

# 1 **Chemical composition of cold-pressed milk thistle seed flour** 2 **extract, and its potential health beneficial properties**

3 Uyory Choe,<sup>a,b</sup> Yanfang Li,<sup>c,d,e</sup> Boyan Gao,<sup>\*d</sup> Lu Yu,<sup>a,b</sup> Thomas T. Y. Wang,<sup>b</sup> Jianghao Sun,<sup>c</sup> Pei  
4 Chen,<sup>c</sup> Liangli (Lucy) Yu<sup>\*a</sup>

5 <sup>a</sup>Department of Nutrition and Food Science, University of Maryland, College Park, MD 20742,  
6 USA

7 <sup>b</sup>Diet, Genomics and Immunology Laboratory, Beltsville Human Nutrition Research Center,  
8 Agricultural Research Service, United States Department of Agriculture, Beltsville, MD 20705,  
9 USA

10 <sup>c</sup>Food Composition and Methods Development Laboratory, Beltsville Human Nutrition Research  
11 Center, Agricultural Research Service, United States Department of Agriculture, Beltsville, MD  
12 20705, USA

13 <sup>d</sup>Institute of Food and Nutraceutical Science, School of Agriculture and Biology, Shanghai Jiao  
14 Tong University, Shanghai 200240, China

15 <sup>e</sup>Beijing Advanced Innovation Center for Food Nutrition and Human Health, Beijing Technology  
16 & Business University (BTBU), Beijing 100048, China

17

18 <sup>\*</sup>Corresponding authors E-mail address: gaoboyan@sjtu.edu.cn (Boyan Gao); and  
19 lyu5@umd.edu (Liangli (Lucy) Yu)

20 **Supplemental materials:**

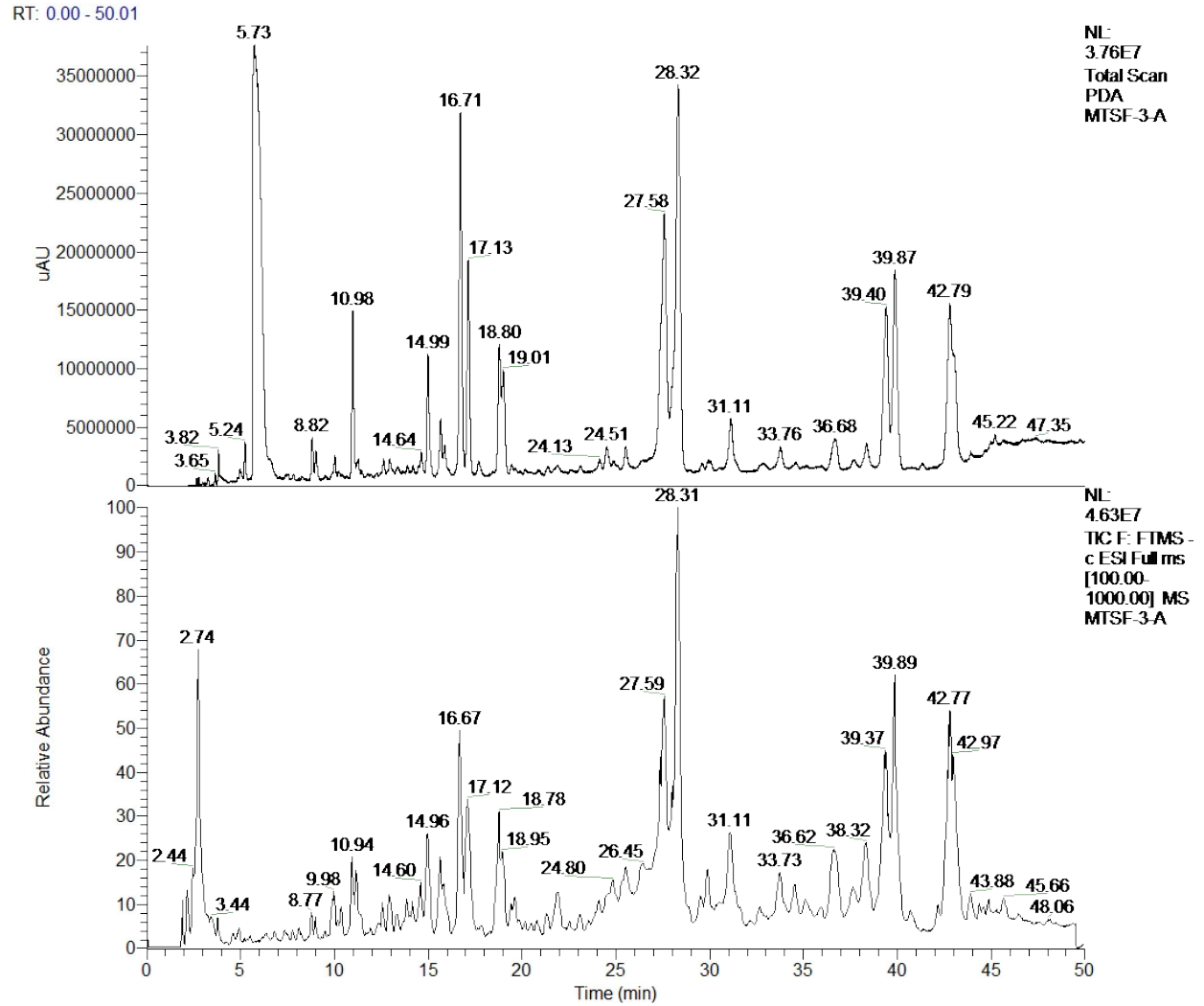
21 **Figure Captions**

22 **Supplementary Fig. S1** Typical UHPLC chromatogram and UHPLC-Orbitrap-MS chromatogram of milk  
23 thistle seed extract.

