

## SUPPLEMENTARY MATERIAL

**Table SI.** Selectivity index values for RAW 264.7 macrophages and different tumor cell lines.

| Cancer cell lines | Methanolic | Aqueous | Acetonitrile | Dichloromethane | Chloroform | Hexanic | Dox  | Ams  | Asu  |
|-------------------|------------|---------|--------------|-----------------|------------|---------|------|------|------|
| HepG2             | 2.92       | 3.05    | 2.99         | 2.84            | 2.76       | 2.67    | ≈1.0 | ≈1.0 | ≈1.0 |
| Jurkat            | 3.82       | 3.61    | 3.45         | 3.36            | 3.21       | 3.10    | ≈1.0 | ≈1.0 | ≈1.0 |
| HL-60             | 4.07       | 3.99    | 3.78         | 3.56            | 3.31       | 3.25    | ≈1.0 | ≈1.0 | ≈1.0 |
| K562              | 3.39       | 3.27    | 3.19         | 3.15            | 2.92       | 2.81    | ≈1.0 | ≈1.0 | ≈1.0 |
| HeLa              | 3.06       | 3.10    | 3.01         | 2.92            | 2.80       | 2.74    | ≈1.0 | ≈1.0 | ≈1.0 |
| HEp-2             | 3.20       | 3.16    | 3.07         | 2.99            | 2.89       | 2.82    | ≈1.0 | ≈1.0 | ≈1.0 |
| HCT-8             | 3.57       | 3.52    | 3.33         | 3.18            | 3.01       | 2.95    | ≈1.0 | ≈1.0 | ≈1.0 |
| HT-29             | 3.42       | 3.38    | 3.26         | 3.09            | 2.93       | 2.84    | ≈1.0 | ≈1.0 | ≈1.0 |
| HCT116            | 3.66       | 3.65    | 3.43         | 3.28            | 3.09       | 3.01    | ≈1.0 | ≈1.0 | ≈1.0 |
| MCF-7             | 3.26       | 3.25    | 3.15         | 3.03            | 2.88       | 2.80    | ≈1.0 | ≈1.0 | ≈1.0 |
| T-47D             | 3.41       | 3.41    | 3.31         | 3.11            | 2.99       | 2.91    | ≈1.0 | ≈1.0 | ≈1.0 |
| NCI-H292          | 3.13       | 3.14    | 3.06         | 2.95            | 2.82       | 2.76    | ≈1.0 | ≈1.0 | ≈1.0 |
| SF-295            | 3.77       | 3.74    | 3.59         | 3.39            | 3.19       | 3.09    | ≈1.0 | ≈1.0 | ≈1.0 |

SI =  $IC_{50}$  RAW 264.7 /  $IC_{50}$  tumor cells; Dox (doxorubicin), Ams (amsacrine) and Asu (asulacrine).

**Table SII.** Selectivity index values for J774-A1 macrophages and different tumor cell lines.

| Cancer cell lines | Methanolic | Aqueous | Acetonitrile | Dichloromethane | Chloroform | Hexanic | Dox  | Ams  | Asu  |
|-------------------|------------|---------|--------------|-----------------|------------|---------|------|------|------|
| HepG2             | 2.87       | 2.99    | 2.94         | 2.80            | 2.69       | 2.63    | ≈1.0 | ≈1.0 | ≈1.0 |
| Jurkat            | 3.76       | 3.53    | 3.39         | 3.33            | 3.12       | 3.06    | ≈1.0 | ≈1.0 | ≈1.0 |
| HL-60             | 4.00       | 3.90    | 3.72         | 3.52            | 3.22       | 3.20    | ≈1.0 | ≈1.0 | ≈1.0 |
| K562              | 3.33       | 3.20    | 3.14         | 3.12            | 2.84       | 2.77    | ≈1.0 | ≈1.0 | ≈1.0 |
| HeLa              | 3.01       | 3.04    | 2.96         | 2.89            | 2.73       | 2.70    | ≈1.0 | ≈1.0 | ≈1.0 |
| HEp-2             | 3.15       | 3.09    | 3.02         | 2.96            | 2.81       | 2.78    | ≈1.0 | ≈1.0 | ≈1.0 |
| HCT-8             | 3.51       | 3.45    | 3.27         | 3.15            | 2.93       | 2.91    | ≈1.0 | ≈1.0 | ≈1.0 |
| HT-29             | 3.36       | 3.31    | 3.21         | 3.06            | 2.86       | 2.80    | ≈1.0 | ≈1.0 | ≈1.0 |
| HCT116            | 3.60       | 3.57    | 3.37         | 3.25            | 3.01       | 2.96    | ≈1.0 | ≈1.0 | ≈1.0 |
| MCF-7             | 3.20       | 3.18    | 3.09         | 3.00            | 2.80       | 2.76    | ≈1.0 | ≈1.0 | ≈1.0 |
| T-47D             | 3.35       | 3.33    | 3.25         | 3.08            | 2.91       | 2.86    | ≈1.0 | ≈1.0 | ≈1.0 |
| NCI-H292          | 3.08       | 3.08    | 3.01         | 2.92            | 2.75       | 2.72    | ≈1.0 | ≈1.0 | ≈1.0 |
| SF-295            | 3.71       | 3.66    | 3.53         | 3.35            | 3.11       | 3.05    | ≈1.0 | ≈1.0 | ≈1.0 |

SI =  $IC_{50}$  J774-A1 /  $IC_{50}$  tumor cells; Dox (doxorubicin), Ams (amsacrine) and Asu (asulacrine).

