

*Supporting Information for*

**Novel anti-hyperuricemia hexapeptides derived from *Apostichopus japonicus* hydrolysate and their modulation effects on gut microbiota and host microRNA profile**

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**Table S1.** Primers used in this study.

|                | Forward primer                                      | Reverse primer  |
|----------------|---|---|
| XOD            | CGCAGAATACTGGATGAGCGAGGT                            | GCCGGTGGGTTTCTTCTTCTTGAA                                |
| ADA            | AATAAGCTCCAAGACCAAGGT                               | GTCCAGCAGACTCAATACACAC                                  |
| GLUT9          | ATGCACTGGCCACAGATTCA                                | TGAGGAACCAGATCGGAGGT                                    |
| URAT1          | ATCATCTCCATGCTGTGCTG                                | AAGTCCACAATCCCGATGAG                                    |
| ABCG2          | GGACCGCGAGAAAGGCATA                                 | ACGTGGTCATTACTGGAAGACA<br>AGTTCTTTATCCCAGTACCGTTGA<br>A |
| MRP4           | GGTGAAACCCAACCCGCTG                                 | A   |
| TNF- $\alpha$  | CCCTCCTAACCCGTTTTGCT                                | GACGTGGAAGTGGCAGAAGA                                    |
| IL-1 $\beta$   | CACAGCAGCACATCAACAAG                                | GTGCTCATGTCCTCATCCTG                                    |
| IL-10          | AGAGGGTTCCCCTACTGTCA                                | GGCCACAGTTTTTCAGGGATG                                   |
| NLRP3          | CAGACCTCCAAGACCACGACTG                              | CATCCGCAGCCAATGAACAGAG                                  |
| ACS            | AACCCAAGCAAGATGCGGAAG                               | TTAGGGCCTGGAGGAGCAAG                                    |
| Caspase-1      | TCCAATAATGCAAGTCAAGCC                               | GCTGTACCCAGATTTTGTAGCA                                  |
| TLR4           | CCGCATGGTGGTGGTTGTT                                 | GGAATCAGTCGCTTCTGTTGGA                                  |
| Myd88          | CTACAGAGCAAGGAATGTGACT<br>CTCAGCGCTGTGCAAATATATATCC | ACCTGATGCCATTTGCTGTCC<br>GGCGTATTGTACCCTGGAAGGG         |
| TRAF6          | C   |   |
| iNOS           | ACAGGAGAAGGGGACGAACT                                | GGCTGGACTTTTCACTCTGC                                    |
| COX-2          | GTGCGACATACTCAAGCAGG                                | TCAGGTGTTGCACGTAGTCT                                    |
| $\beta$ -actin | CAGGCATTGCTGACAGGATG                                | TGCTGATCCACATCTGCTGG                                    |

**Table S2.** The peptide composition in AJOP.

| Sequence      | Content (%) |
|---------------|-------------|
| IEPLV         | 6.382       |
| IIILV         | 2.374       |
| ILLLV         | 1.037       |
| LPELV         | 1.384       |
| <b>GPSGRP</b> | 2.932       |
| KIGRP         | 2.555       |
| <b>GPAGPR</b> | 2.275       |
| GGGKHV        | 2.901       |
| PKGRP         | 2.274       |
| KPGPR         | 1.593       |
| PQGRP         | 3.271       |
| PEILV         | 3.178       |
| PNKPV         | 1.101       |

**Table S3.** The molecular docking results between XOD and GPAGPR and GPSGRP.

| Peptide<br>Abbreviation | Peptide Name            | -CIE<br>(kJ/mol) | -E <sub>VDW</sub><br>(kJ/mol) |
|-------------------------|-------------------------|------------------|-------------------------------|
| GPAGPR                  | Gly-Pro-Ala-Gly-Pro-Arg | 99.500           | 12.484                        |
| GPSGRP                  | Gly-Pro-Ser-Gly-Arg-Pro | 92.096           | 12.293                        |

**Table S4.** Bacterial taxonomic profiles at the phylum level in response to the peptide treatments in the HUA mouse model. Data are represented as the mean  $\pm$  SD (n=5).

|                 | <b>control</b>   | <b>model</b>      | <b>GPAGPR</b>     | <b>GPSGRP</b>     |
|-----------------|------------------|-------------------|-------------------|-------------------|
| Bacteroidetes   | 71.05 $\pm$ 9.16 | 52.66 $\pm$ 10.86 | 29.84 $\pm$ 6.51  | 50.59 $\pm$ 12.13 |
| Firmicutes      | 18.03 $\pm$ 4.76 | 24.34 $\pm$ 8.4   | 31.85 $\pm$ 9.34  | 42.3 $\pm$ 11.31  |
| Verrucomicrobia | 6.02 $\pm$ 5.48  | 8.49 $\pm$ 8.19   | 30.72 $\pm$ 11.22 | 0.36 $\pm$ 0.6    |
| Actinobacteria  | 1.4 $\pm$ 0.65   | 7.45 $\pm$ 5.9    | 2.97 $\pm$ 1.65   | 1.47 $\pm$ 0.52   |
| Proteobacteria  | 1.31 $\pm$ 0.39  | 3.88 $\pm$ 2.29   | 1.47 $\pm$ 0.54   | 3.07 $\pm$ 2.18   |
| Patescibacteria | 1.05 $\pm$ 0.71  | 1.2 $\pm$ 0.51    | 2.52 $\pm$ 0.46   | 1.39 $\pm$ 0.48   |
| others          | 1.15 $\pm$ 0.23  | 1.99 $\pm$ 2.23   | 0.62 $\pm$ 0.07   | 0.81 $\pm$ 0.34   |

