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Novel protein identification method by observation of metastable ions in matrix-assisted laser desorption ionization mass spectrometry

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Abstract

Peptide mass fingerprinting (PMF) is one strategy for protein identification by mass spectrometry (MS) analysis. The input data used in the PMF are mass lists of an enzymatic-digested protein extracted from matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) MS spectra. The MALDI process can lead to post-source decay (PSD) fragmentation, which occurs after the acceleration region of the ionization source. The PSD corresponds to the fragmentation of metastable ions. Herein, we present an enhanced PMF method by introducing a metastable ion relative C-terminal amino acid truncation of peptides derived from LysN protein digested peptides. In comparison to conventional PMF, highly reliable identification of this new method was verified by using three parameters of C-terminal amino acid sequence, N-terminal Lys and mass for database search. A non-probability-based sequence database search algorithm was developed for protein identification, especially for protein mixture, which showed advantageous in a way to common probability-based sequence database search algorithm. Because of its highly desirable property, the novel method could support the conventional PMF to find more applications in the analysis of protein identification.

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Contents

1 Chapter I: General Introduction and Aims

1.1 Mass Spectrometry

Mass spectrometry (MS) is an analytical technology that ionizes chemical substances and systematizes the ions based on the mass-to-charge ratio (*m/z*). MS is applied to many different fields and is acted on pure sample or complex mixtures as well. A mass spectrum is a plot of the intensity of ion signal on the basis of *m/z*. These spectra are used to identify the masses of particles or molecules, to measure the elements or isotopes, and to illustrate the chemical structures of molecules.

MS was invented by J.J. Thomson at the Cavendish Laboratory in the University of Cambridge. Thomson first used his apparatus to measure *m/z* of electrons in 1897, and he received the 1906 Nobel Prize in Physics by this work in “discovering” the electron. He established the foundation of the MS field according to his early work on cathode. The first mass spectrometer to measure the *m/z* of charged atoms was built by Thomson and F. Aston. Mass spectrometers were redesigned by Aston and others in order to improve the resolution in the early decades of the 20th century. Under their efforts on the development of mass spectrometers, elemental isotopes were proved to be existed and separated by MS. Furthermore, MS became a remarkable analytical tool really during the World War II, since the isotopes played significant roles developed by E.O. Lawrence in the Manhattan Project. (Griffiths 2008)

Commercial mass spectrometers were launched in the 1940s, and MS was still dominated by physicists and industrial chemists as a useful technique. K. Biemann made great contributions to apply the MS to chemistry field. He used

MS to determine the structure of complex molecules, founded basic regulation for peptide fragmentation; even designed an early peptide sequencing method. Consequently, A. Marshall and M.Comisarow first applied Fourier transform (TF) to ion cyclotron resonance (ICR) MS. FTICR MS is one of the most important technologies for complex mixture analysis since its ultrahigh resolution power. Then, in 1988, electrospray ionization (ESI) and matrix-assisted laser desorption/ionization (MALDI) appeared on the MS field. These ionization techniques revolutionized biological MS and are served as the predominant types of ionization sources for macromolecule at present (Griffiths 2008).

A common mass spectrometer consists of three main modules: ion source, analyzer, and detector (Figure. 1). Ion source is a device to ionize the analyte molecules to form charged ions by laser, electrospray, plasma, etc. As above mentioned, ESI and MALDI are the most common ion sources used in proteomics. Various analyzers can connect to ion sources to assemble the core part of mass spectrometers, such as time-of-fight (TOF), quadrupole (Q), ion trap (IT) and FTICR. The ion source generated charged ions are separated according to their *m/z* in the analyzer, and the ion intensity at different *m/z* values are recorded by detector. A visual mass spectrum are output by computer finally.

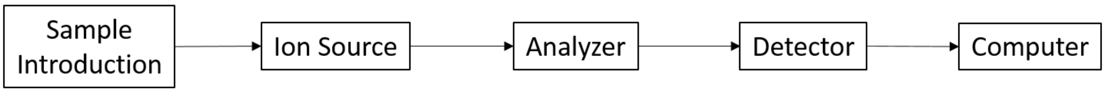


Figure. 1 Simplest form of MS.

1.1.1 Ion Source

1.1.1.1 Matrix-Assisted Laser Desorption Ionization (MALDI).

Matrix-assisted ultraviolet laser desorption/ionization (MALDI) was prensented by Karas and Hillenkamp in 1984, which can rapidly measure the masses of non-volatile biological compounds with a TOF mass spectrometer (Karas et al. 1987, Karas, Gluckmann & Schafer 2000). It has become a widely used and strong technique to ionize a series molecules including proteins, peptides, DNA, RNA, polymers and large inorganic compounds in the gas-phase. One feature of MALDI to be different with other ionization technique is the use of MALDI matrix, which support the desorption and ionization of MALDI. Note that Tanaka and colleagues firstly acquired large proteins' mass spectra by using a combination of nano-metal particles and glycerol, instead of the MALDI matrix, in laser-desorption ionization TOFMS (Tanaka et al. 1988). The advantages of MALDI are the easy sample preparation, fast speed for data analysis, and a good tolerance to contamination (Stump et al. 2002, Chen, Carroll & Beavis 1998).

As to the full name of MALDI, “matrix” indicates a basic characteristic of MALDI MS, which is the appropriate matrix materials supported ionization on analytes. The success of MALDI ionization depends on good sample preparation, thus the matrix selection and the sample preparation optimization control the quality of the ionization results. Several substances have been measured as MALDI matrices. The wavelength of laser used in the MALDI determines the selection of suitable matrix. Small organic molecules with a wavelength absorption in the range of 266-355 nm are the most frequently used matrices in MALDI. α-Cyano-4-hydroxycinnamic acid (CHCA) is one of the most common matrices for peptides/proteins analysis by MALDI (Beavis, 1992).