**Table SIII. Selectivity index values for Vero cells and different tumor cell lines.**

| Cancer cell lines | Methanolic | Aqueous | Acetonitrile | Dichloromethane | Chloroform | Hexanic | Dox  | Ams  | Asu  |
|-------------------|------------|---------|--------------|-----------------|------------|---------|------|------|------|
| HepG2             | 2.74       | 2.85    | 2.82         | 2.69            | 2.59       | 2.51    | ≈1.0 | ≈1.0 | ≈1.0 |
| Jurkat            | 3.59       | 3.37    | 3.25         | 3.19            | 3.01       | 2.91    | ≈1.0 | ≈1.0 | ≈1.0 |
| HL-60             | 3.83       | 3.72    | 3.57         | 3.37            | 3.10       | 3.05    | ≈1.0 | ≈1.0 | ≈1.0 |
| K562              | 3.18       | 3.05    | 3.01         | 2.99            | 2.74       | 2.64    | ≈1.0 | ≈1.0 | ≈1.0 |
| HeLa              | 2.88       | 2.90    | 2.84         | 2.77            | 2.63       | 2.58    | ≈1.0 | ≈1.0 | ≈1.0 |
| HEp-2             | 3.01       | 2.95    | 2.89         | 2.84            | 2.71       | 2.65    | ≈1.0 | ≈1.0 | ≈1.0 |
| HCT-8             | 3.35       | 3.29    | 3.14         | 3.02            | 2.82       | 2.77    | ≈1.0 | ≈1.0 | ≈1.0 |
| HT-29             | 3.21       | 3.15    | 3.07         | 2.93            | 2.75       | 2.67    | ≈1.0 | ≈1.0 | ≈1.0 |
| HCT116            | 3.44       | 3.41    | 3.24         | 3.11            | 2.90       | 2.83    | ≈1.0 | ≈1.0 | ≈1.0 |
| MCF-7             | 3.06       | 3.04    | 2.97         | 2.87            | 2.70       | 2.64    | ≈1.0 | ≈1.0 | ≈1.0 |
| T-47D             | 3.20       | 3.18    | 3.12         | 2.95            | 2.81       | 2.73    | ≈1.0 | ≈1.0 | ≈1.0 |
| NCI-H292          | 2.94       | 2.93    | 2.89         | 2.79            | 2.64       | 2.60    | ≈1.0 | ≈1.0 | ≈1.0 |
| SF-295            | 3.54       | 3.49    | 3.39         | 3.21            | 3.00       | 2.91    | ≈1.0 | ≈1.0 | ≈1.0 |

SI =  $IC_{50}$  Vero cells /  $IC_{50}$  tumor cells; Dox (doxorubicin), Ams (amsacrine) and Asu (asulacrine).

**Table SIV. Selectivity index values for V79 fibroblasts (hamster) and different tumor cell lines.**

| Cancer cell lines | Methanolic | Aqueous | Acetonitrile | Dichloromethane | Chloroform | Hexanic | Dox  | Ams  | Asu  |
|-------------------|------------|---------|--------------|-----------------|------------|---------|------|------|------|
| HepG2             | 2.82       | 2.89    | 2.89         | 2.75            | 2.63       | 2.55    | ≈1.0 | ≈1.0 | ≈1.0 |
| Jurkat            | 3.70       | 3.42    | 3.33         | 3.26            | 3.06       | 2.96    | ≈1.0 | ≈1.0 | ≈1.0 |
| HL-60             | 3.94       | 3.78    | 3.66         | 3.45            | 3.16       | 3.10    | ≈1.0 | ≈1.0 | ≈1.0 |
| K562              | 3.27       | 3.10    | 3.08         | 3.06            | 2.79       | 2.68    | ≈1.0 | ≈1.0 | ≈1.0 |
| HeLa              | 2.96       | 2.94    | 2.91         | 2.83            | 2.68       | 2.62    | ≈1.0 | ≈1.0 | ≈1.0 |
| HEp-2             | 3.10       | 3.00    | 2.96         | 2.90            | 2.76       | 2.69    | ≈1.0 | ≈1.0 | ≈1.0 |
| HCT-8             | 3.45       | 3.34    | 3.22         | 3.08            | 2.87       | 2.82    | ≈1.0 | ≈1.0 | ≈1.0 |
| HT-29             | 3.30       | 3.20    | 3.15         | 2.99            | 2.80       | 2.71    | ≈1.0 | ≈1.0 | ≈1.0 |
| HCT116            | 3.54       | 3.47    | 3.31         | 3.18            | 2.95       | 2.87    | ≈1.0 | ≈1.0 | ≈1.0 |
| MCF-7             | 3.15       | 3.09    | 3.04         | 2.93            | 2.74       | 2.68    | ≈1.0 | ≈1.0 | ≈1.0 |
| T-47D             | 3.29       | 3.23    | 3.19         | 3.02            | 2.86       | 2.78    | ≈1.0 | ≈1.0 | ≈1.0 |
| NCI-H292          | 3.03       | 2.98    | 2.96         | 2.86            | 2.69       | 2.64    | ≈1.0 | ≈1.0 | ≈1.0 |
| SF-295            | 3.65       | 3.55    | 3.47         | 3.28            | 3.05       | 2.95    | ≈1.0 | ≈1.0 | ≈1.0 |

SI =  $IC_{50}$  V79 fibroblasts (hamster) /  $IC_{50}$  tumor cells; Dox (doxorubicin), Ams (amsacrine) and Asu (asulacrine).

