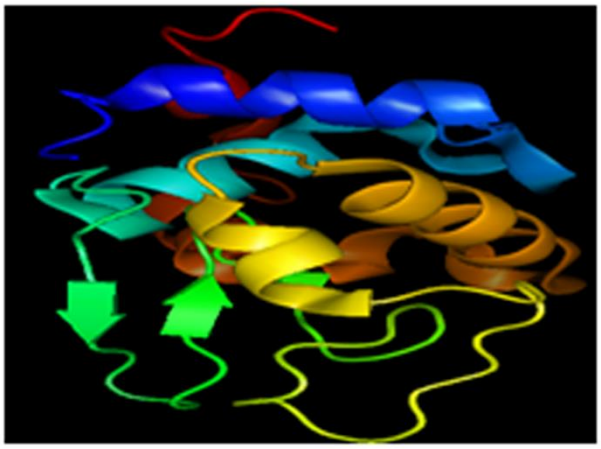
Supplementary Information



SI-I: 3D- Structure of Lysozyme.

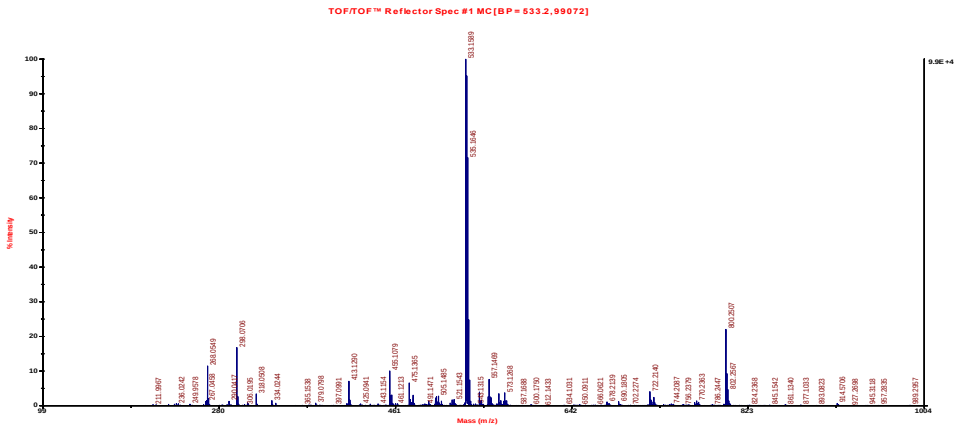
10	20	30	40	50
KVFGRCELAA	AMKRHGLDNY	RGYSLGNWVC	AAKFESNFNT	QATNRNTDGS
60	70	80	90	100
TDYGILQINS	RWWCNDGRTP	GSRNLCNIPC	SALLSSDITA	SVNCAKKIVS
110	120	129		
DGNGMNAWVA	WRNRCKGTDV	QAWIRGCRL		

SI-II: Primary structure of lysozyme.

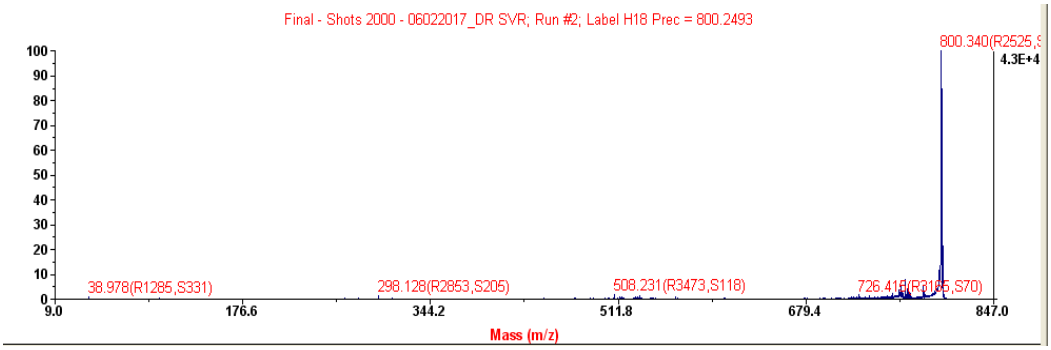
Solutions required	(1) 100% Acetonitrile
	(2) 100 mM Ammonium Bicarbonate
Wash solution	(3) 50% Acetonitrile and 50 mM Ammonium Bicarbonate
Reduction solution	(4) 5 mM DTT and 100 mM Ammonium Bicarbonate
Alkynlation solution	(5) 10 mM iodoacetamide in 100 mM Ammonium Bicarbonate
Trypsin solution	(6) 20 µg/ml
Extraction solution	(7) 0.1% formic acid and Acetonitrile

