



Figure S1 Multiple sequence alignment of DBD domain of SmHsf proteins from *S. miltiorrhiza*. The different colors represent three α -helices and four β -sheets. Sequences were aligned using MEGA with Muscle by default parameters (version 11). Gradient blue colors represent sequence consensus from low to high.

Table S1 Sequence characteristics of heat shock transcription factors (SmHsfs) in *S. miltiorrhiza*.

| Gene name | Gene ID | Strand | CDS (bp) | Exons | Protein length (aa) | MW (kDa) | Instability Index | GRAVY | PI | Localization |
|-----------|------------|--------|----------|-------|------------------------|-------------|----------------------|-------|------|--------------------------|
| SmHsf1 | EVM0002416 | - | 873 | 3 | 291 | 31.36 | 65 | -0.61 | 5.89 | Nuclear |
| SmHsf2 | EVM0014513 | + | 1095 | 2 | 365 | 42.05 | 76.59 | -0.70 | 5.84 | Nuclear |
| SmHsf3 | EVM0027188 | - | 1026 | 2 | 342 | 37.90 | 67.8 | -0.49 | 8.22 | Nuclear |
| SmHsf4 | EVM0020872 | + | 1074 | 4 | 358 | 41.64 | 61.23 | -0.91 | 5.72 | Nuclear |
| SmHsf5 | EVM0022616 | - | 702 | 2 | 234 | 26.51 | 65.32 | -0.80 | 8.74 | Nuclear |
| SmHsf6 | EVM0014116 | - | 951 | 2 | 317 | 34.59 | 78.99 | -0.48 | 4.88 | Nuclear |
| SmHsf7 | EVM0020650 | + | 1038 | 3 | 346 | 37.89 | 66.75 | -0.59 | 5.36 | Nuclear |
| SmHsf8 | EVM0018201 | - | 1005 | 4 | 335 | 38.37 | 65.99 | -0.80 | 5.8 | Nuclear |
| SmHsf9 | EVM0013164 | - | 1527 | 3 | 509 | 56.36 | 76.38 | -0.55 | 5.26 | Nuclear |
| SmHsf10 | EVM0004220 | - | 987 | 2 | 329 | 37.62 | 74.27 | -0.68 | 6.29 | Nuclear |
| SmHsf11 | EVM0003811 | - | 780 | 3 | 260 | 29.74 | 77.53 | -0.72 | 9.19 | Cytoplasmic Nuclear |
| SmHsf12 | EVM0013673 | + | 1449 | 2 | 483 | 53.64 | 66.78 | -0.75 | 5.36 | Nuclear |
| SmHsf13 | EVM0001289 | - | 1059 | 2 | 353 | 39.75 | 71.7 | -0.62 | 5.92 | Nuclear |
| SmHsf14 | EVM0012238 | + | 1167 | 2 | 389 | 43.87 | 68.09 | -0.69 | 5.72 | Nuclear |
| SmHsf15 | EVM0000817 | - | 798 | 2 | 266 | 30.54 | 71.4 | -0.68 | 6.72 | Nuclear |
| SmHsf16 | EVM0024007 | + | 759 | 2 | 253 | 28.70 | 74.33 | -0.58 | 5.9 | Nuclear |
| SmHsf17 | EVM0014653 | + | 783 | 2 | 261 | 30.16 | 78 | -0.66 | 5.77 | Nuclear |
| SmHsf18 | EVM0012567 | + | 564 | 2 | 188 | 21.66 | 64.12 | -0.59 | 9.76 | Mitochondrial Nuclear |
| SmHsf19 | EVM0017861 | - | 873 | 2 | 291 | 33.42 | 71.24 | -0.63 | 6.92 | Nuclear |