**Table S5.** Bacterial taxonomic profiles at the genus level in response to the peptide treatments in the HUA mouse model. Data are represented as the mean  $\pm$  SD (n=5).

|                                    | <b>control</b>   | <b>model</b>     | <b>GPAGRP</b>     | <b>GPSGRP</b>     |
|------------------------------------|------------------|------------------|-------------------|-------------------|
| <i>Muribaculaceae</i>              | 55.26 $\pm$ 7.77 | 39.92 $\pm$ 6.86 | 24.54 $\pm$ 5.42  | 38.59 $\pm$ 11.66 |
| <i>Bacteria</i> unclassified       | 6.02 $\pm$ 5.48  | 8.49 $\pm$ 8.19  | 30.72 $\pm$ 11.22 | 0.36 $\pm$ 0.6    |
| <i>Muribaculum</i>                 | 4.79 $\pm$ 5.27  | 3.48 $\pm$ 2.74  | 9.4 $\pm$ 6.17    | 21.49 $\pm$ 15.13 |
| <i>Muribaculaceae</i> unclassified | 4.02 $\pm$ 1.91  | 3.89 $\pm$ 3     | 11.22 $\pm$ 5     | 1.46 $\pm$ 1.06   |
| <i>Bacteroides</i>                 | 5.67 $\pm$ 4.51  | 4.41 $\pm$ 2.13  | 0.95 $\pm$ 0.69   | 4.5 $\pm$ 2.16    |
| <i>Prevotella</i>                  | 1.63 $\pm$ 1.08  | 6.2 $\pm$ 5.81   | 1.7 $\pm$ 1.78    | 5.86 $\pm$ 6.99   |
| <i>Candidatus Amulumruptor</i>     | 3.84 $\pm$ 1.04  | 3.13 $\pm$ 0.52  | 1.85 $\pm$ 0.62   | 2 $\pm$ 0.71      |
| <i>Akkermansia</i>                 | 1.92 $\pm$ 1.29  | 1.58 $\pm$ 0.82  | 2.55 $\pm$ 0.82   | 1.93 $\pm$ 1.09   |
| <i>Bacteroidales</i> unclassified  | 0.02 $\pm$ 0.02  | 4.28 $\pm$ 3.19  | 1.58 $\pm$ 1.24   | 0.61 $\pm$ 0.35   |
| <i>Bacteria</i> noname             | 2.09 $\pm$ 0.31  | 1.82 $\pm$ 1.11  | 0.71 $\pm$ 0.21   | 1.73 $\pm$ 1.16   |
| <i>Duncaniella</i>                 | 1.05 $\pm$ 0.71  | 1.2 $\pm$ 0.51   | 2.52 $\pm$ 0.46   | 1.39 $\pm$ 0.48   |
| <i>Lachnospiraceae</i> noname      | 0.45 $\pm$ 0.2   | 1.3 $\pm$ 1.31   | 0.88 $\pm$ 0.3    | 1.74 $\pm$ 1.15   |
| <i>Paramuribaculum</i>             | 1.35 $\pm$ 0.71  | 0.81 $\pm$ 0.33  | 0.62 $\pm$ 0.24   | 1.39 $\pm$ 0.43   |
| <i>Bacteroidales</i> noname        | 0.52 $\pm$ 0.29  | 1.92 $\pm$ 1.43  | 0.55 $\pm$ 0.33   | 1.11 $\pm$ 1.22   |
| <i>Alistipes</i>                   | 0.57 $\pm$ 0.6   | 2.57 $\pm$ 2.62  | 0.46 $\pm$ 0.29   | 0.29 $\pm$ 0.33   |
| <i>Clostridium</i>                 | 10.8 $\pm$ 3.32  | 14.99 $\pm$ 7.62 | 9.76 $\pm$ 2      | 15.54 $\pm$ 7.84  |
| Others                             | 55.26 $\pm$ 7.77 | 39.92 $\pm$ 6.86 | 24.54 $\pm$ 5.42  | 38.59 $\pm$ 11.66 |

**Table S6.** The forty-nine differentially abundant genera identified by the Kruskal-Wallis test in this study. Data are represented as the mean (n=5).