SI-III: Protocol for trypsin digestion, this protocol describes the digestion of the protein present with trypsin. The band was excised from the SDS-PAGE. All the reagents used were prepared immediately to prior to use.The water used milliQ water. Acetonitrile and methanol used were of HPLC Grade.

The 'dimeric' band was excised SDS-PAGE and cut into 1 × 1 mm pieces and transferred into a sterile micro centrifuge tube. Washed the gel with 500 µL of wash solution (50% acetonitrile- 50 mM ammonium Bicarbonate) and incubated at room temperature for 15 min. with gentle agitation. Removed the solution with a pipette, till the Coomassie dye was completely removed. Dehydrated the gel in 100% acetonitrile for 5 min. When dehydrate, the gel pieces had an opaque white color and were significantly smaller in size. Removed acetonitrile with a pipette and then completely dried the gel at room temperature for 10-20 mins. in a centrifugal evaporator. Rehydrated the gel pieces in 150 µL reduction solution (10m MDTT, 100 mM ammonium bicarbonate) for 30 min at 60°C. Discarded the reduction solution with a pipette and added 100 µL alkylation solution (50 mM iodoacetamide,100mM ammonium bicarbonate) and incubated for 30 min in the dark at room temperature. Discarded alkylation solution with a pipette and added 500 µL of wash solution and incubated at room temperature for 15 min. with gentle agitation and then discarded wash solution and dehydrated the gel in 100 µL 100% acetonitrile for 5 min. Discarded acetonitrile and completely dried the gel at room temperature in a centrifuge evaporator, while gel was drying prepared protease digestion solution. Re-suspended lyophilized trypsin (20 µg/vial) in 1 ml of 50 mM ammonium bicarbonate, aliquot (50 µL/tube0 and store at -80°C. Ensured that freeze-thaw was not done more than twice. Rehydrated the gel with minimal volume of protease digestion solution. Used 20 µL for small gel plugs. Added more of the solution, if the gel pieces absorbed and all the liquid gel pieces were hydrated throughout the digest. Digested these overnight at 37°C for 18 hr. After overnight incubation, spun down the samples brief centrifugation. Transferred the supernatant (containing additional typical peptide) to a fresh tube. Extracted the gel with an additional 25-50 µl of extraction solution, Agitated the gel pieces by sonicating in a water bath for 10 min or with gentle vortexing. Spun down the sample and transferred the supernatant to a tube. Dried the pooled extracted peptides by centrifugal evaporation to dryness, without using heat and not drying for extended time.



SI-IV-A: MALDI-MS of the new crosslinker



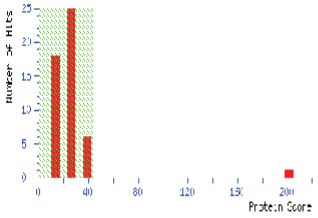
SI-IV-B: MS/ MS of the m/z 800 peak

MATRIX SCIENCE **Mascot Search Results**

User :
Email :
Search title :
Database : SwissProt 57.15 (515203 sequences; 181334896 residues)
Timestamp : 8 Mar 2017 at 10:28:32 GMT
Protein hits : LYSC_CHICK Lysozyme C OS=Gallus gallus GN=LYZ PE=1 SV=1

Mascot Score Histogram

Ions score is -10*Log(P), where P is the probability that the observed match is a random event.
Individual ions scores > 44 indicate identity or extensive homology (p<0.05).
Protein scores are derived from ions scores as a non-probabilistic basis for ranking protein hits.