**Table SV. Immunomodulatory activity results of the methanolic extract against mouse splenocytes.**

| Parameters           | Control       | 3.9 µg/mL    | 7.81 µg/mL   | 15.63 µg/mL  | 31.25 µg/mL  | 62.5 µg/mL   | 125 µg/mL    | 250 µg/mL    | 500 µg/mL    | 1000 µg/mL   |
|----------------------|---------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| PI (%)               | 100.00 ± 0.20 | 99.59 ± 0.1  | 99.28 ± 0.2  | 98.19 ± 0.12 | 97.65 ± 0.23 | 97.63 ± 0.1  | 96.82 ± 0.1  | 96.04 ± 0.3  | 95.69 ± 0.2  | 95.00 ± 0.21 |
| Annexin V (%)        | 100.00 ± 0.20 | 99.99 ± 0.1  | 99.05 ± 0.1  | 98.64 ± 0.6  | 97.75 ± 0.0  | 97.25 ± 0.3  | 97.15 ± 0.2  | 96.49 ± 0.1  | 95.95 ± 0.21 | 95.70 ± 0.25 |
| CFSE (%)             | 7.92 ± 0.0    | 7.91 ± 0.0   | 8.07 ± 0.0   | 9.23 ± 0.0   | 10.41 ± 0.06 | 11.43 ± 0.06 | 11.86 ± 0.06 | 11.95 ± 0.06 | 12.10 ± 0.06 | 12.62 ± 0.06 |
| IL-2                 | 11.84 ± 0.13  | 12.39 ± 0.13 | 12.81 ± 0.13 | 13.38 ± 0.13 | 13.76 ± 0.13 | 14.59 ± 0.13 | 15.04 ± 0.13 | 15.64 ± 0.13 | 15.91 ± 0.13 | 17.39 ± 0.13 |
| IL-4                 | 10.47 ± 0.17  | 11.08 ± 0.17 | 11.59 ± 0.17 | 11.87 ± 0.17 | 11.97 ± 0.17 | 13.51 ± 0.17 | 13.61 ± 0.17 | 13.67 ± 0.17 | 14.47 ± 0.17 | 14.90 ± 0.17 |
| IL-10                | 5.75 ± 0.06   | 6.32 ± 0.06  | 7.10 ± 0.06  | 7.82 ± 0.06  | 8.38 ± 0.06  | 8.88 ± 0.06  | 8.96 ± 0.06  | 9.31 ± 0.06  | 9.43 ± 0.06  | 11.30 ± 0.06 |
| IFN-γ                | 8.11 ± 0.12   | 8.72 ± 0.12  | 9.20 ± 0.12  | 10.15 ± 0.12 | 10.34 ± 0.12 | 10.45 ± 0.12 | 11.38 ± 0.12 | 12.47 ± 0.12 | 12.95 ± 0.12 | 13.86 ± 0.12 |
| IL-12 (pg/mL)        | 10.23 ± 0.16  | 10.90 ± 0.16 | 11.09 ± 0.16 | 11.71 ± 0.16 | 11.82 ± 0.16 | 13.50 ± 0.16 | 13.99 ± 0.16 | 14.11 ± 0.16 | 14.40 ± 0.16 | 14.48 ± 0.16 |
| IL-17 (pg/mL)        | 7.29 ± 0.07   | 7.68 ± 0.07  | 8.14 ± 0.07  | 9.17 ± 0.07  | 10.06 ± 0.07 | 10.17 ± 0.07 | 10.76 ± 0.07 | 11.82 ± 0.07 | 11.85 ± 0.07 | 12.14 ± 0.07 |
| TNF-α (pg/mL)        | 7.28 ± 0.10   | 8.12 ± 0.10  | 8.14 ± 0.10  | 8.76 ± 0.10  | 8.98 ± 0.10  | 10.20 ± 0.10 | 10.39 ± 0.10 | 11.01 ± 0.10 | 11.94 ± 0.10 | 12.20 ± 0.10 |
| TGF-β (pg/mL)        | 10.19 ± 0.09  | 10.22 ± 0.09 | 11.54 ± 0.09 | 11.81 ± 0.09 | 11.95 ± 0.09 | 12.40 ± 0.09 | 13.01 ± 0.09 | 14.12 ± 0.09 | 14.41 ± 0.09 | 14.86 ± 0.09 |
| NO                   | 0.0100 ± 0.0  | 0.0100 ± 0.0 | 0.0088 ± 0.0 | 0.0075 ± 0.0 | 0.0062 ± 0.0 | 0.0050 ± 0.0 | 0.0038 ± 0.0 | 0.0025 ± 0.0 | 0.0012 ± 0.0 | 0.0000 ± 0.0 |
| ROS mit.             | 8.68 ± 0.01   | 9.86 ± 0.01  | 9.88 ± 0.02  | 10.44 ± 0.08 | 10.91 ± 0.01 | 11.18 ± 0.00 | 12.45 ± 0.01 | 13.41 ± 0.01 | 13.55 ± 0.01 | 13.68 ± 0.04 |
| ROS cit.             | 11.45 ± 0.11  | 11.77 ± 0.15 | 12.62 ± 0.18 | 12.78 ± 0.1  | 13.95 ± 0.1  | 14.12 ± 0.1  | 14.57 ± 0.1  | 14.88 ± 0.1  | 15.71 ± 0.16 | 16.73 ± 0.15 |
| ΔΨ (mV)              | 13.17 ± 0.06  | 14.35 ± 0.06 | 14.63 ± 0.06 | 15.20 ± 0.06 | 15.25 ± 0.06 | 15.50 ± 0.06 | 17.08 ± 0.06 | 17.17 ± 0.06 | 17.69 ± 0.06 | 18.48 ± 0.06 |
| Ca <sup>2+</sup>     | 6.62 ± 0.09   | 7.73 ± 0.01  | 7.80 ± 0.06  | 8.67 ± 0.01  | 8.76 ± 0.03  | 8.89 ± 0.04  | 10.46 ± 0.09 | 11.14 ± 0.1  | 11.16 ± 0.0  | 11.42 ± 0.0  |
| CD4 <sup>+</sup> (%) | 8.25 ± 0.14   | 9.13 ± 0.14  | 9.95 ± 0.14  | 10.09 ± 0.14 | 10.91 ± 0.14 | 10.57 ± 0.14 | 11.64 ± 0.14 | 11.44 ± 0.14 | 12.30 ± 0.14 | 13.69 ± 0.14 |
| CD8 <sup>+</sup> (%) | 12.95 ± 0.0   | 13.33 ± 0.0  | 15.38 ± 0.0  | 14.62 ± 0.01 | 14.43 ± 0.0  | 15.68 ± 0.01 | 15.78 ± 0.1  | 17.31 ± 0.01 | 17.41 ± 0.04 | 17.88 ± 0.05 |

Mean ± Standard deviation. PI = propidium iodide; Annexin V = early apoptosis; CFSE = cell proliferation; IL (2,4,10,12 and 17) = interleukins; IFN-γ = interferon gamma; TNF-α = tumor necrosis factor alpha; TGF-β = transforming growth factor beta; NO = nitric oxide; ROS mit = mitochondrial reactive oxygen species; ROS cit. = cytosolic reactive oxygen species; ΔΨ = mitochondrial membrane potential; Ca<sup>2+</sup> = intracellular calcium; CD4<sup>+</sup> = helper T cells; CD8<sup>+</sup> = cytotoxic.