| Gene name | Gene ID | Strand | CDS (bp) | Exons | Protein length (aa) | MW (kDa) | Instability Index | GRAVY | PI | Localization |
|-----------|------------|--------|----------|-------|------------------------|-------------|----------------------|--------|------|------------------------|
| SmHsf20 | EVM0007953 | + | 1278 | 2 | 426 | 48.24 | 68.35 | -0.75 | 5.44 | Nuclear |
| SmHsf21 | EVM0002514 | + | 1509 | 2 | 503 | 55.09 | 62.01 | -0.62 | 4.78 | Nuclear |
| SmHsf23 | EVM0012123 | - | 1152 | 2 | 384 | 43.55 | 71.31 | -0.65 | 4.64 | Nuclear |
| SmHsf24 | EVM0006191 | - | 684 | 2 | 228 | 26.43 | 68.77 | -0.73 | 6.97 | Nuclear |
| SmHsf25 | EVM0021531 | + | 942 | 2 | 314 | 36.13 | 66.68 | -0.79 | 5.11 | Nuclear |
| SmHsf26 | EVM0028007 | + | 1200 | 2 | 400 | 45.23 | 68.67 | -0.73 | 5.16 | Nuclear |
| SmHsf27 | EVM0004954 | + | 564 | 2 | 188 | 22.13 | 65.24 | -0.90 | 9.93 | Nuclear |
| SmHsf28 | EVM0003128 | - | 1011 | 2 | 337 | 37.66 | 66.76 | -0.50 | 4.89 | Nuclear |
| SmHsf29 | EVM0007734 | - | 1062 | 2 | 354 | 39.55 | 64.67 | -0.72 | 5.7 | Nuclear |
| SmHsf30 | EVM0022005 | - | 1464 | 2 | 488 | 54.26 | 66.45 | -0.57 | 5 | Cytoplasmic Nuclear |
| SmHsf31 | EVM0010800 | + | 1026 | 2 | 342 | 39.15 | 62.02 | -0.73 | 5.39 | Nuclear |
| SmHsf32 | EVM0006922 | - | 675 | 2 | 225 | 25.71 | 78.75 | -0.63 | 8.32 | Nuclear |
| SmHsf33 | EVM0013214 | + | 810 | 2 | 270 | 29.51 | 60.26 | -0.68 | 6.34 | Nuclear |
| SmHsf34 | EVM0003304 | - | 882 | 2 | 294 | 33.68 | 57.61 | -0.87 | 8.43 | Nuclear |
| SmHsf35 | EVM0012493 | + | 1089 | 3 | 363 | 40.94 | 70.25 | - 0.66 | 4.84 | Nuclear |

CDS, coding sequece; MW, molecular weight; GRAVY, grand average of hydropathicity; pI, isoelectric point

Table S2 Functional domains analysis of SmHsfs in *S. miltiorrhiza*

| Gene name | DBD | HR-A/B | NLS | NES | RD | AHA |
|-----------|--------|---------|---------------------------------------|------------------------|------------|-----------------------|
| SmHsf1 | 22-115 | 165-198 | (249) LKRLKRSENGNGGGGERMAKDVKSE | N.D. | (242) LFGV | N.D. |
| SmHsf2 | 56-149 | 166-209 | (248) GEVLKKRRRLA | (199) SHLAAVEKRLRITE | N.D. | N.D. |
| SmHsf3 | 21-114 | 175-202 | (305) KKRLHPECNSTAATTNKTRLVLEKDDL | N.D. | (297) LFGV | N.D. |
| SmHsf4 | 40-133 | 172-206 | (312) EDLLKQGFDEGEKLVKDEDAEVYHKNQRF | (271) QMSELEALALQMQ | N.D. | N.D. |
| SmHsf5 | 18-111 | 140-182 | (210) EKEKKRKRAHE | N.D. | (200) LFGV | N.D. |
| SmHsf6 | 28-121 | 155-188 | (116) KTIKRRRNTS | (274) DPITDDVLEMLLSEED | N.D. | N.D. |
| SmHsf7 | 23-116 | 171-205 | (6) VDRSGSGESTAGDAPRSLPTPFLTKTYQL | N.D. | (281) LFGV | N.D. |
| SmHsf8 | 34-127 | 167-202 | (220) HKEKRKELEEAIKKRRRPI | (161) KLVLVTELVKLRQQQ | N.D. | N.D. |
| SmHsf9 | 14-107 | 134-171 | (212) RHISGANKKRRLPNQDEESLARKLSITS | (167) SQLQTVGQRVHVMEQ | N.D. | (444)NDVFWEQFLPESPLI |
| SmHsf10 | 38-131 | 153-248 | (235) IGKKRRKKID | N.D. | N.D. | N.D. |
| SmHsf11 | 38-94 | 107-202 | (189) IGKKRRKKID | (226) EAQDFDDLKVGWD | N.D. | N.D. |
| SmHsf12 | 16-109 | 125-180 | (207) YNKKRRRLPQ | N.D. | N.D. | (429)DVFWEQFLTERPGC |
| SmHsf13 | 46-139 | 145-181 | (23) ESLPLRPPPSVASFDSRPLASPPIPRFLSKTY | N.D. | N.D. | N.D. |
| SmHsf14 | 10-103 | 120-148 | (193) EFQRKKRRSVALNF | N.D. | N.D. | (362) NDVFWQQFLTEAP |
| SmHsf15 | 24-118 | 136-183 | (114) NMKRRSLQMSPTPPRQKNSLSP | (245) QHAIAIAQDFQVWLTE | N.D. | N.D. |
| SmHsf16 | 22-116 | 125-162 | (111) SNMKRRALQM | N.D. | N.D. | N.D. |
| SmHsf17 | 13-107 | 125-172 | (102) PNMKRRSEQNNPTPPRQKNSLSP | (242) QHAIAIAQDFQVWLIE | N.D. | N.D. |
| SmHsf18 | 16-108 | 130-159 | (6)RVGSKPHKMKCPAPFLSKTYDLLDAVERVVS | (152) QISHFKAVEMRLLE | N.D. | N.D. |
| SmHsf19 | 21-114 | 159-189 | (258) KKRLHSEFNSSNKSRLVLEKDDLGLNLMP | N.D. | (249) LFGV | N.D. |
| SmHsf20 | 10-103 | 114-171 | (193) FPNKKRRLLALQY | N.D. | N.D. | (358)DMFWQQFLTEAPAS |
| SmHsf21 | 29-122 | 150-181 | (232) EGNKKRRRLKQD | (265) EAAKAMLRLQIMKLD | N.D. | (440)SDPFWEKFLQSPPPV |
| SmHsf22 | 2-88 | 146-185 | N.D. | N.D. | N.D. | N.D. |
| SmHsf23 | 10-103 | 130-183 | N.D. | (177) QQMLSFIVMAMQSPE | N.D. | (304) NDAVLEQLLLPYPST |
| SmHsf24 | 22-115 | 129-160 | (172)RARACKLYMTENVKPLELLPMTRFCQEMTR | (210) SAEHHMDLELRPLGD | N.D. | N.D. |