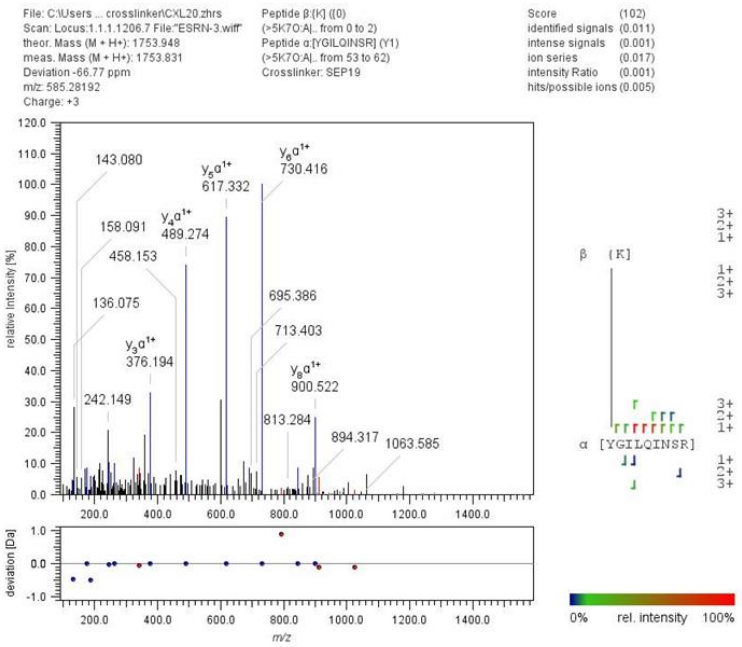
Peptide Summary Report

Format As: Peptide Summary — Help
Significance threshold p< 0.05 — Max. number of hits: AUTO
Standard scoring — MudPIT scoring — Ions score or expect cut-off: 0 — Show sub-sets: 0
Show pop-ups — Suppress pop-ups — Sort unassigned: Decreasing Score — Require bold red
Select All | Select None | Search Selected | Error tolerant | Archive Report

1. LYSC_CHICK Mass: 16228 Score: 202 Matches: 6(4) Sequences: 3(3)
Lysozyme C OS=Gallus gallus GN=LYZ PE=1 SV=1
Check to include this hit in error tolerant search or archive report

Query	Observed	Mr(expt)	Mr(calc)	Delta	Miss	Score	Expect	Rank	Unique	Peptide
16	1045.4727	1044.4654	1044.5352	-0.0699	0	59	0.0025	1	U	K.GTDVQAWIR.G
17	1045.4860	1044.4787	1044.5352	-0.0566	0	(21)	17	8	U	K.GTDVQAWIR.G
19	1428.5587	1427.5514	1427.6429	-0.0915	0	75	5.3e-005	1	U	K.FESNFNTQATNR.N
20	1428.5647	1427.5574	1427.6429	-0.0855	0	(31)	1.6	1	U	K.FESNFNTQATNR.N
21	1675.7053	1674.6980	1674.7937	-0.0956	0	67	0.00037	1	U	K.IVSDGNGMNAWVAWR.N
22	1675.7086	1674.7013	1674.7937	-0.0923	0	(45)	0.054	1	U	K.IVSDGNGMNAWVAWR.N

SI-V: Mascot analysis of the trypsin digested ‘dimeric’ band.



SI-VI: The ‘b’ and ‘y’ ions obtained for fragment m/z.1753.831 via StavroX 3.6.0.1.