**Table SVI. Immunomodulatory activity results of the aqueous extract against mouse splenocytes.**

| Parameters           | Control       | 3.9 µg/mL    | 7.81 µg/mL   | 15.63 µg/mL  | 31.25 µg/mL  | 62.5 µg/mL   | 125 µg/mL    | 250 µg/mL    | 500 µg/mL    | 1000 µg/mL   |
|----------------------|---------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| PI (%)               | 100.00 ± 0.20 | 99.61 ± 0.20 | 98.68 ± 0.20 | 97.81 ± 0.1  | 97.47 ± 0.2  | 97.15 ± 0.2  | 96.77 ± 0.0  | 96.30 ± 0.1  | 95.47 ± 0.0  | 95.00 ± 0.1  |
| Annexin V (%)        | 100.00 ± 0.2  | 100.07 ± 0.0 | 99.46 ± 0.1  | 98.47 ± 0.2  | 98.28 ± 0.3  | 97.77 ± 0.8  | 96.91 ± 0.7  | 96.75 ± 0.3  | 96.14 ± 0.1  | 95.17 ± 0.3  |
| CFSE (%)             | 11.97 ± 0.15  | 11.99 ± 0.06 | 13.72 ± 0.04 | 13.86 ± 0.0  | 14.05 ± 0.0  | 14.87 ± 0.0  | 15.41 ± 0.0  | 15.86 ± 0.0  | 16.84 ± 0.0  | 17.69 ± 0.0  |
| IL-2                 | 12.02 ± 0.16  | 12.52 ± 0.13 | 13.20 ± 0.18 | 13.63 ± 0.1  | 13.73 ± 0.1  | 14.02 ± 0.1  | 15.42 ± 0.0  | 15.47 ± 0.2  | 16.44 ± 0.1  | 16.75 ± 0.1  |
| IL-4                 | 7.87 ± 0.1    | 8.16 ± 0.1   | 8.97 ± 0.1   | 9.05 ± 0.1   | 9.89 ± 0.1   | 11.25 ± 0.1  | 11.40 ± 0.1  | 11.67 ± 0.1  | 12.02 ± 0.1  | 14.06 ± 0.1  |
| IL-10                | 12.26 ± 0.0   | 13.33 ± 0.0  | 13.68 ± 0.0  | 14.11 ± 0.0  | 14.70 ± 0.0  | 14.97 ± 0.0  | 15.30 ± 0.0  | 15.35 ± 0.0  | 16.31 ± 0.0  | 18.09 ± 0.0  |
| IFN-γ                | 6.56 ± 0.1    | 6.82 ± 0.1   | 7.22 ± 0.1   | 8.66 ± 0.1   | 9.38 ± 0.1   | 9.50 ± 0.1   | 9.77 ± 0.1   | 10.70 ± 0.1  | 11.19 ± 0.1  | 11.92 ± 0.1  |
| IL-12 (pg/mL)        | 5.14 ± 0.1    | 5.00 ± 0.1   | 6.08 ± 0.1   | 7.32 ± 0.1   | 7.42 ± 0.1   | 7.70 ± 0.1   | 7.81 ± 0.1   | 8.58 ± 0.1   | 8.96 ± 0.1   | 9.49 ± 0.1   |
| IL-17 (pg/mL)        | 11.35 ± 0.1   | 11.79 ± 0.1  | 12.18 ± 0.0  | 13.00 ± 0.2  | 14.30 ± 0.1  | 14.71 ± 0.1  | 15.12 ± 0.1  | 15.12 ± 0.3  | 15.82 ± 0.4  | 16.67 ± 0.7  |
| TNF-α (pg/mL)        | 8.88 ± 0.1    | 9.44 ± 0.1   | 9.63 ± 0.1   | 11.41 ± 0.1  | 11.56 ± 0.1  | 11.67 ± 0.1  | 12.41 ± 0.1  | 12.70 ± 0.1  | 13.07 ± 0.1  | 13.98 ± 0.1  |
| TGF-β (pg/mL)        | 13.45 ± 0.1   | 13.64 ± 0.1  | 14.33 ± 0.1  | 15.55 ± 0.1  | 16.36 ± 0.1  | 16.38 ± 0.1  | 17.01 ± 0.1  | 17.05 ± 0.1  | 18.33 ± 0.1  | 18.73 ± 0.1  |
| NO                   | 0.0100 ± 0.0  | 0.0100 ± 0.0 | 0.0088 ± 0.0 | 0.0075 ± 0.0 | 0.0062 ± 0.0 | 0.0050 ± 0.0 | 0.0038 ± 0.0 | 0.0025 ± 0.0 | 0.0012 ± 0.0 | 0.0000 ± 0.0 |
| ROS mit.             | 12.51 ± 0.1   | 12.81 ± 0.1  | 13.70 ± 0.19 | 14.14 ± 0.19 | 15.00 ± 0.19 | 15.16 ± 0.19 | 16.27 ± 0.19 | 16.30 ± 0.19 | 17.37 ± 0.19 | 17.37 ± 0.19 |
| ROS cit.             | 14.51 ± 0.1   | 14.70 ± 0.1  | 15.80 ± 0.1  | 16.53 ± 0.1  | 16.67 ± 0.1  | 17.04 ± 0.1  | 17.79 ± 0.1  | 18.42 ± 0.1  | 19.33 ± 0.1  | 19.84 ± 0.1  |
| ΔΨ (mV)              | 5.91 ± 0.1    | 6.06 ± 0.1   | 6.49 ± 0.1   | 6.97 ± 0.1   | 8.51 ± 0.1   | 8.98 ± 0.1   | 9.62 ± 0.1   | 10.09 ± 0.1  | 10.69 ± 0.1  | 11.03 ± 0.1  |
| Ca <sup>2+</sup>     | 12.06 ± 0.1   | 12.66 ± 0.1  | 13.45 ± 0.1  | 13.70 ± 0.1  | 13.92 ± 0.1  | 14.35 ± 0.1  | 15.49 ± 0.1  | 16.35 ± 0.1  | 16.72 ± 0.1  | 17.06 ± 0.1  |
| CD4 <sup>+</sup> (%) | 5.56 ± 0.1    | 6.70 ± 0.1   | 7.01 ± 0.1   | 7.43 ± 0.1   | 7.79 ± 0.1   | 7.59 ± 0.1   | 8.45 ± 0.1   | 9.26 ± 0.1   | 10.23 ± 0.1  | 10.59 ± 0.2  |
| CD8 <sup>+</sup> (%) | 5.74 ± 0.1    | 6.73 ± 0.1   | 7.04 ± 0.1   | 7.39 ± 0.1   | 9.07 ± 0.1   | 8.69 ± 0.1   | 8.84 ± 0.1   | 10.54 ± 0.1  | 10.78 ± 0.1  | 11.65 ± 0.1  |