|                                      | <b>control</b> | <b>model</b> | <b>GPAGRP</b> | <b>GPSGRP</b> |
|--------------------------------------|----------------|--------------|---------------|---------------|
| <i>Paramuribaculum</i>               | 2.0946         | 1.822        | 0.7084        | 1.7322        |
| <i>Alloprevotella</i>                | 5.6651         | 4.41         | 0.9475        | 4.4967        |
| <i>Muribaculaceae unclassified</i>   | 55.2566        | 39.9233      | 24.5444       | 38.5892       |
| <i>Muribaculum</i>                   | 3.8424         | 3.135        | 1.8459        | 2.0016        |
| <i>Akkermansia</i>                   | 6.0168         | 8.4875       | 30.725        | 0.3646        |
| <i>Dubosiella</i>                    | 4.0192         | 3.8882       | 11.2168       | 1.4577        |
| <i>Candidatus Saccharimonas</i>      | 1.0456         | 1.1953       | 2.5185        | 1.3941        |
| <i>Lactobacillus</i>                 | 4.7899         | 3.4837       | 9.4006        | 21.4872       |
| <i>Alistipes</i>                     | 1.3536         | 0.8071       | 0.6154        | 1.3926        |
| <i>Desulfovibrio</i>                 | 0.352          | 1.3478       | 0.2343        | 1.5619        |
| <i>Coriobacteriaceae UCG-002</i>     | 0.0209         | 4.2828       | 1.5841        | 0.6119        |
| <i>Bifidobacterium</i>               | 0.5709         | 2.5672       | 0.46          | 0.2898        |
| <i>Bacteroidetes unclassified</i>    | 0.0133         | 0.0969       | 0             | 0.0406        |
| <i>Anaeroplasma</i>                  | 0.0095         | 0.0031       | 0             | 0.0183        |
| <i>Odoribacter</i>                   | 0.2678         | 0.3747       | 0.0877        | 0.375         |
| <i>Enterococcus</i>                  | 0.0988         | 0.0188       | 0.0121        | 0             |
| <i>Insolitispirillum</i>             | 0.0226         | 0.0022       | 0.0029        | 0.0029        |
| <i>Prevotellaceae UCG-001</i>        | 0.5083         | 0.3918       | 0.1809        | 0.219         |
| <i>Escherichia-Shigella</i>          | 0.0647         | 0.0903       | 0.0279        | 0.0314        |
| <i>Methylobacterium</i>              | 0.0067         | 0            | 0             | 0.002         |
| <i>Parabacteroides</i>               | 0.321          | 0.168        | 0.1063        | 0.1217        |
| <i>Turicibacter</i>                  | 0.449          | 0.0447       | 0.0268        | 0             |
| <i>Bacteroides</i>                   | 0.7858         | 0.6714       | 0.4452        | 0.4586        |
| <i>Acidaminococcus</i>               | 0.0871         | 0.0554       | 0.0229        | 0.0302        |
| <i>Fusobacterium</i>                 | 0.0405         | 0.0269       | 0.009         | 0.0101        |
| <i>Eggerthella</i>                   | 0.0368         | 0.0105       | 0.0543        | 0.018         |
| <i>Dorea</i>                         | 0.0437         | 0            | 0.0054        | 0.0367        |
| <i>Rikenellaceae RC9 gut group</i>   | 0.4958         | 0.3836       | 0.1459        | 0.7621        |
| <i>Ruminococcaceae UCG-004</i>       | 0.0281         | 0.0183       | 0.1339        | 0.8643        |
| <i>Eubacterium xylanophilum</i>      | 0.349          | 0.1362       | 0.2233        | 1.1964        |
| <i>group</i>                         |                |              |               |               |
| <i>Ruminococcaceae unclassified</i>  | 0.0104         | 0.0572       | 0.0249        | 0.0679        |
| <i>Stenotrophomonas</i>              | 0.0055         | 0.0065       | 0.0045        | 0.04          |
| <i>Candidatus Bacilloplasma</i>      | 0              | 0            | 0.0093        | 0.0267        |
| <i>Eisenbergiella</i>                | 0              | 0.0018       | 0.0008        | 0.0239        |
| <i>Oligella</i>                      | 0              | 0            | 0             | 0.0322        |
| <i>Pygmaibacter</i>                  | 0              | 0            | 0             | 0.0423        |
| <i>Oxyphotobacteria unclassified</i> | 0              | 0            | 0             | 0.0069        |
| <i>Eubacterium coprostanoligenes</i> | 0.0937         | 0.1092       | 1.325         | 0.1567        |
| <i>group</i>                         |                |              |               |               |

|   |        |        |        |        |
|---|--------|--------|--------|--------|
| <i>Blautia</i>                          | 0      | 0.0017 | 0.0253 | 0.0057 |
| <i>Candidatus_Stoquefichus</i>          | 0      | 0.0076 | 0.0172 | 0      |
| <i>Prevotellaceae NK3B31 group</i>      | 0      | 0.0092 | 0.0516 | 0      |
| <i>Enterorhabdus</i>                    | 0.6853 | 0.3503 | 0.5928 | 0.4296 |
| <i>Christensenellaceae_R-7_group</i>    | 0.0949 | 0.0376 | 0.0808 | 0.0351 |
| <i>Olsenella</i>                        | 0      | 0.1549 | 0.0241 | 0.0039 |
| <i>Parasutterella</i>                   | 0.3578 | 0.7386 | 0.5704 | 0.1745 |
| <i>Erysipelotrichaceae_unclassified</i> | 0.0379 | 0.104  | 0.0678 | 0.0301 |
| <i>Eubacterium_nodatum_group</i>        | 0.0268 | 0.0156 | 0.0384 | 0.0038 |
| <i>Erysipelatoclostridium</i>           | 0.0067 | 0.0326 | 0.0525 | 0      |
| <i>Catabacter</i>                       | 0.0044 | 0.0262 | 0.0406 | 0.0035 |

**Table S7.** Overview of reads from raw data to cleaned sequences. Data are represented as the mean  $\pm$  SD (n=3).

|             | <b>control</b>             | <b>model</b>               | <b>GPSGRP</b>              |
|-------------|----------------------------|----------------------------|----------------------------|
| Raw reads   | 14,949,618 $\pm$ 5,098,975 | 11,585,807 $\pm$ 1,121,781 | 16,114,506 $\pm$ 3,922,372 |
| Clean reads | 3,718,484 $\pm$ 1,271,019  | 2,369,893 $\pm$ 174,885    | 2,800,077 $\pm$ 689,950    |



