**Quality** **uniformity evaluation for Babao Dan capsules based on fourteen main chemical components and *in vitro* anti-inflammatory activity**

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Table A.1 Results of precision of 14 index compounds

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Analytes | Precision (*n*=6) | | | | | | RSD (%) |
| The ratio of the peak area of the analyte to ISTD | | | | | |
| Cholic acid | 19.82 | 19.73 | 19.34 | 19.33 | 19.86 | 19.68 | 3.0 |
| Glycodeoxycholic acid | 11.71 | 11.48 | 11.55 | 10.87 | 11.75 | 11.45 | 2.8 |
| Ursodeoxycholic acid | 3.01 | 3.08 | 2.96 | 2.82 | 3.15 | 2.93 | 3.9 |
| Chenodeoxycholic acid | 0.67 | 0.66 | 0.63 | 0.60 | 0.68 | 0.66 | 4.1 |
| Deoxycholic acid | 3.60 | 3.63 | 3.49 | 3.41 | 3.70 | 3.61 | 2.9 |
| Glycocholic acid | 39.04 | 39.86 | 37.81 | 36.83 | 38.97 | 38.79 | 2.8 |
| Taurocholic acid | 0.48 | 0.49 | 0.47 | 0.46 | 0.50 | 0.46 | 2.7 |
| Bilirubin | 226.95 | 221.53 | 213.22 | 204.60 | 218.19 | 209.86 | 3.8 |
| Ginsenoside Rb1 | 14.27 | 14.42 | 13.87 | 13.63 | 14.71 | 14.59 | 3.0 |
| Ginsenoside Re | 1.00 | 1.05 | 1.01 | 0.99 | 1.05 | 1.02 | 2.6 |
| Ginsenoside Rd | 15.22 | 15.56 | 15.11 | 14.62 | 15.84 | 15.60 | 2.8 |
| Ginsenoside Rg1 | 7.38 | 7.36 | 7.26 | 6.99 | 7.53 | 7.41 | 2.5 |
| Notoginsenoside R1 | 9.62 | 9.73 | 9.64 | 9.18 | 10.08 | 9.96 | 3.2 |
| Muskone | 0.07 | 0.07 | 0.06 | 0.07 | 0.07 | 0.07 | 2.0 |

Table A.2 Results of stability of 14 index compounds

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Analytes | The ratio of the peak area of the analyte to ISTD | | | | | RSD (%) |
| 0h | 2h | 4h | 10h | 12h |
| Cholic acid | 13.88 | 12.61 | 12.73 | 12.95 | 12.99 | 3.8 |
| Glycodeoxycholic acid | 6.27 | 5.95 | 6.28 | 6.06 | 6.01 | 2.5 |
| Ursodeoxycholic acid | 1.51 | 1.49 | 1.48 | 1.52 | 1.55 | 1.9 |
| Chenodeoxycholic acid | 0.66 | 0.69 | 0.71 | 0.65 | 0.63 | 4.8 |
| Deoxycholic acid | 3.15 | 3.24 | 3.05 | 2.92 | 2.95 | 4.4 |
| Glycocholic acid | 18.44 | 17.61 | 17.83 | 16.24 | 17.08 | 4.8 |
| Taurocholic acid | 0.74 | 0.66 | 0.67 | 0.70 | 0.69 | 4.5 |
| Bilirubin | 112.55 | 118.56 | 121.58 | 123.98 | 118.71 | 3.6 |
| Ginsenoside Rb1 | 20.19 | 19.73 | 20.68 | 21.01 | 20.71 | 2.5 |
| Ginsenoside Re | 0.41 | 0.41 | 0.42 | 0.45 | 0.42 | 4.3 |
| Ginsenoside Rd | 6.26 | 5.86 | 6.11 | 6.56 | 6.31 | 4.2 |
| Ginsenoside Rg1 | 10.79 | 11.41 | 11.41 | 11.94 | 12.12 | 4.6 |
| Notoginsenoside R1 | 2.14 | 2.28 | 2.35 | 2.40 | 2.35 | 4.4 |
| Muskone | 0.26 | 0.26 | 0.27 | 0.28 | 0.28 | 3.9 |