The process of MALDI is separated into two steps. There are two ways of the first step, dropping the analyte, which is dissolved in a matrix containing solvent, on a MALDI plate before analysis till the mixture is dried; or dropping the solution of matrix on the plate first, then dropping the sample compound on the dried matrix. After drying, the analyte-matrix mixture form crystals named “solid solution”. The analyte molecules are isolated from each other completed by embedding the matrix all over. In the second step, the majority of the “solid solution” is desorbed by strong laser impulse in the vacuum inside the ion source of MS in a short time (Knochenmuss 2002, Karas, Krüger 2003). The excitation of the matrix leads to the large energy accumulation in the condensed phase, consequently rapid heating of the crystals is induced by irradiation of the laser pulse. Then the matrix is sublimated and expanded into the gas phase caused by the rapid heating, along with the desorption of the surface of crystals. Finally, the analyte is entrained with the gaseous matrix and joins the expanding matrix plume (Dreisewerd 2003).

There are many different pathways, either chemical or physical, to explain the origin of ions generated in MALDI, such as excited state proton transfer, gas-phase photo ionization, desorption of performed ions and in-molecule reactions, but is not completed understood yet. (Zenobi, Knochenmuss 1998, Knochenmuss, Zenobi 2003). Proton transfer and gas-phase photoionization are two most widely accepted mechanisms of ion formation in MALDI. The differences between these two mechanisms are the time. The former occurs in the solid phase, and the latter proceeds in the sublimated matrix plume. An electrostatic field supplied by high voltage accelerate the gas-phase ions towards the analyzer (Figure. 2). In comparison to other laser ionization techniques, MALDI is more sensitive and general. The highly increased sensitivity reflects on two points. The first point is the formation of analyte sample clusters are prevented by a huge number of matrix molecules separate the embedded analyte molecules exhaustively. Second, since the matrix absorb most of the energy from the incident laser pulse, the damage to the samples is decreased, and prompt the laser transfer energy to the analyte efficiently. As to the generalizability, there are also two aspects. First, since the matrix molecules absorb the laser pulse, the adjustment of wavelength of laser to match the absorption range of every sample molecules is unnecessary. In the second place, macromolecules with high masses around 100,000 Da can be allowed by MALDI because the ionization process does not depends on the properties of analyte, such as absorption and size (Spengler, Cotter 1990).

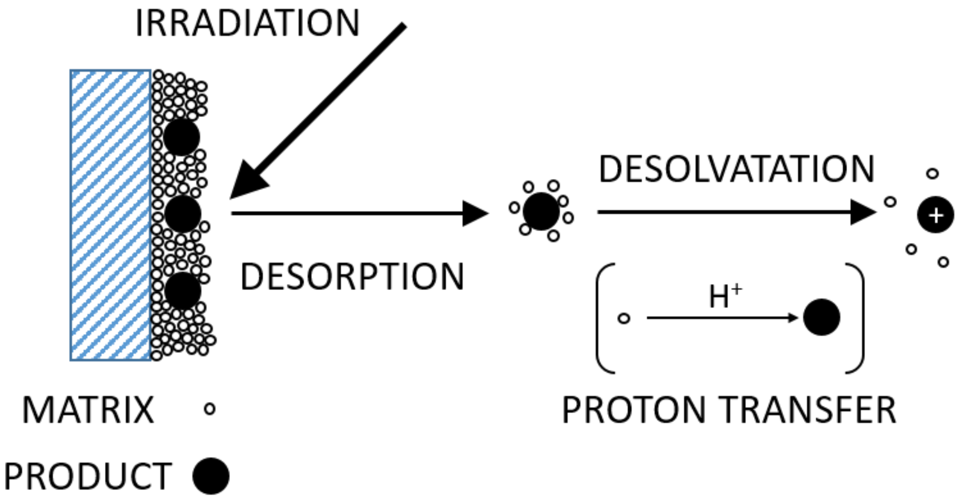


Fig. 2 Diagram of the principle of MALDI.

1.1.1.2 Electrospray Ionization (ESI)

ESI was first described by J. B. Fenn and colleagues who showed that the low molecular weight (low to 2000 Th) of multiply charged ions generated from proteins can be measured with instruments (Fenn et al. 1989). In the early stage, ESI was considered to ionize protein for proteomics, then the application field was expanded to the small polar molecules, polymers and biopolymers.

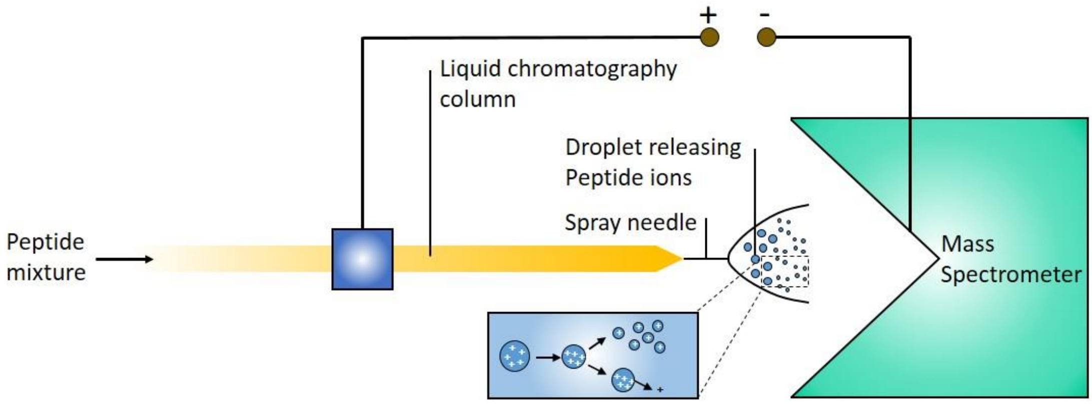


Figure. 3 Diagram of electrospray ionization (ESI)

Figure. 3 describes the structure of ESI. A biomolecules containing solution is pumped through a needle with a very large voltage applied to it (2-5 kC). This forms small, highly charged droplets of solvent upon exit of the needle. As the droplet travels towards the orifice of the mass spectrometer, the solvent evaporates aided by the drying gas, and it shrinks and the surface charge density increase. This causes the droplet to blow apart and the process repeats itself until charged biomolecules emerge.