**Table S8.** Twenty-one nonrepetitive and DE-miRNAs were identified among the three groups in this study.

| miR              | Sequence                | P-value | Control | Model    | GPSGRP  | Expression level |
|------------------|-------------------------|---------|---------|----------|---------|------------------|
| mmu-miR-188-5p   | CATCCCTTGCATGGTGGAGGGT  | 0.0245  | 14.21   | 4.31     | 3.10    | middle           |
| mmu-miR-598-3p   | TACGTCATCGTCGTCATCGTTAT | 0.0437  | 22.59   | 27.83    | 8.03    | middle           |
| mmu-miR-411-3p   | TATGTAACACGGTCCACTAAC   | 0.0080  | 1.27    | 0.1      | 7.48    | middle           |
| mmu-miR-3084-3p  | CTGCCAGTCTCCTTCAGACAAAA | 0.0082  | 4.97    | 0.1      | 1.33    | low              |
| mmu-miR-20a-3p   | ACTGCATTACGAGCACTTAAAGT | 0.0094  | 9.06    | 0.1      | 2.09    | middle           |
| mmu-miR-152-5p   | TAGGTTCTGTGATACTCCGACT  | 0.0233  | 11.65   | 0.1      | 5.43    | middle           |
| mmu-miR-15a-3p   | CAGGCCATACTGTGCTGCCTCA  | 0.0261  | 8.68    | 0.1      | 2.03    | middle           |
| tch-miR-26a-2-5p | TTCAAGTAACCCAGGATAGGCT  | 0.0436  | 8.08    | 0.1      | 2.68    | middle           |
| mmu-miR-125b-5p  | TCCCTGAGACCCTAACTTGTGA  | 0.0144  | 8776.15 | 10531.86 | 7902.99 | high             |
| hsa-miR-378c     | ACTGGACTTGGAGTCAGAAGGCT | 0.0087  | 2153.94 | 1794.13  | 1537.99 | middle           |
| mmu-miR-200c-3p  | TAATACTGCCGGGTAATGATGGA | 0.0417  | 3717.20 | 3637.58  | 2552.11 | high             |
| hsa-mir-636-p5   | CGCGCGGGCGGGGCCGGG      | 0.0451  | 56.03   | 44.99    | 125.61  | middle           |
| cgr-miR-1973     | TATCCTGACCGTGCAAAGGTAGC | 0.0487  | 98.66   | 63.38    | 135.54  | middle           |
|                  | A                       |         |         |          |         |                  |
| cgr-miR-1260_2   | ATCCCACCGCTGCCACCAT     | 0.0179  | 38.70   | 55.67    | 74.48   | middle           |
| cgr-miR-1260_1   | ATCCCACCGCTGCCACCAA     | 0.0179  | 38.70   | 55.67    | 74.48   | middle           |
| mmu-mir-3535-p3  | GATAGTCAAGTTCTGATC      | 0.0325  | 67.79   | 38.06    | 42.04   | middle           |
| mmu-miR-501-5p   | AATCCTTTGTCCCTGGGTGA    | 0.0339  | 65.95   | 43.24    | 62.55   | middle           |
| mmu-miR-501-3p   | AATGCACCCGGGCAAGGATTTGG | 0.0303  | 600.60  | 713.68   | 493.76  | middle           |
| bta-miR-378      | ACTGGACTTGGAGTCAGAAGGAT | 0.0045  | 260.23  | 179.46   | 148.79  | middle           |
| mmu-miR-128-3p   | TCACAGTGAACCGGTCTCTTT   | 0.0087  | 262.80  | 276.45   | 350.38  | middle           |
| mmu-miR-345-5p   | TGCTGACCCCTAGTCCAGTGCT  | 0.0464  | 412.32  | 342.57   | 365.76  | middle           |

**Table S9.** miRNAs related to uric acid metabolism and immune response identified among three groups in this study.

| <b>miR</b>  | <b>Reported target mRNA</b>     | <b>Control</b> | <b>Model</b> | <b>GPSGRP</b> | <b>Expression level</b> |
|-------------|---------------------------------|----------------|--------------|---------------|-------------------------|
| miR-143-3p  | GLUT9                           | 27346.75       | 25895.85     | 28048.07      | High                    |
| miR-34a     | URAT1                           | 570.82         | 680.99       | 507.83        | Middle                  |
| miR-223     | NLRP3 inflammasome              | 168.33         | 204.76       | 626.83        | Middle                  |
| miR-17-5p   | Thioredoxin-interacting protein | 808.94         | 730.23       | 918.29        | Middle                  |
| miR-186     | TLR4/ NF- $\kappa$ B pathway    | 513.34         | 521.51       | 695.31        | Middle                  |
| miR-125b-5p | related to renal injury         | 8776.15        | 10531.86     | 7902.99       | High                    |
| miR-152-5p  | related to renal injury         | 11.65          | 0.1          | 5.43          | Middle                  |
| miR-188-5p  | PTEN/PI3K/AKT                   | 14.21          | 4.31         | 3.10          | Middle                  |
| miR-128-3p  | SIRT1                           | 262.80         | 276.45       | 350.38        | Middle                  |
| miR-30b     | IL-6 receptor                   | 13363.08       | 10188.33     | 9902.47       | High                    |

