

Supplementary information

Limited proteolysis data: Digestion fragments produced in the limited proteolysis experiments performed on Hsp27 or on the complexes HAA/Hsp27 or **16**/Hsp27 were analyzed by MALDI/MS instrument operating in reflectron or in liner mode, depending on the m/z range. Achieved results are summarized in the tables S1-S3. An example of the MALDI spectra acquired in these analyses is reported (figure S1).

Table S1: Digestion fragments observed by MALDI/MS in the limited proteolysis experiments performed on Hsp27 or on the complexes HAA/Hsp27 or **16**/Hsp27 using trypsin.

Molecular mass	Peptide	Hydrolysis site(s)
1372.8	1-11	R11
21296^a	12-204	R11
1901.9 ^b	12-26	R11; R26
3046.5 ^b	12-36	R11; R36
5265	12-55	R11; R55
20593	12-197	R11; K197
3256.6 ^b	1-26	R26
19411^b	27-204	R26
1162.6 ^b	27-36	R26; R36
3377.7 ^b	27-55	R26; R55
18708^b	27-197	R26; K197
4404^b	1-36	R36
18265^b	37-204	R36
2233.1 ^b	37-55	R36; R55
17563^b	37-197	R36; K197
6620	1-55	R55
16049	56-204	R55
15346	56-197	R55; K197
21949	1-197	K197

^a Masses reported in bold were observed by MALDI/MS instrument operating in linear mode

^b These species were not detected in the digestion performed on the HAA/Hsp27 complex.

Table S2: Digestion fragments observed by MALDI/MS in the limited proteolysis experiments performed on Hsp27 or on the complexes HAA/Hsp27 or 16/Hsp27 using chymotrypsin.

Molecular mass	Peptide	Hydrolysis site(s)
3369.7 ^b	1-27	L27
19297^b	28-204	L27
11710^b	28-132	L27; Y132
3978.0 ^b	1-32	F32
18689^b	33-204	F32
4404^b	33-132	F32; Y132
15064	1-132	Y132
7606	133-204	Y132

^a Masses reported in bold were observed by MALDI/MS instrument operating in linear mode

^b These species were not detected in the digestion performed on the HAA/Hsp27 complex

Table S3: Digestion fragments observed by MALDI/MS in the limited proteolysis experiments performed on Hsp27 or on the complexes HAA/Hsp27 or 16/Hsp27 using GluC.

Molecular mass	Peptide	Hydrolysis site(s)
1915.0	1-16	D16
20238^a	17-204	D16
19780	17-194	D16; E194
21278	1-194	E194

^a Masses reported in bold were observed by MALDI/MS instrument operating in linear mode

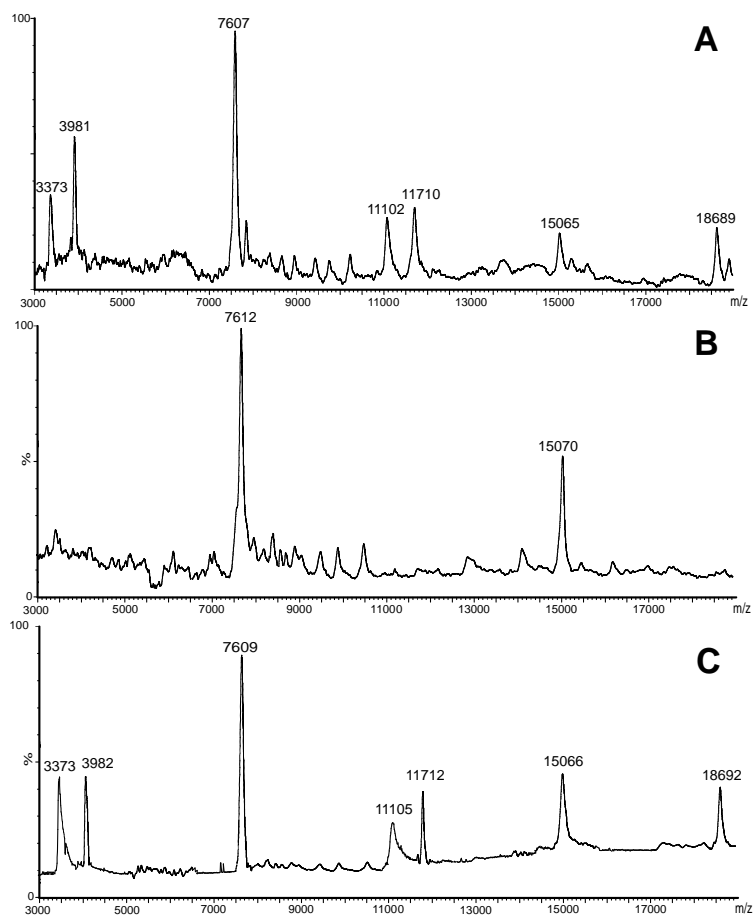


Figure S1: MALDI/MS spectra of the digestion fragments produced in the limited proteolysis experiments performed on Hsp27 (A), on the HAA/Hsp27 complex (B) or on the 16/Hsp27 complex using chymotrypsin as a proteolytic agent.