The peptides containing liquid effluent is electrostatically dispersed by eluting from a syringe or HPLC eluent through a needle with a high voltage (several kV) applied to it. It forms highly charged droplets of solvent departure the needle. The solvent evaporates easily by the aid of drying gas during the droplets travel to the orifice, it increases the surface charge density of the droplets and decreases their size. High electrical fields allow the desorption of analyte ions on the surface of droplet to generate desolvated ions, and repetitive droplet fission, which sustain to each droplet contains only one analyte ion averagely, leads to the formation of tiny droplets.

1.1.2 Analyzer

1.1.2.1 Time-of-Flight (TOF)

The time-of-flight (TOF) analyzer was first come up with in 1946 by W. E. Stephens (Anonymous 1946). The linear TOF mass spectrometer was designed in 1955 and became the first commercial instrument later(Wiley, McLaren 1955). Since the end of 1980s, the development of MALDI settled new applications for both biomolecules and synthetic polymers. The basic principle of separation for TOF analyzer is based on velocities (flight time) of ions.

1.1.2.1.1 Linear Time-of-Flight Mass Spectrometer

Ions are separated by the TOF analyzer according to their velocity that depends on their *m/z* ratios when they travel through a free-field region (flight tube), after the electrostatic field induced acceleration followed by their formation during a laser pulse (Figure. 4). One advantage of TOF spectrometer is that it is suitable for soft ionization techniques, such as masses above 300 kDa samples can be observed by MALDI-TOF (Imrie, Pentney & Cottrell 1995, Moniatte et al. 1996), due to there is no limit for the upper mass range of it. Another benefit is the very high sensitivity results from their high transmission efficiency. For example, 10^{-15} mol of gramicidin (Lange et al. 1986) and 10^{-16} mol of protein mixture (Onnerfjord et al. 1998) have been detected by TOF analyzers. The third advantage is the analysis speed of TOF is very fast, a spectrum can be obtained in micro-seconds.

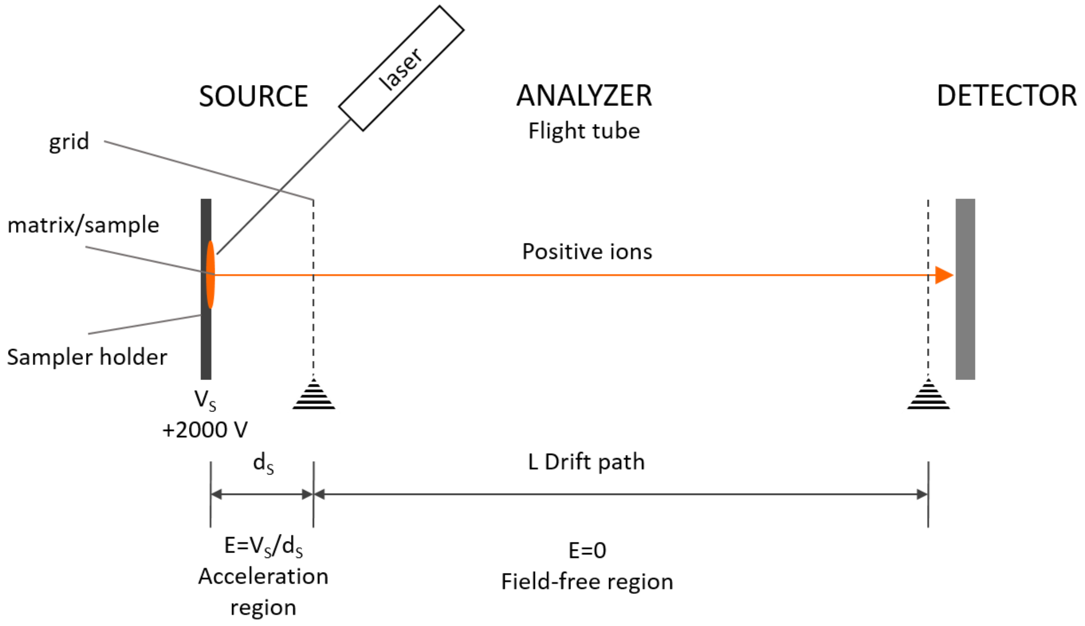


Figure. 4 Diagram of Linear TOF

1.1.1.1.2 Reflectron Time-of-Flight Mass Spectrometer

Electrostatic reflector, or reflection, is another way to improve mass resolution of TOF spectrometer. It was first described by Mamyrin (Mamyrin et al. 1973). The basic principle of reflectron is the ions are deflected by an ion mirror (retarding field) and are sent back through the field-free region. The reflectron is set at the end of the flight tube and the detector is set on the opposite side of the reflectron to receive the ions reflected from the reflectron (Figure. 5). The kinetic energy of the ions with the same *m/z* are corrected by reflectron thereby they can fly along different pathways in the reflectron. The distance penetrated in the reflectron of ions with less kinetic energy and less velocity is shallower than ions with more kinetic energy. Accordingly, the time cost in the reflectron by slower ions is short than the faster ions, and they arrive at the detector simultaneously.