**Table S10.** Spearman’s correlation analysis between these 10 miRNAs and 49 differentially abundant genera.

|                                       | miR-143-3p | miR-223 | miR-17-5p | miR-186 | miR-125b-5p | miR-152-5p | miR-188-5p | miR-128-3p | miR-30b | miR-34a |
|---------------------------------------|------------|---------|-----------|---------|-------------|------------|------------|------------|---------|---------|
| <i>Paramuribaculum</i>                | -0.042     | -0.742  | -0.328    | -0.72   | 0.049       | 0.752      | 0.99       | -0.789     | 0.986   | 0.084   |
| <i>Alloprevotella</i>                 | 0.257      | -0.508  | -0.032    | -0.481  | -0.251      | 0.914      | 0.987      | -0.57      | 0.991   | -0.216  |
| <i>Muribaculaceae_unclassified</i>    | 0.126      | -0.619  | -0.165    | -0.594  | -0.119      | 0.852      | 1          | -0.675     | 1       | -0.084  |
| <i>Muribaculum</i>                    | -0.442     | -0.949  | -0.682    | -0.939  | 0.448       | 0.422      | 0.849      | -0.97      | 0.835   | 0.48    |
| <i>Akkermansia</i>                    | -0.913     | -0.931  | -0.992    | -0.942  | 0.916       | -0.254     | 0.316      | -0.902     | 0.293   | 0.929   |
| <i>Dubosiella</i>                     | -0.72      | -1      | -0.889    | -1      | 0.725       | 0.09       | 0.62       | -0.995     | 0.6     | 0.748   |
| <i>Candidatus_Saccharimonas</i>       | 0.395      | 0.932   | 0.643     | 0.92    | -0.402      | -0.468     | -0.875     | 0.956      | -0.863  | -0.434  |
| <i>Lactobacillus</i>                  | 0.792      | 0.991   | 0.934     | 0.995   | -0.796      | 0.021      | -0.529     | 0.978      | -0.508  | -0.817  |
| <i>Alistipes</i>                      | 0.965      | 0.49    | 0.848     | 0.517   | -0.963      | 0.809      | 0.357      | 0.424      | 0.38    | -0.953  |
| <i>Desulfovibrio</i>                  | -0.032     | 0.69    | 0.257     | 0.667   | 0.025       | -0.799     | -0.998     | 0.742      | -0.996  | -0.011  |
| <i>Coriobacteriaceae_UCG-002</i>      | -0.899     | -0.318  | -0.735    | -0.348  | 0.896       | -0.905     | -0.525     | -0.247     | -0.546  | 0.88    |
| <i>Bifidobacterium</i>                | -0.978     | -0.536  | -0.876    | -0.562  | 0.976       | -0.777     | -0.306     | -0.472     | -0.329  | 0.968   |
| <i>Bacteroidetes_unclassified</i>     | -0.795     | -0.126  | -0.587    | -0.157  | 0.791       | -0.971     | -0.682     | -0.052     | -0.7    | 0.769   |
| <i>Anaeroplasma</i>                   | 0.958      | 0.876   | 1         | 0.89    | -0.96       | 0.379      | -0.189     | 0.838      | -0.165  | -0.97   |
| <i>Odoribacter</i>                    | -0.195     | 0.563   | 0.096     | 0.536   | 0.188       | -0.886     | -0.995     | 0.622      | -0.997  | 0.153   |
| <i>Enterococcus</i>                   | 0.018      | -0.7    | -0.271    | -0.677  | -0.011      | 0.79       | 0.997      | -0.751     | 0.994   | 0.024   |
| <i>Insolitispirillum</i>              | 0.227      | -0.535  | -0.064    | -0.508  | -0.22       | 0.901      | 0.992      | -0.596     | 0.995   | -0.185  |
| <i>Prevotellaceae_UCG-001</i>         | -0.423     | -0.943  | -0.666    | -0.932  | 0.43        | 0.441      | 0.86       | -0.965     | 0.847   | 0.461   |
| <i>Escherichia-Shigella</i>           | -0.963     | -0.868  | -1        | -0.883  | 0.965       | -0.393     | 0.174      | -0.829     | 0.149   | 0.973   |
| <i>Methylobacterium</i>               | 0.474      | -0.296  | 0.2       | -0.265  | -0.467      | 0.983      | 0.923      | -0.365     | 0.933   | -0.436  |
| <i>Parabacteroides</i>                | -0.025     | -0.73   | -0.312    | -0.708  | 0.032       | 0.763      | 0.992      | -0.779     | 0.989   | 0.068   |
| <i>Turicibacter</i>                   | 0.108      | -0.633  | -0.183    | -0.608  | -0.101      | 0.842      | 1          | -0.689     | 1       | -0.065  |
| <i>Bacteroides</i>                    | -0.477     | -0.961  | -0.71     | -0.952  | 0.483       | 0.386      | 0.828      | -0.979     | 0.813   | 0.514   |
| <i>Acidaminococcus</i>                | -0.256     | -0.869  | -0.524    | -0.853  | 0.263       | 0.592      | 0.936      | -0.903     | 0.927   | 0.297   |
| <i>Fusobacterium</i>                  | -0.376     | -0.924  | -0.627    | -0.912  | 0.383       | 0.486      | 0.885      | -0.95      | 0.873   | 0.415   |
| <i>Eggerthella</i>                    | 0.461      | -0.31   | 0.186     | -0.279  | -0.455      | 0.98       | 0.929      | -0.379     | 0.938   | -0.423  |
| <i>Dorea</i>                          | 0.889      | 0.298   | 0.72      | 0.328   | -0.886      | 0.914      | 0.543      | 0.227      | 0.564   | -0.869  |
| <i>Rikenellaceae_RC9</i>              | 0.909      | 0.934   | 0.991     | 0.945   | -0.912      | 0.246      | -0.324     | 0.906      | -0.301  | -0.926  |
| <i>Ruminococcaceae_UCG-004</i>        | 0.757      | 0.997   | 0.913     | 0.999   | -0.762      | -0.034     | -0.575     | 0.988      | -0.555  | -0.784  |
| <i>Eubacterium_xylanophilum_group</i> | 0.862      | 0.966   | 0.972     | 0.974   | -0.866      | 0.146      | -0.419     | 0.944      | -0.396  | -0.883  |
| <i>Ruminococcaceae_unclassified</i>   | -0.023     | 0.697   | 0.266     | 0.674   | 0.016       | -0.793     | -0.997     | 0.748      | -0.995  | -0.02   |
| <i>Stenotrophomonas</i>               | 0.733      | 0.999   | 0.898     | 1       | -0.738      | -0.07      | -0.604     | 0.993      | -0.584  | -0.762  |
| <i>Candidatus_Bacilloplasma</i>       | 0.75       | 0.997   | 0.909     | 0.999   | -0.755      | -0.044     | -0.583     | 0.989      | -0.563  | -0.778  |
| <i>Eisenbergiella</i>                 | 0.704      | 1       | 0.879     | 1       | -0.709      | -0.112     | -0.637     | 0.997      | -0.618  | -0.734  |

