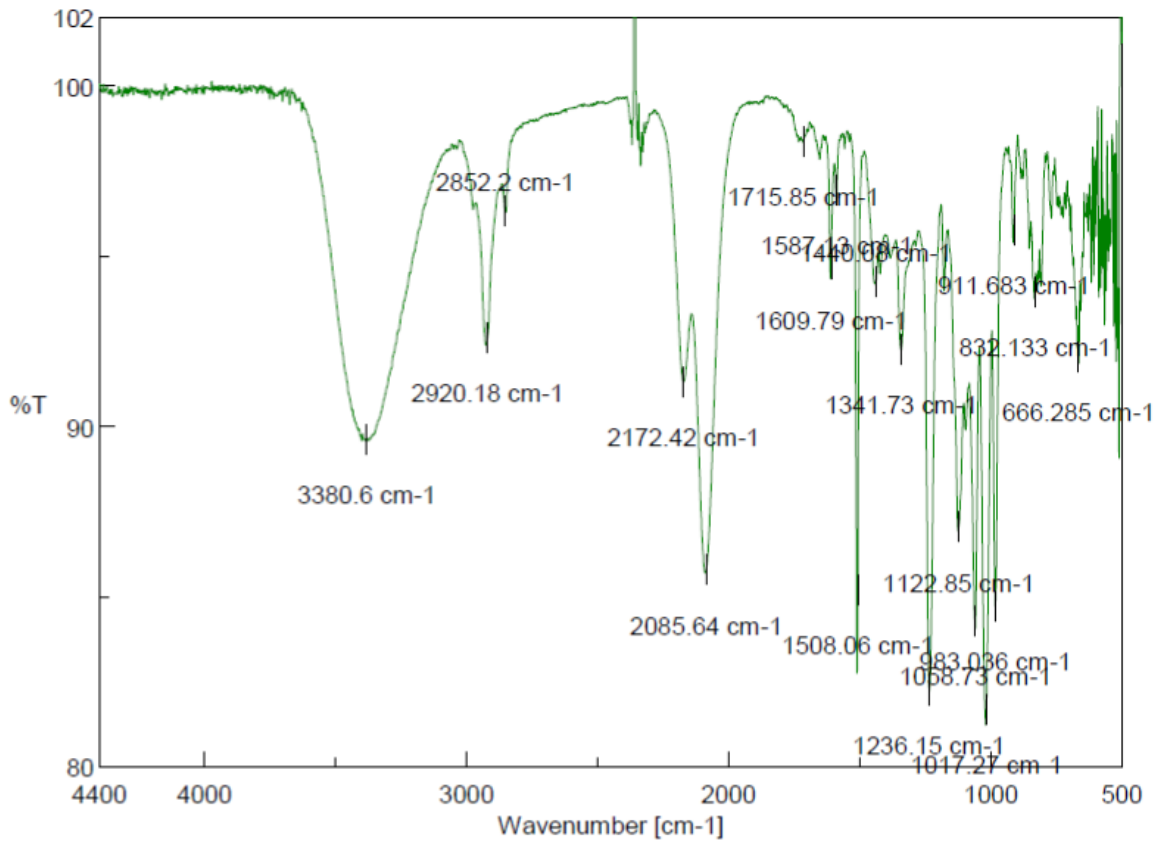
Mass spectrum, TOF MS ES

ESI-MS 334.0723 [M + Na]<sup>+</sup>, 100%, (theoretical C<sub>14</sub>H<sub>17</sub>NO<sub>5</sub>Na 334.0720); 335.0750 [M +1+Na]<sup>+</sup>, 20%;  
336.0710 [M +2+Na]<sup>+</sup>, 4%.