Mean ± Standard deviation. PI = propidium iodide; Annexin V = early apoptosis; CFSE = cell proliferation; IL (2,4,10,12 and 17) = interleukins; IFN-γ = interferon gamma; TNF-α = tumor necrosis factor alpha; TGF-β = transforming growth factor beta; NO = nitric oxide; ROS mit = mitochondrial reactive oxygen species; ROS cit. = cytosolic reactive oxygen species; ΔΨ = mitochondrial membrane potential; Ca<sup>2+</sup> = intracellular calcium; CD4<sup>+</sup> = helper T cells; CD8<sup>+</sup> = cytotoxic.

**Table SVII. Immunomodulatory activity results of the acetonitrile extract against mouse splenocytes.**

| Parameters           | Control       | 3.9 µg/mL    | 7.81 µg/mL   | 15.63 µg/mL  | 31.25 µg/mL  | 62.5 µg/mL   | 125 µg/mL    | 250 µg/mL    | 500 µg/mL    | 1000 µg/mL   |
|----------------------|---------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| PI (%)               | 100.00 ± 0.20 | 99.40 ± 0.20 | 98.58 ± 0.2  | 98.11 ± 0.0  | 97.08 ± 0.1  | 97.00 ± 0.2  | 96.38 ± 0.2  | 96.20 ± 0.1  | 95.84 ± 0.2  | 95.00 ± 0.1  |
| Annexin V (%)        | 100.00 ± 0.1  | 99.76 ± 0.2  | 99.71 ± 0.1  | 99.36 ± 0.1  | 97.87 ± 0.1  | 97.87 ± 0.1  | 96.91 ± 0.1  | 96.46 ± 0.1  | 96.37 ± 0.2  | 96.10 ± 0.2  |
| CFSE (%)             | 14.87 ± 0.18  | 15.38 ± 0.01 | 15.52 ± 0.01 | 16.67 ± 0.06 | 17.01 ± 0.01 | 17.52 ± 0.0  | 17.76 ± 0.0  | 18.32 ± 0.0  | 18.86 ± 0.0  | 19.82 ± 0.0  |
| IL-2                 | 13.13 ± 0.10  | 13.60 ± 0.1  | 14.91 ± 0.1  | 14.95 ± 0.1  | 15.08 ± 0.1  | 15.16 ± 0.1  | 15.76 ± 0.1  | 17.27 ± 0.1  | 18.10 ± 0.1  | 18.90 ± 0.1  |
| IL-4                 | 6.14 ± 0.05   | 6.76 ± 0.0   | 7.82 ± 0.0   | 8.23 ± 0.01  | 8.97 ± 0.02  | 9.10 ± 0.0   | 10.03 ± 0.0  | 10.53 ± 0.0  | 10.63 ± 0.0  | 10.66 ± 0.0  |
| IL-10                | 12.55 ± 0.1   | 12.92 ± 0.1  | 14.36 ± 0.1  | 14.52 ± 0.1  | 14.61 ± 0.1  | 15.00 ± 0.1  | 15.13 ± 0.1  | 17.15 ± 0.1  | 17.43 ± 0.1  | 17.48 ± 0.1  |
| IFN-γ                | 10.14 ± 0.1   | 10.40 ± 0.1  | 11.87 ± 0.1  | 12.26 ± 0.1  | 12.49 ± 0.1  | 12.88 ± 0.1  | 13.75 ± 0.1  | 13.85 ± 0.1  | 13.87 ± 0.1  | 15.14 ± 0.1  |
| IL-12 (pg/mL)        | 11.33 ± 0.0   | 12.03 ± 0.0  | 12.33 ± 0.0  | 12.92 ± 0.0  | 12.97 ± 0.0  | 14.32 ± 0.0  | 15.02 ± 0.0  | 15.06 ± 0.0  | 16.17 ± 0.0  | 16.80 ± 0.0  |
| IL-17 (pg/mL)        | 6.92 ± 0.0    | 7.01 ± 0.0   | 7.70 ± 0.0   | 7.83 ± 0.0   | 9.01 ± 0.0   | 9.09 ± 0.0   | 9.70 ± 0.0   | 10.74 ± 0.0  | 11.38 ± 0.0  | 12.18 ± 0.0  |
| TNF-α (pg/mL)        | 13.74 ± 0.1   | 14.84 ± 0.1  | 15.05 ± 0.1  | 15.23 ± 0.1  | 15.56 ± 0.1  | 16.78 ± 0.1  | 16.78 ± 0.1  | 17.00 ± 0.1  | 18.58 ± 0.1  | 19.36 ± 0.1  |
| TGF-β (pg/mL)        | 9.63 ± 0.11   | 10.20 ± 0.11 | 10.56 ± 0.11 | 11.27 ± 0.11 | 11.89 ± 0.11 | 12.30 ± 0.11 | 12.84 ± 0.11 | 12.98 ± 0.11 | 14.22 ± 0.11 | 14.27 ± 0.11 |
| NO                   | 0.0100 ± 0.0  | 0.0100 ± 0.0 | 0.0088 ± 0.0 | 0.0075 ± 0.0 | 0.0062 ± 0.0 | 0.0050 ± 0.0 | 0.0038 ± 0.0 | 0.0025 ± 0.0 | 0.0012 ± 0.0 | 0.0000 ± 0.0 |
| ROS mit.             | 10.87 ± 0.0   | 11.15 ± 0.0  | 12.30 ± 0.0  | 12.72 ± 0.0  | 12.72 ± 0.0  | 13.53 ± 0.0  | 13.79 ± 0.0  | 14.32 ± 0.0  | 15.78 ± 0.0  | 15.88 ± 0.0  |
| ROS cit.             | 6.02 ± 0.1    | 6.58 ± 0.1   | 6.85 ± 0.1   | 7.33 ± 0.2   | 8.02 ± 0.2   | 8.56 ± 0.2   | 8.92 ± 0.2   | 9.21 ± 0.2   | 9.62 ± 0.2   | 11.36 ± 0.1  |
| ΔΨ (mV)              | 6.12 ± 0.0    | 6.82 ± 0.0   | 7.14 ± 0.0   | 7.86 ± 0.0   | 8.44 ± 0.0   | 8.51 ± 0.0   | 9.17 ± 0.0   | 10.26 ± 0.0  | 10.66 ± 0.0  | 12.14 ± 0.0  |
| Ca <sup>2+</sup>     | 12.69 ± 0.1   | 13.47 ± 0.1  | 13.81 ± 0.1  | 14.00 ± 0.1  | 14.07 ± 0.1  | 15.02 ± 0.1  | 15.77 ± 0.1  | 16.37 ± 0.1  | 16.41 ± 0.1  | 18.09 ± 0.1  |
| CD4 <sup>+</sup> (%) | 6.54 ± 0.1    | 8.43 ± 0.1   | 6.80 ± 0.11  | 8.09 ± 0.1   | 8.52 ± 0.1   | 8.99 ± 0.1   | 10.23 ± 0.1  | 10.97 ± 0.1  | 11.36 ± 0.1  | 11.18 ± 0.1  |
| CD8 <sup>+</sup> (%) | 8.49 ± 0.01   | 10.21 ± 0.07 | 9.64 ± 0.03  | 10.16 ± 0.07 | 10.92 ± 0.05 | 12.03 ± 0.03 | 11.74 ± 0.0  | 12.42 ± 0.07 | 12.63 ± 0.01 | 13.78 ± 0.04 |