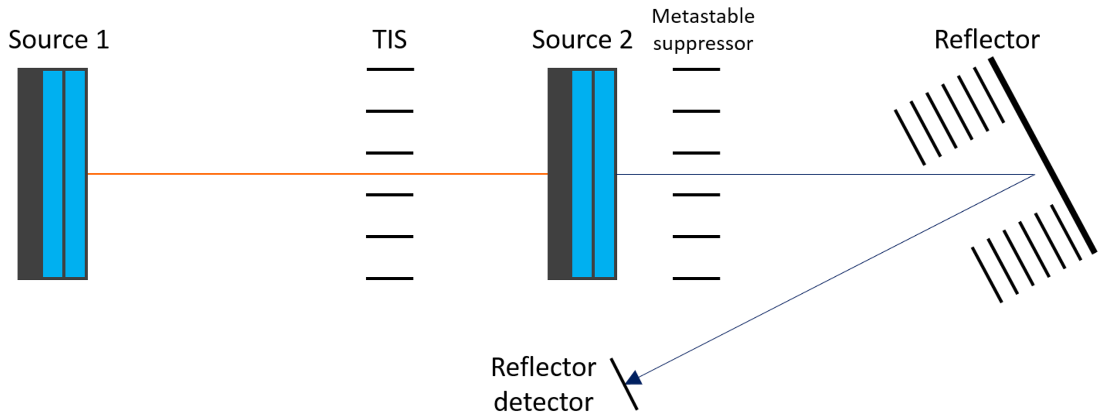


Figure. 5 Diagram of Reflectron TOF. Ion source 1: MALDI ion source. Ion source 2: re-acceleration cell. TIS: Timed ion selector.

1.2 Proteomics

The Human Genome Project (HGP) aiming to sequence the entire human genome was initiated by the US Department of Energy and National Institute of Health (NIH) from the late 1980 (Venter et al. 2001), and it has been achieved almost in 2003 by sequencing the human complete genome (Dunham et al. 1999). One major objective of HGP is to reveal and understand the linkage between genes and their related proteins by sequencing of the complete human genome. Whenever we mention the features of a protein, always including localization, expression, modification, interaction, domain structure, and activity. For an organism, not all encoded proteins express from genome at any time, and the protein expression changes at different stages of development, while the genome of each cell is almost same. For a single cell, different stimuli may change the dynamic proteome. The word “PROTEOME” is defined as the “total PROTEin complement of a genOME” (Wasinger et al. 1995), and “PROTEOMICS” started to attracted enormous attention after the completion of HGP (Figure. 6). The goal of proteomics is obtaining complete characterization of all proteins, understanding the protein-protein interactions, and illustrating the functions of those proteins. Proteomics is the bridge connected genomics and biology, the relationship between the proteome to physiological changes under the healthy or diseased conditions became one popular topic in modern biomedical research.

Figure. 6 Proteomics is a study of how the genome is expressed in proteins, and of how these proteins function and interaction.

1.1.2 Protein Identification

Analysis of the protein expression in organisms is one of the tasks of proteomics research. The 2D gel electrophoresis (2DE) is the first analysis tool in proteomics, proteins can be separated along two dimensions according to molecular masses and isoelectric points by gel electrophoresis (Bienvenut et al. 1999). With the development of MS, the 2DE-MS has been used for protein identification. 2DE resolved proteins can be addressed by in-gel digestion or membrane digestion (on immobilized trypsin containing membranes). Trypsin is the most commonly used protease in proteomics research, which cleaves at the C-terminal of lysine (Lys) and arginine (Arg). In additional, other proteases like AspN which cleaves before aspartate (Asp), GluC (V8) which cleave after glutamic acid (Glu) and LysC which cleave after Lys specifically are also used. The results of mass spectrometric analysis depend on the quality of the digestion, the digestion with high specificity and high cleavage efficiency lead to a good identification of protein. The development of newly engineered proteases promotes the improvement of cleavage methods (Willett et al. 1995). Moreover, the protein identification strategies of bottom-up sequencing and topdown sequencing are developed by measurement of intact proteins and peptide fragments together by MS.

1.1.2.1 Peptide Mass Fingerprinting

Peptide mass fingerprinting (PMF) is an analytical technology for protein identification. PMF was developed by many groups independently in 1993 (Henzel et al. 1993, James et al. 1993, Mann, Hojrup & Roepstorff 1993, Pappin, Hojrup & Bleasby 1993, Yates et al. 1993), and it is still the most common way of identifying proteins separated by 2DE or one-dimensional SDS-PAGE. Basically, the unknown proteins of interest are commonly digested by trypsin into smaller peptides, which cleaves at the C-terminal end of Lys and Arg specifically. The experimental data in the PMF based protein identification strategy is the mass list derived from MALDI-TOF or ESI-TOF mass spectrum of the enzymatic-digested protein. The experimental masses of the resulting tryptic peptides from unknown proteins are calculated to compare the theoretical peptide masses of each protein in a database *in silico* for acquiring a best match. MALDI-TOF is commonly used for PMF since in the spectrum of MALDI-TOF MS, the peptides display as singly charged ions, which is simple to interpret. However, an ESI mass spectrum appears peptides as multiply charged ions, the peptides masses should be deconvoluted for every search (Yates et al. 1993b). It is why ESI spectra is seldom used for PMF, though PMF can be used for protein identification by ESI. Another reason is that as the preferred instrument for PMF, MALDI-TOF has the ability to analyze several proteins in a single MS experiment by its high sample throughput.