|  |        |        |        |        |        |        |        |        |        |        |
|--|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|
| <i>Oligella</i>                            | 0.75   | 0.997  | 0.909  | 0.999  | -0.755 | -0.044 | -0.583 | 0.989  | -0.563 | -0.778 |
| <i>Pygmaibacter</i>                        | 0.75   | 0.997  | 0.909  | 0.999  | -0.755 | -0.044 | -0.583 | 0.989  | -0.563 | -0.778 |
| <i>Oxyphotobacteria_unclassified</i>       | 0.75   | 0.997  | 0.909  | 0.999  | -0.755 | -0.044 | -0.583 | 0.989  | -0.563 | -0.778 |
| <i>Eubacterium_coprostanoligenes_group</i> | 0.573  | 0.986  | 0.785  | 0.98   | -0.579 | -0.279 | -0.759 | 0.996  | -0.742 | -0.607 |
| <i>Blautia</i>                             | 0.526  | 0.975  | 0.749  | 0.968  | -0.532 | -0.333 | -0.794 | 0.989  | -0.779 | -0.562 |
| <i>Candidatus_Stoquefichus</i>             | -0.948 | -0.437 | -0.815 | -0.465 | 0.945  | -0.843 | -0.412 | -0.369 | -0.434 | 0.933  |
| <i>Prevotellaceae_NK3B31_group</i>         | -0.948 | -0.437 | -0.815 | -0.465 | 0.945  | -0.843 | -0.412 | -0.369 | -0.434 | 0.933  |
| <i>Enterorhabdus</i>                       | 0.414  | -0.359 | 0.134  | -0.329 | -0.408 | 0.969  | 0.947  | -0.426 | 0.954  | -0.375 |
| <i>Christensenellaceae_R-7_group</i>       | 0.161  | -0.591 | -0.13  | -0.565 | -0.154 | 0.87   | 0.998  | -0.649 | 0.999  | -0.119 |
| <i>Olsenella</i>                           | -0.94  | -0.417 | -0.802 | -0.446 | 0.938  | -0.855 | -0.432 | -0.349 | -0.454 | 0.925  |
| <i>Parasutterella</i>                      | -1     | -0.701 | -0.957 | -0.723 | 1      | -0.628 | -0.1   | -0.646 | -0.125 | 0.999  |
| <i>Erysipelotrichaceae_unclassified</i>    | -0.974 | -0.521 | -0.867 | -0.548 | 0.972  | -0.787 | -0.322 | -0.457 | -0.346 | 0.963  |
| <i>Eubacterium_nodatum_group</i>           | -0.334 | -0.906 | -0.591 | -0.892 | 0.34   | 0.525  | 0.905  | -0.935 | 0.894  | 0.373  |
| <i>Erysipelatoclostridium</i>              | -0.992 | -0.604 | -0.912 | -0.629 | 0.991  | -0.722 | -0.226 | -0.543 | -0.251 | 0.985  |
| <i>Catabacter</i>                          | -0.958 | -0.468 | -0.835 | -0.496 | 0.956  | -0.824 | -0.379 | -0.402 | -0.402 | 0.945  |

## SUPPORTING FIGURE LEGENDS

**Fig. S1 *In vitro* XOD inhibitory rate of allopurinol.** Data are shown as mean  $\pm$  SD, n=3.

**Fig. S2 The effects of GPAGPR and GPSGRP on body weight.** **A.** body weight curve; **B.** Body weight gain. Data are shown as mean  $\pm$  SD, n=5. \*\*  $p < 0.01$ .

**Fig. S3 Effects of peptides on the mRNA levels of the NF- $\kappa$ B signaling pathway.** **A.** TLR4. **B.** MyD88. **C.** TRAF6. **D.** iNOS. **E.** COX-2. Data are shown as mean  $\pm$  SD, n=5. \*  $p < 0.05$ , \*\*  $p < 0.01$ , and \*\*\*  $p < 0.001$ .

**Fig. S4 DE-miRNAs (nonrepetitive) in the pairwise comparison among the three treatments ( $p < 0.05$ ).** The abundance of DE-miRNAs increased in the model group and reduced by GPSGRP treatment is marked in red, and the abundance of DE-miRNAs decreased in the model group and enhanced by the GPSGRP treatment is marked in green.