Mean ± Standard deviation. PI = propidium iodide; Annexin V = early apoptosis; CFSE = cell proliferation; IL (2,4,10,12 and 17) = interleukins; IFN-γ = interferon gamma; TNF-α = tumor necrosis factor alpha; TGF-β = transforming growth factor beta; NO = nitric oxide; ROS mit = mitochondrial reactive oxygen species; ROS cit. = cytosolic reactive oxygen species; ΔΨ = mitochondrial membrane potential; Ca<sup>2+</sup> = intracellular calcium; CD4<sup>+</sup> = helper T cells; CD8<sup>+</sup> = cytotoxic.

**Table SVIII. Immunomodulatory activity results of the dichloromethane extract against mouse splenocytes.**

| Parameters           | Control       | 3.9 µg/mL    | 7.81 µg/mL   | 15.63µg/mL   | 31.25µg/mL   | 62.5 µg/mL   | 125 µg/mL    | 250 µg/mL    | 500 µg/mL    | 1000 µg/mL   |
|----------------------|---------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| PI (%)               | 100.00 ± 0.10 | 99.59 ± 0.19 | 99.28 ± 0.23 | 98.19 ± 0.20 | 97.65 ± 0.19 | 97.63 ± 0.16 | 96.82 ± 0.21 | 96.04 ± 0.16 | 95.69 ± 0.27 | 95.00 ± 0.29 |
| Annexin V (%)        | 100.00 ± 0.10 | 99.99 ± 0.15 | 99.05 ± 0.23 | 98.64 ± 0.05 | 97.75 ± 0.13 | 97.25 ± 0.09 | 97.15 ± 0.07 | 96.49 ± 0.10 | 95.95 ± 0.14 | 95.70 ± 0.15 |
| CFSE (%)             | 7.92 ± 0.10   | 7.91 ± 0.16  | 8.07 ± 0.06  | 9.23 ± 0.19  | 10.41 ± 0.16 | 11.43 ± 0.16 | 11.86 ± 0.13 | 11.95 ± 0.10 | 12.10 ± 0.20 | 12.62 ± 0.12 |
| IL-2                 | 11.84 ± 0.10  | 12.39 ± 0.19 | 12.81 ± 0.23 | 13.38 ± 0.12 | 13.76 ± 0.18 | 14.59 ± 0.27 | 15.04 ± 0.27 | 15.64 ± 0.08 | 15.91 ± 0.10 | 17.39 ± 0.06 |
| IL-4                 | 10.47 ± 0.10  | 11.08 ± 0.29 | 11.59 ± 0.19 | 11.87 ± 0.29 | 11.97 ± 0.23 | 13.51 ± 0.22 | 13.61 ± 0.10 | 13.67 ± 0.29 | 14.47 ± 0.05 | 14.90 ± 0.11 |
| IL-10                | 5.75 ± 0.10   | 6.32 ± 0.11  | 7.10 ± 0.27  | 7.82 ± 0.10  | 8.38 ± 0.28  | 8.88 ± 0.17  | 8.96 ± 0.20  | 9.31 ± 0.24  | 9.43 ± 0.18  | 11.30 ± 0.12 |
| IFN-γ                | 8.11 ± 0.10   | 8.72 ± 0.27  | 9.20 ± 0.13  | 10.15 ± 0.26 | 10.34 ± 0.06 | 10.45 ± 0.08 | 11.38 ± 0.20 | 12.47 ± 0.18 | 12.95 ± 0.15 | 13.86 ± 0.13 |
| IL-12 (pg/mL)        | 10.23 ± 0.10  | 10.90 ± 0.07 | 11.09 ± 0.24 | 11.71 ± 0.16 | 11.82 ± 0.23 | 13.50 ± 0.29 | 13.99 ± 0.18 | 14.11 ± 0.18 | 14.40 ± 0.07 | 14.48 ± 0.12 |