Many database search software can be used for PMF searches (Perkins et al. 1999, Zhang, Chait 2000). The outcome output from those software is usually a ranked list of proteins according to the size of database, the size of input experimental data, the abundance of a specific peptide mass derived from a certain protein, the tolerance to the molecular mass, the allowed number of missed cleavages in one peptide sequence, the expected modifications, and other possible parameters The protein ranks at the first place is the unknown protein in the sample with the highest reliability. On the other hand, the algorithm design is also important for the database search result. Probability-based database search algorithm is popular used in many software. The ranking list of proteins can be produced after a series complicated calculation and computational modification.

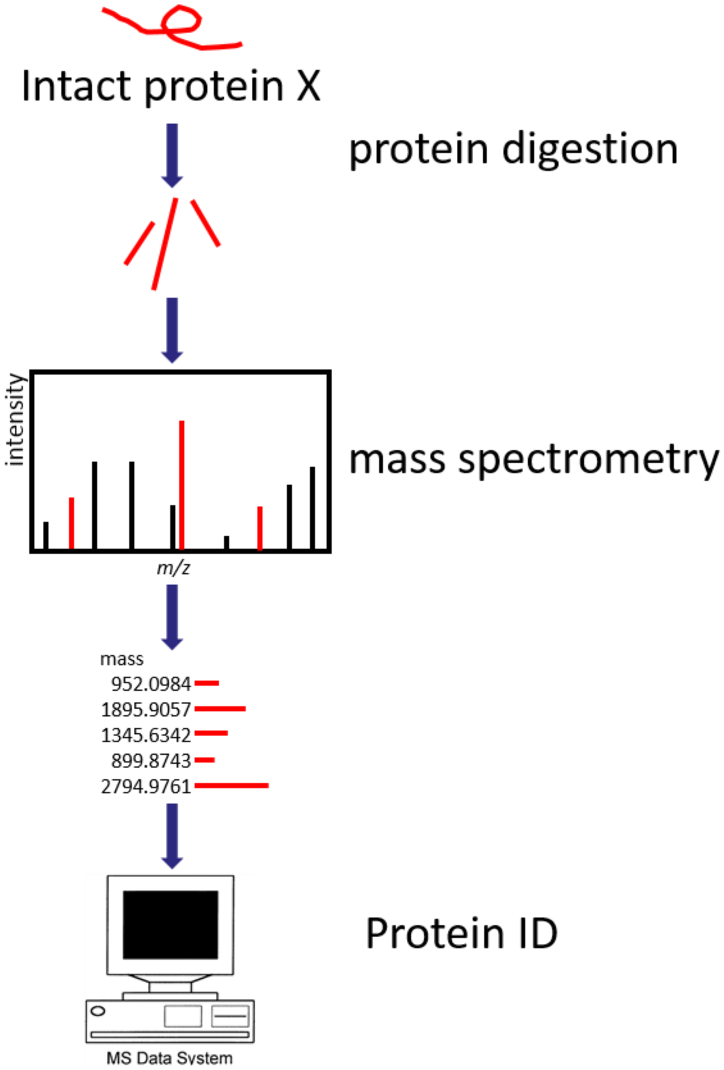


Figure. 7 Scheme of PMF. 1. Make proteolytic peptide fragments. 2. Measure peptide masses. 3. Match peptide masses to protein or nucleotide sequence database.

1.2 Aims

In the present study, I demonstrate a MALDI-based post-source fragmentation method that allows the C-terminal amino acid sequence in peptide sequences can be inferred for the digested peptides, which are produced by LysN or AspN proteases, to be performed. The data obtained for the constitute peptides are molecular masses, N-terminal amino acid sequences and C-terminal amino acid sequences are unique in the present method. Here I present an enhanced PMF approach based on the new method that employs MALDI-TOF/TOF-MS of protein digest by LysN, and demonstrate to be useful for reliable identification, of either single protein or those in a mixture.

2 Chapter II: C-terminal Fragmentation of Peptides

2.1 Introduction

2.1.1 In-Source Decay (ISD) and Post-Source Decay (PSD)

In the MALDI, the excess energy transfers to the analyte during the ionization desorption process can lead to the ion fragmentation. In MALDI spectra, fragment ions are generated by different types of fragmentations, which are distinguished from the different places they occur.

In general, the types of fragmentation consist of in-source decay (ISD) and post-source decay (PSD). Moreover, ISD contains prompt fragmentation and fast fragmentation. ISD is a kind of fragmentation taking place in the source and can be separated to two steps by time scale. First, the fragmentation occurs is before or during the desorption is prompt fragmentation; secondly, fragmentation occurs in the source after the desorption but before the acceleration is fast fragmentation. PSD is a type of fragmentation that occurs in the field-free region after the acceleration region of the MALDI. It corresponds to the fragmentation of metastable ions. Metastable ions, as the name suggests, are not “stable”. They are stable enough to leave the source after ionization like the normal molecule ions, but due to the excess energy they received from laser pulse, they can fragment in the field free region of analyzer (Figure. 8).

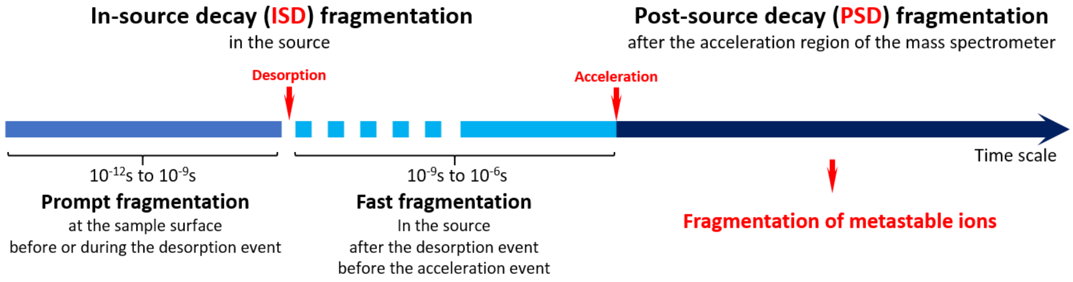


Figure. 8 Diagram of the fragmentation of MALDI.