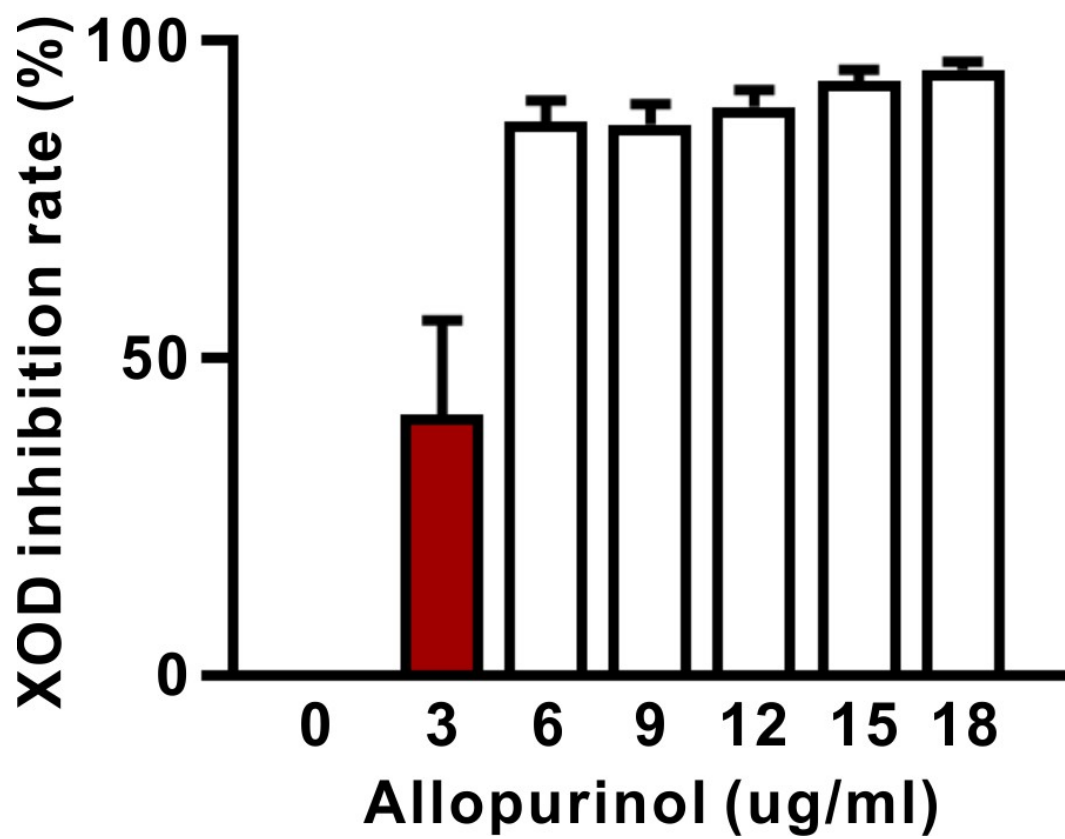


Fig. S1

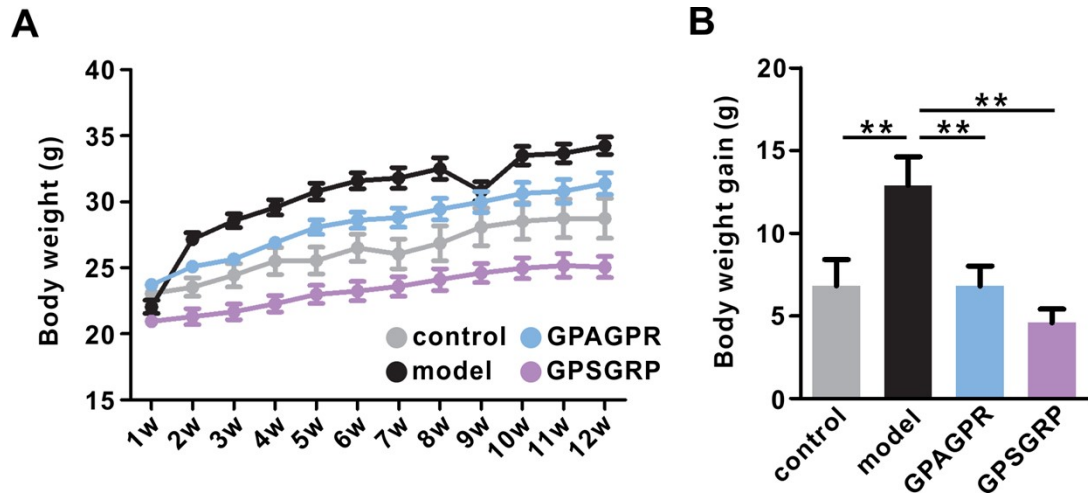


Fig. S2