| Gene name | DBD | HR-A/B | NLS | NES | RD | AHA |
|-----------|--------|---------|-------------------------------------|------------------------|------------|----------------------|
| SmHsf25 | 19-112 | 145-195 | (105) LLKTIKRRRKQT | N.D. | N.D. | N.D. |
| SmHsf26 | 11-104 | 121-177 | (198) HDRKRRFPG | (243) ELLDQIDSSMSLWE | N.D. | (335)NDVFWEQFLTENPGS |
| SmHsf27 | 18-116 | 135-170 | (13) RRKMKSAPFVSKTYDLLEAEAEERRGGEK | (162) QIAHFKALQLSLLQ | N.D. | N.D. |
| SmHsf28 | 21-114 | 137-173 | N.D. | N.D. | N.D. | N.D. |
| SmHsf29 | 15-108 | 118-164 | (190) RLTPDKKRKLM | N.D. | N.D. | N.D. |
| SmHsf30 | 93-186 | 212-259 | N.D. | N.D. | N.D. | N.D. |
| SmHsf31 | 63-156 | 180-229 | (256) GMSKKRRRVACE | (207) DMAAVRERLRVTE | (231) LFGV | N.D. |
| SmHsf32 | 17-110 | 134-180 | (150) ENKRLKRENDALNSELLLLKRKCQQL | (200) QALKLFGVRLEVGSE | (204) LFGV | N.D. |
| SmHsf33 | 6-99 | 149-186 | (238) KKRGCCSGGGADLCGPHKKQMKPW | (184)DELIAFLTQCVKVAPDE | N.D. | N.D. |
| SmHsf34 | 61-154 | 175-220 | (26) RVKEEEESFMFSAAAAEGDQNQPPKAMPME | (201) SSMAAVRERLRVTE | N.D. | N.D. |
| SmHsf35 | 39-132 | 155-205 | (125) LLKTIKRRRRNVM | N.D. | N.D. | N.D. |

DBD, DNA binding domain; HR-A/B, Oligomerization domain; NLS, Nuclear localization signal; NES, Nuclear export signal; AHA, Transcriptional activation domain; RD, Repressor domain; N.D., not detected. For the NLS, NES, RD and AHA column, the numbers in parenthesis are the start site of the functional domain.

Table S3 The detailed information of conserved motifs in SmHsfs by MEME tools

| Motif | E-value | Width | Best Possible Match |
|-------|-----------|-------|---|
| 1 | 5.7e-1034 | 41 | FVVWDPPEFSRDLLPKYFKHNNFSSFVRQLNTYGFRKVDPD |
| 2 | 4.2e-513 | 29 | RWEFANEGFLRGQKHLLKNIRRRKTSSSS |
| 3 | 7.9e-473 | 29 | PPPFLTKTYEMVDDPSTDSIISWSRDGNS |
| 4 | 5.7e-255 | 41 | CVESGRFGLEEENERLRREKNVLMaelVKLRQQQQSTRAQL |
| 5 | 9.6e-152 | 29 | ZRLQGTEQKQQQMMSFLAKALQNPDFLQQ |
| 6 | 3.1e-022 | 11 | PQPM EGLHEAG |
| 7 | 1.0e-019 | 15 | NDVFWEQFLTEAPGS |
| 8 | 1.3e-014 | 23 | JVQHKEKKKEIEEAIGKKRRKKI |
| 9 | 3.0e-016 | 50 | VPHHHHPITGGFFPYPSRLSISPTDSDDHPWGESPPPLASPTGGGYHSV |
| 10 | 1.1e-009 | 41 | DHQELVFQDREGGDDHGNYISFGGGVKVEAQDFDDLKVG EW |