Tandem mass spectrometry (MS/MS) is a type of spectrometer with additional analyzer(s) connect to the first analyzer to measure the fragment ions further. In the MALDI-TOF MS with a linear TOF analyzer, an extra reflectron time-of-flight instruments can be used to construct a MALDI-TOF-TOF MS/MS (Figure. 9). Fragment ions and neutral fragments can be fragmented by acquiring enough internal energy by ions.

Figure. 9 Simplest form of MS/MS

Since the ISD occurs before acceleration in MALDI, the ISD induced fragment ions (ISD fragment ions) have the same kinetic energy to the molecule ions (precursor ions) from the static electric field, thus they have the different velocities according to their m/z ratio. The different velocities allow both of the precursor ions and ISD fragment ions to be analyzed by linear TOF and reflectron TOF respectively. However, if a device named "timed ion selector" (TIS) is equipped, they cannot be analyzed in one experiment because they cannot pass the TIS in the same time.

On the other way, the velocity of the PSD induced fragment ions (PSD ions) and precursor ions are same because the PSD occurs after acceleration, and the kinetic energy of them are different. If the fragment ions have the same velocity to the precursor ions, the kinetic energy of the fragment ions is less than the precursors due to the less masses of the fragment ions. Thus they have the same flight time in TOF 1. Therefore, in TOF1, although the precursor ions and their PSD fragment ions have different kinetic energies, they can pass the TIS at the same time. Consequently, the precursor ions and their TOF 1 produced PSD fragment ions are reaccelerated in Ion Source 2, the kinetic energies of TOF 1 produced PSD fragment ions increase significantly to reach the kinetic energies of precursor ions, thus the velocity of the TOF 1 produced PSD fragment ions become faster than the precursors due to their lower masses. In the following field-free region (TOF 2), PSD occurs on precursor ions again, as well as in TOF 1. The TOF 2 produced PSD fragment ions have the same velocity as their precursor due to their lower kinetic energies and lower masses as mentioned above. The TOF 1 produced PSD fragment ions reach to the reflectron firstly, followed with the precursor ions and TOF 2 produced fragment ions. Since the precursor ions and TOF 1 produced PSD fragment ions are reaccelerated in Ion Source 2 together, they share a similar orbit in reflectron. However, the TOF 2 produced PSD fragment ions with less masses penetrate the reflectron shallower and spend less time than the precursor ions, thus the TOF 2 produced PSD fragment ions reached to the detector before the precursor ions. Because the velocities of TOF 1 produced PSD fragment ions are faster than the precursor ions, the TOF 1 produced PSD fragment ions reach to the detector before the precursor ions too (Figure 10). These ions reach the detector according to their *m/z* by different time of flight, and due to the reacceleration and refocusing of TOF 1 produced PSD fragment ions, the TOF 1 and TOF 2 produced PSD fragment ions perform as different type of peaks in the mass spectra.

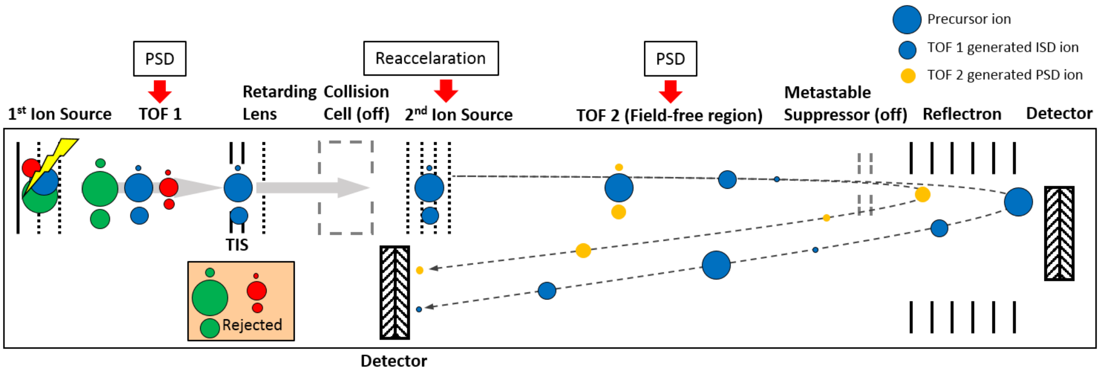


Figure. 10 Diagram of MALDI-TOF/TOF MS.

2.1.2 Collision Induced Dissociation (CID)

MS/MS requires the first analyzer to select fragment ions of precursor ions towards the second analyzer to measure the fragment ions. Collision Induced Dissociation (CID) is a technology to induce ion fragmentation in gas phase (Sleno, Volmer 2004, Mitchell Wells, McLuckey 2005). In a MALDI-TOF-TOF MS/MS, CID is usually equipped between TOF 1 and ion source 2 to increase the probability of ion fragmentation. The gas-phase precursor ions enter the CID cell which is full of collision gas (nitrogen, helium or argon) and then are activated by high electrical potential. Those ions collide with the neutral molecules of the collision gas and fragment to form small fragment ions. These accelerated fragment ions can then be analyzed by TOF 2 (Figure. 11).