| IL-17 (pg/mL)        | 7.29 ± 0.10   | 7.68 ± 0.27  | 8.14 ± 0.29  | 9.17 ± 0.27  | 10.06 ± 0.18 | 10.17 ± 0.11 | 10.76 ± 0.05 | 11.82 ± 0.16 | 11.85 ± 0.15 | 12.14 ± 0.18 |
| TNF-α (pg/mL)        | 7.28 ± 0.10   | 8.12 ± 0.05  | 8.14 ± 0.18  | 8.76 ± 0.17  | 8.98 ± 0.08  | 10.20 ± 0.09 | 10.39 ± 0.10 | 11.01 ± 0.15 | 11.94 ± 0.11 | 12.20 ± 0.07 |
| TGF-β (pg/mL)        | 10.19 ± 0.10  | 10.22 ± 0.24 | 11.54 ± 0.06 | 11.81 ± 0.21 | 11.95 ± 0.24 | 12.40 ± 0.17 | 13.01 ± 0.11 | 14.12 ± 0.10 | 14.41 ± 0.24 | 14.86 ± 0.09 |
| NO                   | 0.0100 ± 0.0  | 0.0100 ± 0.0 | 0.0088 ± 0.0 | 0.0075 ± 0.0 | 0.0062 ± 0.0 | 0.0050 ± 0.0 | 0.0038 ± 0.0 | 0.0025 ± 0.0 | 0.0012 ± 0.0 | 0.0000 ± 0.0 |
| ROS mit.             | 8.68 ± 0.10   | 9.86 ± 0.09  | 9.88 ± 0.24  | 10.44 ± 0.12 | 10.91 ± 0.18 | 11.18 ± 0.05 | 12.45 ± 0.28 | 13.41 ± 0.28 | 13.55 ± 0.06 | 13.68 ± 0.29 |
| ROS cit.             | 11.45 ± 0.10  | 11.77 ± 0.24 | 12.62 ± 0.11 | 12.78 ± 0.26 | 13.95 ± 0.29 | 14.12 ± 0.29 | 14.57 ± 0.16 | 14.88 ± 0.20 | 15.71 ± 0.24 | 16.73 ± 0.21 |
| ΔΨ (mV)              | 13.17 ± 0.10  | 14.35 ± 0.18 | 14.63 ± 0.24 | 15.20 ± 0.27 | 15.25 ± 0.05 | 15.50 ± 0.13 | 17.08 ± 0.29 | 17.17 ± 0.18 | 17.69 ± 0.13 | 18.48 ± 0.18 |
| Ca <sup>2+</sup>     | 6.62 ± 0.10   | 7.73 ± 0.26  | 7.80 ± 0.09  | 8.67 ± 0.06  | 8.76 ± 0.14  | 8.89 ± 0.12  | 10.46 ± 0.18 | 11.14 ± 0.13 | 11.16 ± 0.13 | 11.42 ± 0.08 |
| CD4 <sup>+</sup> (%) | 8.25 ± 0.10   | 9.13 ± 0.11  | 9.95 ± 0.18  | 10.09 ± 0.19 | 10.91 ± 0.06 | 10.57 ± 0.14 | 11.64 ± 0.11 | 11.44 ± 0.22 | 12.30 ± 0.09 | 13.69 ± 0.07 |
| CD8 <sup>+</sup> (%) | 12.95 ± 0.10  | 13.33 ± 0.12 | 15.38 ± 0.18 | 14.62 ± 0.10 | 14.43 ± 0.07 | 15.68 ± 0.25 | 15.78 ± 0.21 | 17.31 ± 0.21 | 17.41 ± 0.19 | 17.88 ± 0.06 |

Mean ± Standard deviation. PI = propidium iodide; Annexin V = early apoptosis; CFSE = cell proliferation; IL (2,4,10,12 and 17) = interleukins; IFN-γ = interferon gamma; TNF-α = tumor necrosis factor alpha; TGF-β = transforming growth factor beta; NO = nitric oxide; ROS mit = mitochondrial reactive oxygen species; ROS cit. = cytosolic reactive oxygen species; ΔΨ = mitochondrial membrane potential; Ca<sup>2+</sup> = intracellular calcium; CD4<sup>+</sup> = helper T cells; CD8<sup>+</sup> = cytotoxic.