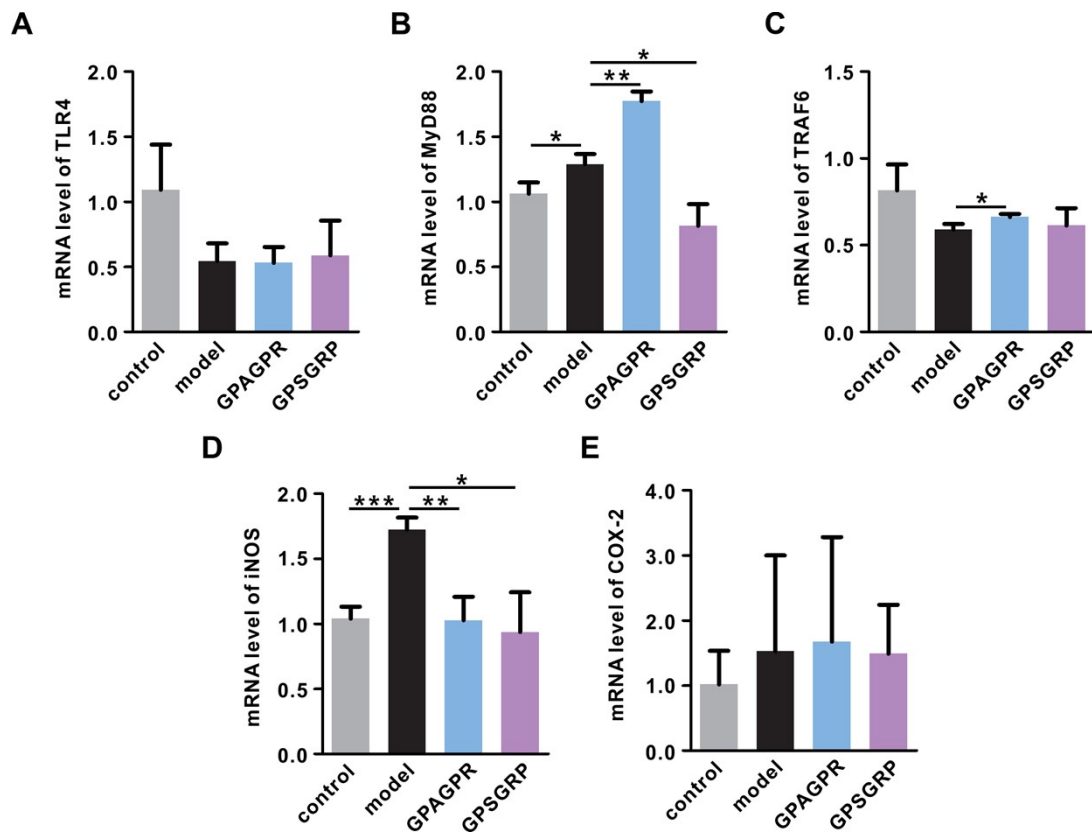


Fig. S3

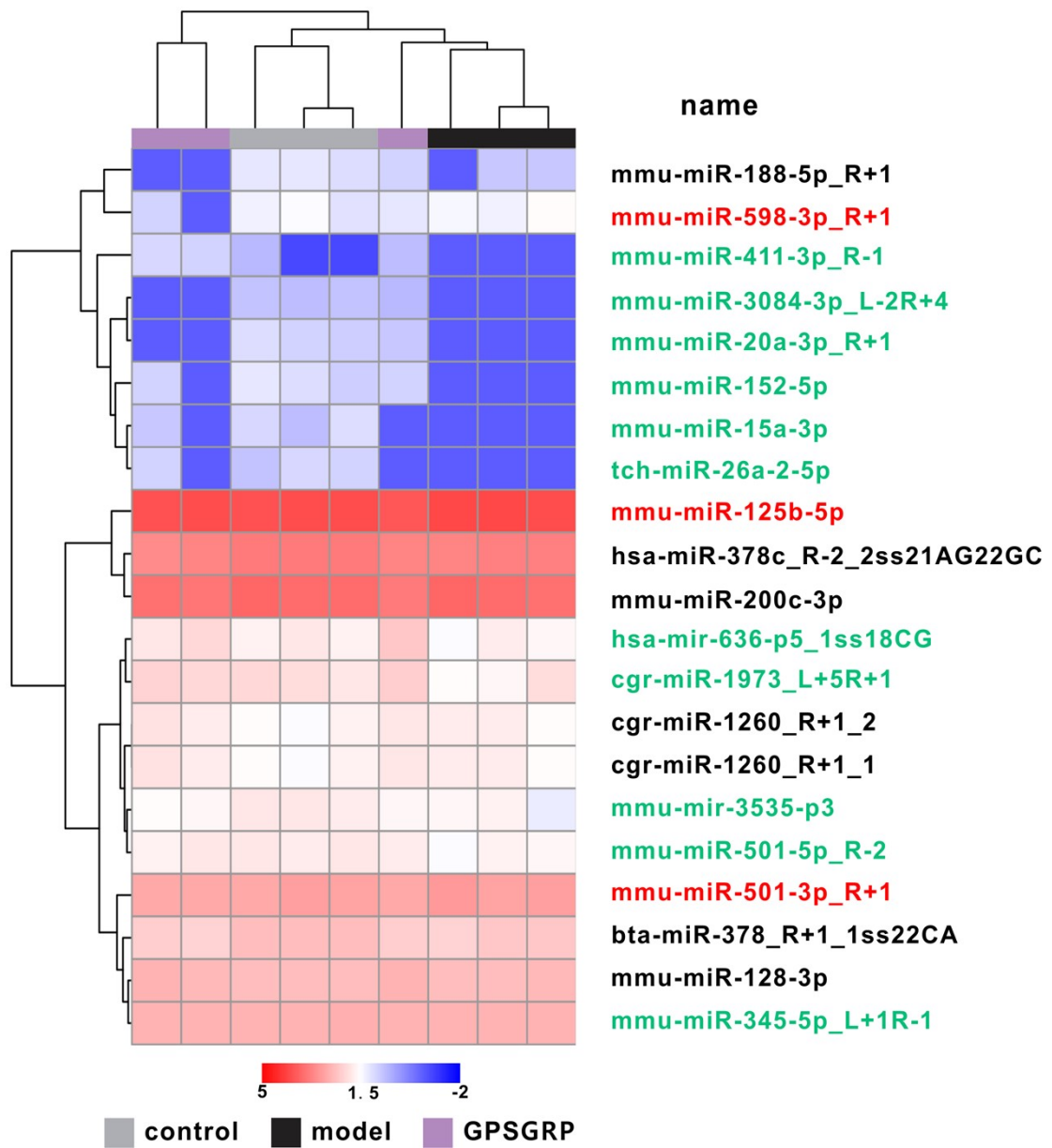


Fig. S4