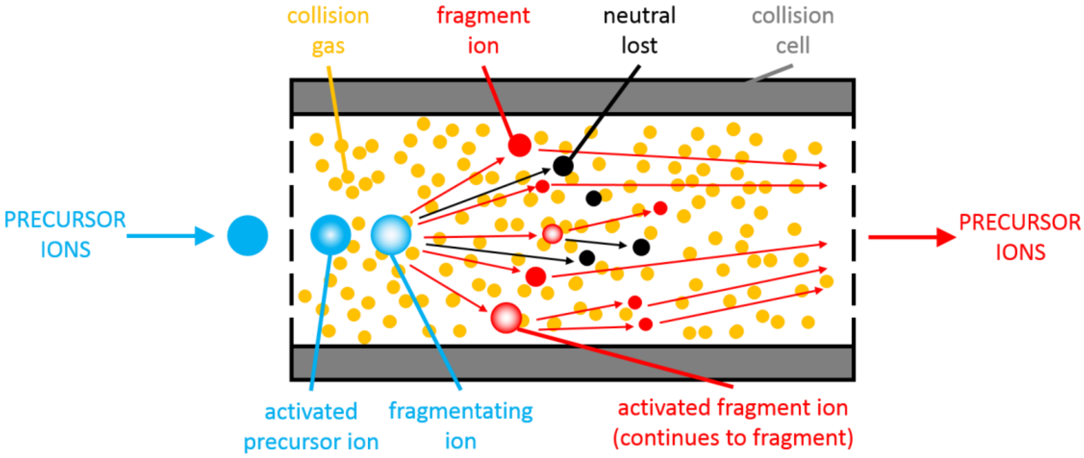


Figure. 11 Diagram of CID.

Figure. 12 shows the fragmentation specifies (the Roepstorff–Fohlmann–Biemann nomenclature) on the chemical structure of a peptide backbone by transmitting energy into the molecule (Roepstorff, Fohlman 1984, Biemann 1992). In the mass spectrometers for proteomics research, peptide fragmentation is induced by CID, and bond breakage mainly occurs at the amide bonds, where is the pathway with the lowest energy. If the charge retains on the N-terminal fragment, so called “b ions” are generated; if the charge retains on the C-terminal fragment, another kind of “y ion” are produced.

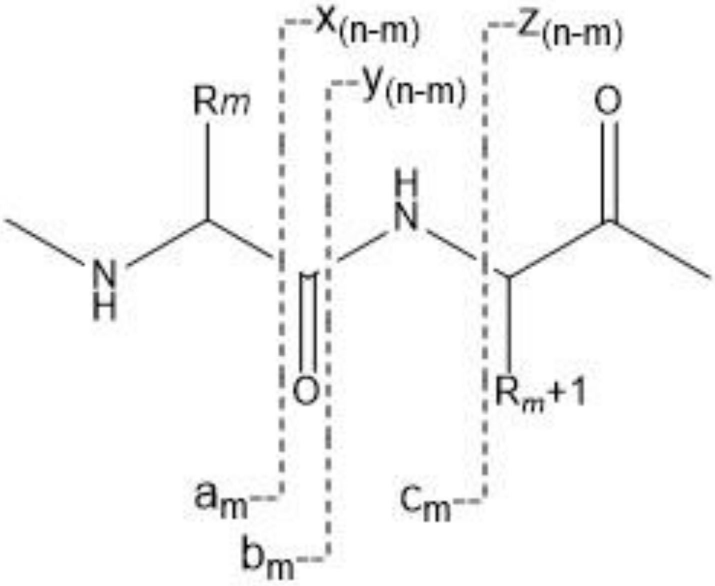


Fig. 12 Diagram of Roepstorff–Fohlmann–Biemann nomenclature.

2.1.3 C-terminal Rearrangement of Peptides.

Previous reports showed and illustrated a phenomenon of C-terminal rearrangement ions [b_{n-1}+H_2O] (n is the total number of amino acid residues in peptides) are frequently observed from internal Arg-containing peptides in PSD spectra (Takao et al. 1993, Gonzalez et al. 1996, Fang et al. 2000). (Renner, Spiteller 1988, Tang et al. 1988, Grese, Cerny & Gross 1989), Arg may play a similar role as the most basic amino acid residue. Such a C-terminal rearrangement could provide the information of the C-terminal amino acid, and a new idea for enhanced PMF originated from this phenomenon, thus through introducing the confirmed C-terminal amino acid sequences of peptides into the conventional PMF thereby improving the reliability of PMF method.

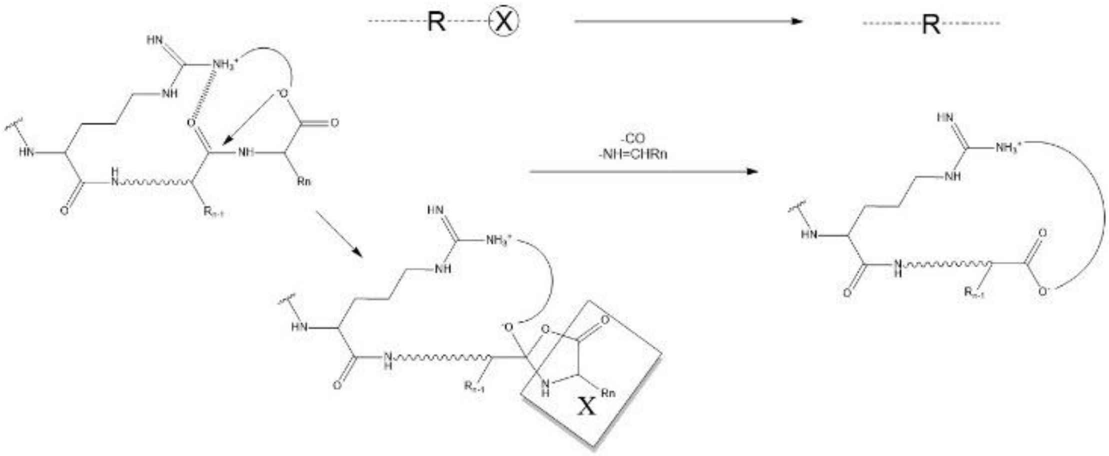


Figure. 13 Mechanism of C-terminal rearrangement of Arg-containing peptides.