**Table SIX.** Immunomodulatory activity results of the hexanic extract against mouse splenocytes.

| Parameters           | Control      | 3.9 µg/mL    | 7.81 µg/mL   | 15.63 µg/mL  | 31.25 µg/mL  | 62.5 µg/mL   | 125 µg/mL    | 250 µg/mL    | 500 µg/mL    | 1000 µg/mL   |
|----------------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| PI (%)               | 100 ± 0.20   | 99.73 ± 0.12 | 99.41 ± 0.18 | 98.54 ± 0.10 | 97.83 ± 0.25 | 97.44 ± 0.18 | 96.52 ± 0.12 | 95.77 ± 0.15 | 95.23 ± 0.20 | 94.61 ± 0.22 |
| Annexin V (%)        | 100 ± 0.20   | 99.78 ± 0.10 | 99.23 ± 0.14 | 98.60 ± 0.11 | 97.92 ± 0.20 | 97.35 ± 0.18 | 96.77 ± 0.12 | 96.01 ± 0.14 | 95.55 ± 0.20 | 95.12 ± 0.23 |
| CFSE (%)             | 7.92 ± 0.0   | 8.01 ± 0.0   | 8.27 ± 0.0   | 9.45 ± 0.06  | 10.56 ± 0.06 | 11.12 ± 0.06 | 11.75 ± 0.06 | 12.04 ± 0.06 | 12.32 ± 0.06 | 12.81 ± 0.06 |
| IL-2                 | 11.84 ± 0.13 | 12.18 ± 0.13 | 12.65 ± 0.13 | 13.02 ± 0.13 | 13.78 ± 0.13 | 14.11 ± 0.13 | 14.77 ± 0.13 | 15.10 ± 0.13 | 15.83 ± 0.13 | 16.54 ± 0.13 |
| IL-4                 | 10.47 ± 0.17 | 10.95 ± 0.17 | 11.36 ± 0.17 | 11.68 ± 0.17 | 12.18 ± 0.17 | 12.86 ± 0.17 | 13.37 ± 0.17 | 13.95 ± 0.17 | 14.23 ± 0.17 | 14.71 ± 0.17 |
| IL-10                | 5.75 ± 0.06  | 6.18 ± 0.06  | 6.77 ± 0.06  | 7.43 ± 0.06  | 8.09 ± 0.06  | 8.63 ± 0.06  | 8.92 ± 0.06  | 9.20 ± 0.06  | 9.66 ± 0.06  | 10.85 ± 0.06 |
| IFN-γ                | 8.11 ± 0.12  | 8.58 ± 0.12  | 9.09 ± 0.12  | 9.96 ± 0.12  | 10.25 ± 0.12 | 10.78 ± 0.12 | 11.23 ± 0.12 | 11.91 ± 0.12 | 12.47 ± 0.12 | 13.32 ± 0.12 |
| IL-12 (pg/mL)        | 10.23 ± 0.16 | 10.71 ± 0.16 | 11.02 ± 0.16 | 11.45 ± 0.16 | 11.97 ± 0.16 | 12.85 ± 0.16 | 13.44 ± 0.16 | 13.97 ± 0.16 | 14.18 ± 0.16 | 14.45 ± 0.16 |
| IL-17 (pg/mL)        | 7.29 ± 0.07  | 7.55 ± 0.07  | 7.94 ± 0.07  | 8.86 ± 0.07  | 9.73 ± 0.07  | 10.01 ± 0.07 | 10.48 ± 0.07 | 11.12 ± 0.07 | 11.63 ± 0.07 | 11.97 ± 0.07 |
| TNF-α (pg/mL)        | 7.28 ± 0.10  | 7.85 ± 0.10  | 8.09 ± 0.10  | 8.64 ± 0.10  | 8.96 ± 0.10  | 9.73 ± 0.10  | 10.12 ± 0.10 | 10.80 ± 0.10 | 11.55 ± 0.10 | 11.88 ± 0.10 |
| TGF-β (pg/mL)        | 10.19 ± 0.09 | 10.35 ± 0.09 | 10.87 ± 0.09 | 11.43 ± 0.09 | 11.78 ± 0.09 | 12.10 ± 0.09 | 12.91 ± 0.09 | 13.64 ± 0.09 | 14.03 ± 0.09 | 14.52 ± 0.09 |
| NO                   | 0.0100 ± 0.0 | 0.0095 ± 0.0 | 0.0087 ± 0.0 | 0.0074 ± 0.0 | 0.0065 ± 0.0 | 0.0053 ± 0.0 | 0.0041 ± 0.0 | 0.0028 ± 0.0 | 0.0015 ± 0.0 | 0.0000 ± 0.0 |
| ROS mit.             | 8.68 ± 0.01  | 9.24 ± 0.01  | 9.76 ± 0.02  | 10.33 ± 0.08 | 10.82 ± 0.01 | 11.35 ± 0.00 | 11.99 ± 0.01 | 12.84 ± 0.01 | 13.42 ± 0.01 | 13.56 ± 0.04 |
| ROS cit.             | 11.45 ± 0.11 | 11.68 ± 0.15 | 12.33 ± 0.18 | 12.97 ± 0.10 | 13.75 ± 0.10 | 14.02 ± 0.10 | 14.42 ± 0.10 | 14.89 ± 0.10 | 15.43 ± 0.16 | 16.20 ± 0.15 |
| ΔΨ (mV)              | 13.17 ± 0.06 | 14.06 ± 0.06 | 14.58 ± 0.06 | 15.07 ± 0.06 | 15.43 ± 0.06 | 15.89 ± 0.06 | 16.57 ± 0.06 | 17.05 ± 0.06 | 17.46 ± 0.06 | 18.21 ± 0.06 |
| Ca <sup>2+</sup>     | 6.62 ± 0.09  | 7.31 ± 0.01  | 7.85 ± 0.06  | 8.33 ± 0.01  | 8.70 ± 0.03  | 9.04 ± 0.04  | 9.97 ± 0.09  | 10.73 ± 0.10 | 11.05 ± 0.0  | 11.29 ± 0.0  |
| CD4 <sup>+</sup> (%) | 8.25 ± 0.14  | 8.77 ± 0.14  | 9.42 ± 0.14  | 9.95 ± 0.14  | 10.44 ± 0.14 | 10.82 ± 0.14 | 11.19 ± 0.14 | 11.57 ± 0.14 | 12.05 ± 0.14 | 12.89 ± 0.14 |
| CD8 <sup>+</sup> (%) | 12.95 ± 0.0  | 13.11 ± 0.0  | 13.77 ± 0.0  | 14.25 ± 0.01 | 14.89 ± 0.0  | 15.27 ± 0.01 | 15.83 ± 0.1  | 16.47 ± 0.01 | 17.03 ± 0.04 | 17.49 ± 0.05 |

Mean ± Standard deviation. PI = propidium iodide; Annexin V = early apoptosis; CFSE = cell proliferation; IL (2,4,10,12 and 17) = interleukins; IFN-γ = interferon gamma; TNF-α = tumor necrosis factor alpha; TGF-β = transforming growth factor beta; NO = nitric oxide; ROS mit = mitochondrial reactive oxygen species; ROS cit. = cytosolic reactive oxygen species; ΔΨ = mitochondrial membrane potential; Ca<sup>2+</sup> = intracellular calcium; CD4<sup>+</sup> = helper T cells; CD8<sup>+</sup> = cytotoxic.