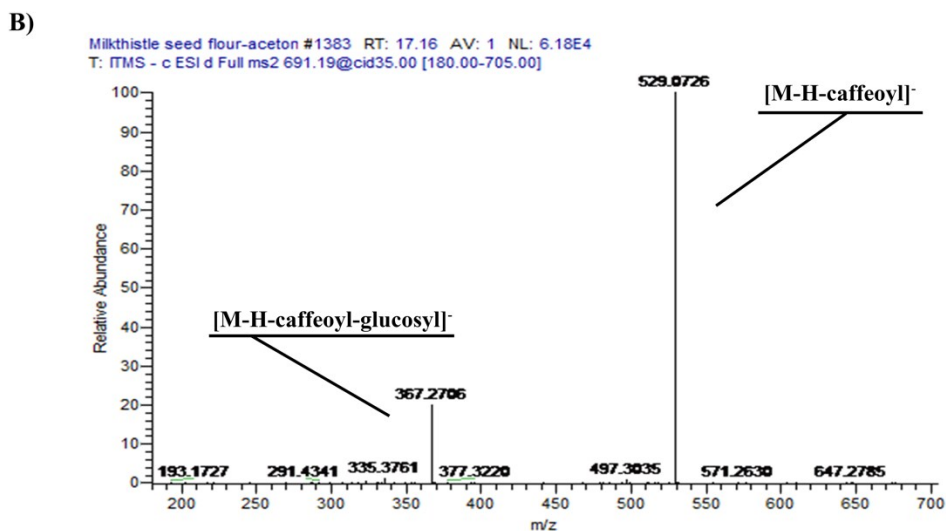
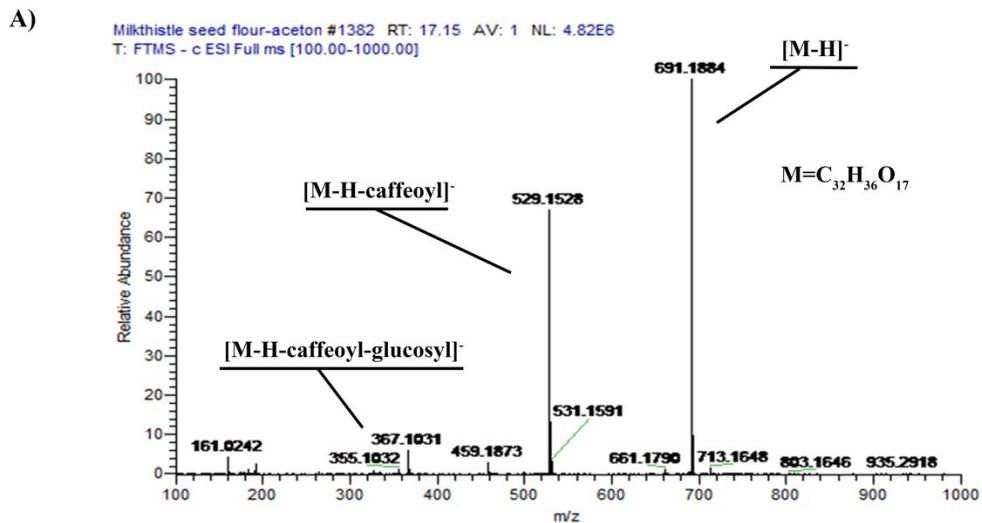
24 **Supplementary Fig. S2** Identification of compound 5, Methyl 5-(6-caffeoyl-glucopyranosyl)-  
25 caffeoylquinic acid. (A) MS spectrum; (B) MS<sup>2</sup> spectrum in negative mode. The high resolution  
26 ESI-MS of Methyl 5-(6-caffeoyl-glucopyranosyl)-caffeoylquinic acid showed the 691.1884 of  
27 [M-H]<sup>-</sup> corresponding to the formula of C<sub>32</sub>H<sub>36</sub>O<sub>17</sub> (0 ppm). The detailed analysis of the  
28 fragmental ions of Methyl 5-(6-caffeoyl-glucopyranosyl)-caffeoylquinic acid showed peaks of  
29 529.1528 [M-H-caffeoyl]<sup>-</sup> (C<sub>23</sub>H<sub>29</sub>O<sub>14</sub>) and 367.1031 [M-H-caffeoyl-glucosyl]<sup>-</sup> (C<sub>17</sub>H<sub>19</sub>O<sub>9</sub>) in  
30 negative mode.

31 **Supplementary Fig. S3** Anti-proliferative capacity of milk thistle seed flour extract in LNCaP  
32 prostate cancer cells. A final concentration of 0.4 mg flour equivalents/mL milk thistle seed  
33 flour extract was treated in LNCaP prostate cancer cells. Relative LNCaP prostate cancer cell  
34 numbers were measured every 24 h. Each column represents the mean ± SD (n = 3). Columns  
35 marked with different letters indicate significant difference ( $P \leq 0.05$ ).

**Supplementary Fig. S1** Typical UHPLC chromatogram and UHPLC-Obitrap-MS chromatogram of milk thistle seed extract.



**Supplementary Fig. S2** Identification of compound 5, Methyl 5-(6-caffeoyl-glucoyranosyl)-caffeoylquinic acid. (A) MS spectrum; (B) MS<sup>2</sup> spectrum in negative mode.



**Supplementary Fig. S3** Anti-proliferative capacity of milk thistle seed flour extract in LNCaP prostate cancer cells.