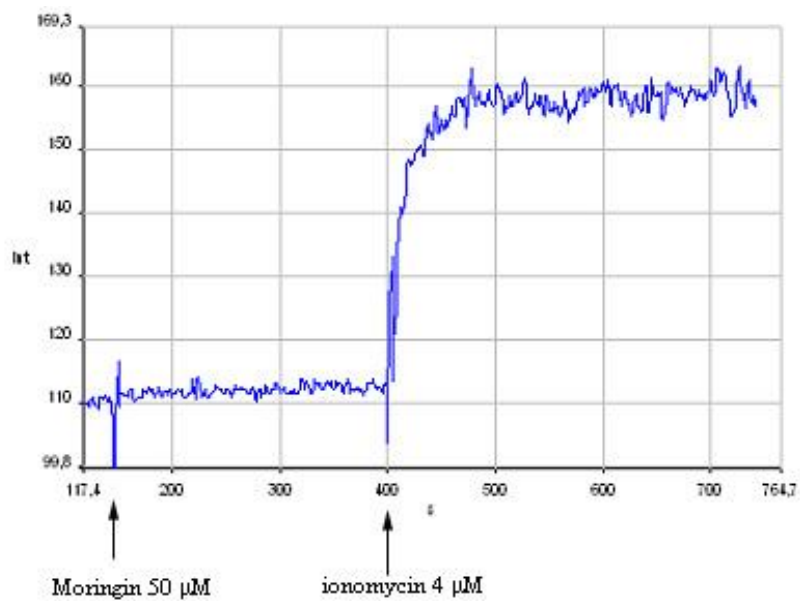
FTIR spectrum

FTIR  $\nu_{\text{max}}$  3381, (br. OH), 2920, 2172, 2086, 1609, 1609, 1508, 1236, 1119, 1123, 1058, 983  $\text{cm}^{-1}$ .



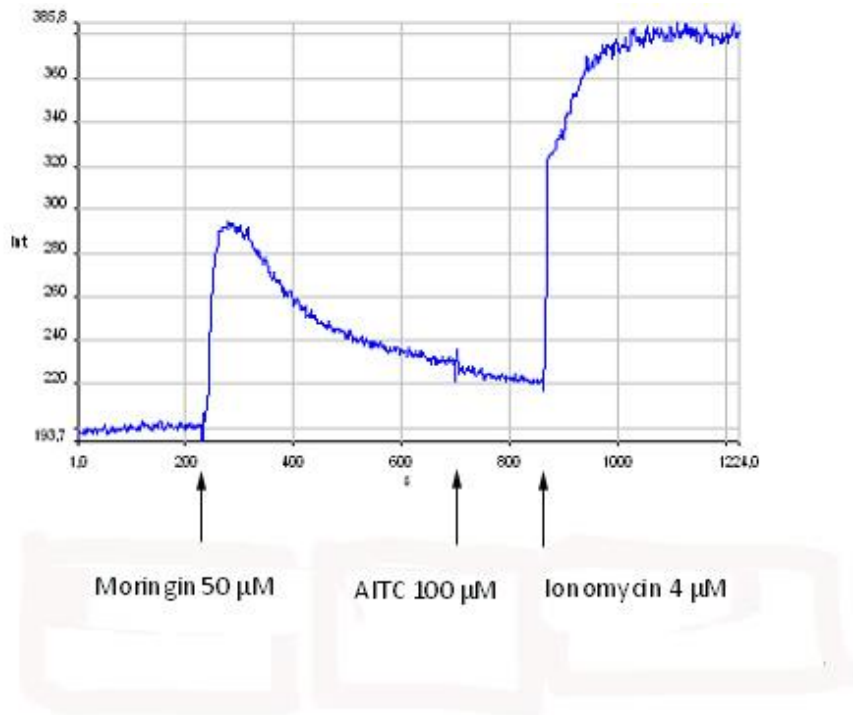
## Time-course of calcium bound Fluo-4 fluorescence intensity in rTRPA1 experiments

In our experiment on rTRPA1 HEK-293 cells 100  $\mu\text{M}$  allyl isothiocyanate (AITC) exerts an efficacy of  $52.4 \pm 4.4$  % of the response to ionomycin 4  $\mu\text{M}$ . Moringin in not transfected HEK-293 cells exert only a very little increase of  $[\text{Ca}^{2+}]_i$  ( $\ll 10$  % of the response in rTRPA1 HEK-293 cells) and this effect was used as baseline and subtracted from the values obtained from transfected cells.



HEK-293 wt

The effect of AITC 100  $\mu\text{M}$  administered after a pre-incubation of TRPA1-HEK293 cells with moringin is completely inhibited.



TRPA1-HEK-293