Table A.3 Results of repeatability of 14 index compounds

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Analytes | Content (mg/g) | | | | | | RSD (%) |
| 1 | 2 | 3 | 4 | 5 | 6 |
| Cholic acid | 19.51 | 19.21 | 19.32 | 17.24 | 19.20 | 17.60 | 5.3 |
| Glycodeoxycholic acid | 1.92 | 1.88 | 1.89 | 1.89 | 1.89 | 1.90 | 0.7 |
| Ursodeoxycholic acid | 3.52 | 3.39 | 3.50 | 3.38 | 3.53 | 3.43 | 1.9 |
| Chenodeoxycholic acid | 4.62 | 4.41 | 4.60 | 4.51 | 4.53 | 4.46 | 1.8 |
| Deoxycholic acid | 9.18 | 8.74 | 8.98 | 8.84 | 8.61 | 8.60 | 2.6 |
| Glycocholic acid | 9.01 | 8.49 | 8.76 | 8.42 | 7.43 | 8.08 | 3.7 |
| Taurocholic acid | 1.80 | 1.79 | 1.84 | 1.84 | 1.74 | 1.83 | 2.1 |
| Bilirubin | 5.27 | 5.37 | 5.25 | 4.70 | 5.28 | 5.10 | 4.7 |
| Ginsenoside Rb1 | 59.61 | 56.86 | 58.87 | 52.93 | 60.07 | 53.73 | 5.4 |
| Ginsenoside Re | 3.41 | 3.25 | 3.41 | 3.15 | 3.29 | 3.25 | 3.1 |
| Ginsenoside Rd | 4.55 | 4.33 | 4.54 | 4.52 | 4.78 | 4.47 | 4.1 |
| Ginsenoside Rg1 | 66.35 | 61.97 | 63.02 | 61.26 | 64.90 | 59.35 | 4.0 |
| Notoginsenoside R1 | 4.56 | 4.11 | 4.32 | 4.43 | 4.49 | 4.18 | 4.1 |
| Muskone | 1.12 | 1.25 | 1.20 | 1.25 | 1.16 | 1.15 | 4.5 |

Table A.4 Recovery results of 14 index compounds

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Analytes | Spiked amount (μg) | Original amount (μg) | Detected amount (μg) | Recovery (%) | Mean recovery (%) | RSD (%) |
| Cholic acid | 474.08 | 494.90 | 991.61 | 104.57 | 103.9 | 2.1 |
| 473.71 | 494.90 | 980.13 | 102.33 |
| 467.06 | 494.90 | 992.40 | 106.15 |
| 468.35 | 494.90 | 969.00 | 101.16 |
| 469.83 | 494.90 | 978.51 | 102.78 |
| 473.16 | 494.90 | 1000.52 | 106.56 |
| Glycodeoxycholic acid | 49.18 | 50.00 | 96.61 | 94.85 | 96.9 | 1.9 |
| 49.14 | 50.00 | 97.92 | 97.56 |
| 49.00 | 50.00 | 98.04 | 98.07 |
| 49.35 | 50.00 | 96.51 | 94.31 |
| 49.43 | 50.00 | 98.42 | 97.97 |
| 48.71 | 50.00 | 98.02 | 98.62 |
| Ursodeoxycholic acid | 90.50 | 63.80 | 153.32 | 98.45 | 99.5 | 2.3 |
| 90.43 | 63.80 | 152.00 | 96.50 |
| 89.16 | 63.80 | 154.61 | 102.59 |
| 89.41 | 63.80 | 154.46 | 101.96 |
| 89.69 | 63.80 | 152.71 | 98.78 |
| 90.33 | 63.80 | 153.44 | 98.93 |
| Deoxycholic acid | 100.88 | 137.60 | 239.44 | 100.70 | 99.8 | 2.1 |
| 100.80 | 137.60 | 233.05 | 96.11 |
| 100.52 | 137.60 | 241.58 | 102.52 |
| 101.24 | 137.60 | 238.76 | 99.94 |
| 101.40 | 137.60 | 238.12 | 99.36 |
| 99.92 | 137.60 | 237.66 | 100.10 |
| Glycocholic acid | 216.66 | 241.40 | 444.90 | 94.55 | 96.0 | 2.8 |
| 216.49 | 241.40 | 443.60 | 94.08 |
| 213.45 | 241.40 | 453.08 | 99.27 |
| 214.04 | 241.40 | 443.65 | 95.12 |
| 214.71 | 241.40 | 454.96 | 99.52 |
| 216.24 | 241.40 | 441.94 | 93.50 |
| Taurocholic acid | 187.38 | 228.00 | 407.13 | 96.38 | 93.1 | 4.5 |
| 187.24 | 228.00 | 409.12 | 97.32 |
| 186.72 | 228.00 | 388.52 | 88.51 |
| 188.05 | 228.00 | 387.45 | 87.45 |
| 188.35 | 228.00 | 405.53 | 95.25 |
| 185.60 | 228.00 | 398.86 | 93.54 |
| Chenodeoxycholic acid | 46.41 | 43.41 | 86.85 | 93.16 | 95.2 | 4.7 |
| 46.37 | 43.41 | 86.24 | 91.83 |
| 45.72 | 43.41 | 87.58 | 96.42 |
| 45.85 | 43.41 | 90.27 | 102.32 |
| 45.99 | 43.41 | 88.21 | 97.24 |
| 46.32 | 43.41 | 85.41 | 90.04 |
| Bilirubin | 91.98 | 140.80 | 234.56 | 101.26 | 97.7 | 3.0 |
| 92.60 | 140.80 | 224.06 | 93.36 |
| 91.98 | 140.80 | 232.13 | 99.54 |
| 91.62 | 140.80 | 227.6 | 96.59 |
| 92.27 | 140.80 | 232.31 | 99.46 |
| 91.51 | 140.80 | 226.83 | 96.11 |
| Ginsenoside Rb1 | 1079.44 | 740.80 | 1800.74 | 97.37 | 103.0 | 3.6 |
| 1078.60 | 740.80 | 1839.13 | 102.66 |
| 1063.45 | 740.80 | 1870.38 | 108.93 |
| 1066.39 | 740.80 | 1824.58 | 102.35 |
| 1069.76 | 740.80 | 1835.29 | 103.34 |
| 1077.34 | 740.80 | 1843.79 | 103.46 |
| Ginsenoside Re | 72.56 | 60.37 | 130.83 | 96.52 | 100.3 | 2.2 |
| 72.50 | 60.37 | 132.21 | 98.90 |
| 71.49 | 60.37 | 132.83 | 101.62 |
| 71.68 | 60.37 | 132.77 | 101.18 |
| 71.91 | 60.37 | 133.20 | 101.52 |
| 72.42 | 60.37 | 134.19 | 102.32 |
| Ginsenoside Rd | 131.14 | 137.60 | 282.53 | 110.02 | 108.8 | 1.8 |
| 131.04 | 137.60 | 283.12 | 110.53 |
| 130.68 | 137.60 | 280.89 | 109.16 |
| 131.61 | 137.60 | 279.83 | 107.72 |
| 131.82 | 137.60 | 276.89 | 105.43 |
| 129.90 | 137.60 | 281.01 | 109.82 |
| Ginsenoside Rg1 | 1352.25 | 763.47 | 2069.16 | 93.90 | 98.8 | 2.6 |
| 1351.20 | 763.47 | 2104.61 | 98.68 |
| 1332.21 | 763.47 | 2102.63 | 100.91 |
| 1335.90 | 763.47 | 2090.01 | 98.77 |
| 1340.12 | 763.47 | 2100.98 | 99.66 |
| 1349.62 | 763.47 | 2119.42 | 100.83 |
| Notoginsenoside R1 | 112.72 | 146.46 | 253.19 | 95.91 | 101.6 | 3.9 |
| 112.54 | 146.46 | 260.51 | 101.03 |
| 112.23 | 146.46 | 266.7 | 105.45 |
| 111.78 | 146.46 | 256.2 | 98.63 |
| 113.16 | 146.46 | 263.04 | 102.33 |
| 112.01 | 146.46 | 267.67 | 106.28 |
| Muskone | 26.67 | 16.21 | 43.23 | 102.16 | 101.7 | 3.7 |
| 26.64 | 16.21 | 43.79 | 105.79 |
| 26.27 | 16.21 | 43.05 | 103.50 |
| 26.34 | 16.21 | 43.26 | 104.36 |
| 26.43 | 16.21 | 41.99 | 96.00 |
| 26.61 | 16.21 | 42.52 | 98.13 |

Table A.5 The concentrations of NO based on the RAW264.7 and the inhibited counts by luciferase on NF-ĸB-RE-luc HEK293 from different groups *in vitro* (n=6).

|  |  |  |
| --- | --- | --- |
| Samples | NO (μM） | Counts |
| C | 1.29 | 174 |
| M | 35.32 | 120457 |
| S1-1 | 23.44 | 45756 |
| S1-2 | 23.25 | 41876 |
| S1-3 | 24.16 | 40881 |
| S1-4 | 23.10 | 37170 |
| S1-5 | 22.14 | 47361 |
| S1 | 23.44 | 45206 |
| S2 | 21.68 | 50498 |
| S3 | 20.12 | 48305 |
| S4 | 21.57 | 45714 |
| S5 | 22.08 | 45566 |
| S6 | 19.76 | 52003 |
| S7 | 29.20 | 51636 |
| S8 | 21.11 | 45185 |
| S9 | 20.99 | 48774 |
| S10 | 18.33 | 52503 |

Table A.6 The coefficients of association (*r*) between inhibition of NO, luciferase counts and the content of 14 main compounds, respectively.

|  |  |  |
| --- | --- | --- |
| Sample | *r*-NO | *r*-Counts |
| Cholic acid | 0.6098 | 0.2779 |
| Glycodeoxycholic acid | 0.4892 | -0.3255 |
| Ursodeoxycholic acid | 0.4231 | -0.2480 |
| Deoxycholic acid | 0.1849 | -0.0932 |
| Glycocholic acid | 0.4917 | -0.3808 |
| Taurocholic acid | 0.2268 | 0.4508 |
| Chenodeoxycholic acid | 0.3848 | -0.2531 |
| Bilirubin | 0.2351 | -0.1412 |
| Ginsenoside Rb1 | 0.4022 | -0.2732 |
| Ginsenoside Re | 0.0505 | 0.4481 |
| Ginsenoside Rd | 0.1929 | 0.7315 |
| Ginsenoside Rg1 | 0.4704 | 0.4590 |
| Notoginsenoside R1 | 0.0401 | 0.5545 |
| Muscone | -0.1091 | -0.3947 |