2.2 Materials and Methods

2.2.1 Proteins and Chemicals

Bovine Serum Albumin (BSA), apo-Transferrin from bovine (TF), Cytochrome c (CYC) from equine heart, myoglobin from horse (MB), Myosin from rabbit (MYH), Phosphorylase b from rabbit (PGYM), Carbonic anhydrase from bovine (CA), ammonium bicarbonate, glycine, trifluoroacetic acid (TFA), LC/MS grade distilled water, and acetonitrile were purchased from Sigma-Aldrich (St. Louis, MO, USA). Synthetic peptides were purchased from GL Biochem (Shanghai, China). SDS-PAGE Molecular Weight Standards was purchased from Bio-Rad (Hercules, CA, USA). Sodium lauryl sulfate (SDS), sucrose and bromophenol blue were purchased from Nacalai Tesque (Kyoto, Japan). Tris base, dithiotheitol (DTT) and iodoacetic acid (IAA) were purchased from Wako (Osaka, Japan). LysN protease was purchased from Thermo Scientific (Rockford, IL, USA). Ultrapure water was supplied by a PURIC-MX II system (Organo, Tokyo, Japan).

2.2.2 Protein in Solution/Gel Digestion

10 mg protein was dissolved in 1mL 50mM NH_4HCO_3, 0.1% SDS solution (pH 8.0). Reduction of disulfide bonds was carried out by adding 20.41μL of 1M DTT

into protein solution to a final conc. of 20mM (1:50 dilution) and incubated at 37˚C for 45min. Alkylation was carried out by adding 42.52μL of 1M IAA into reduced protein solution to a final conc. of 40mM (1:25 dilute) and incubated at R.T. for15min in dark. The alkylated protein solution has been quenched by adding 10.74μL of 1M DTT to a final conc. of 10mM (1:100 dilution). 2μL of 0.5μ/μL LysN protease was applied to 8.5μL quenched protein solution (80μg) which diluted 10 times to conc. of 1μg/μL by adding 76.5μL 50mM NH_4HCO_3, 0.1% SDS solution, and incubated at 50 ˚C for 2h for digestion. The digests were stored in -20 ˚C to stop reaction, and desalted by Bond Elut-C18 (Agilent, Santa Clara, CA, USA). Purified peptides were dried by rotary vacuum micro-concentrator RMC-24 (KPI, Itami, Hyogo, Japan), and dissolved in 5% acetonitrile.

Gel bands were excised from the Coomassie blue-stained gel carefully, and prepared for in gel digestion. Gel pieces were washed three times with 50% MeOH/50 mM NH_4HCO_3 for 1h to overnight shanking at room temperature, removing the supernatant after each wash. Reduction of disulfide bonds was carried out by adding 0.5 mL of 10 mM DTT/100 mM NH_4HCO_3 into protein solution and incubated at 60˚C for 1h. Alkylation was carried out by adding 100 μL of 50 mM IAA/100 mM NH4HCO3 into reduced protein and incubated at R.T. for 30min in dark. Reduction and alkylation solution were removed after reaction. Alkylated gels were washed by water and ACN twice respectively. 30 μL of 1 μg LysN protease were added to a gel band, and incubated at 50 ˚C for 2h for digestion. 30 μL of ACN/0.1% TFA was added to the gel digestion to a final conc. of 50% ACN, following with a sonication for 10 min, then supernatant was collected. 100 μL of 50% ACN/0.1% TFA was added with a following 10 min sonication, supernatant was collected, this step was repeated twice.

2.2.3 nanoHPLC Separation

Analysis of the LysN digest was addressed by offline nanoHPLC/MALDI-TOF/TOF MS, a nanoHPLC system (Ultimate 3000; Thermo Scientific, Idstein, Germany) and an MALDI-TOF/TOF 4700 mass spectrometer (AB Sciex, Foster City, CA, USA) were utilized in the experiments. Chromatographic separation of the peptides was performed by loading 10μL sample onto a trapping column (μ-Precolumn holder, 5 mm, with connecting tubing, 30 μm internal diameter (i.d.); Dionex, Sunnyvale, CA, USA), and washed with 0.1% TFA at a flow rate of 20 μL/min for 25 min. The trapped peptides were then eluted into the analysis column (IntegraFrit C18, 3 μm, 100 Å, 75 μm i.d. × 100 mm; New Objective, Woburn, MA, USA), which had been equilibrated with 95% solvent A (5% ACN/95% H_2O containing 0.05% TFA). A solvent system contained solvent A and solvent B (100% ACN containing 0.5% TFA) mixed according to the linear gradient of 5–85% solvent B in 60 min, maintenance at 85% solvent B for 20 min, and linear gradient of 100–5% solvent B in 2 min were used for separation of peptides. The flow rate of the column was 200 nL/min. UV absorption at 220 nm and 280 nm was used for signal detection. 156 fractions of the effluent (each of 30 s) were spotted onto an Opti-TOF™ MALDI Plate System (192 spots; AB Sciex, Foster City, CA, USA) using a Probot microfraction collector (Dionex, Sunnyvale, CA, USA) from 20 min to 90 min of the run,.

2.2.4 MS/MS Measurement (MALDI-TOF/TOF)

The MALDI-TOF MS analyses were carried out on a 4700 MALDI TOF/TOF analyzer (Applied Biosystems, Framingham, MA) with a pulsed laser. The MS/MS was set in the positive mode and reflectron mode. The average number of laser shots was set at 800 on each sample well. The accelerating voltage in ion source 1 was 8 keV, and 15 keV in ion source 2. Metastable suppressor was set at “OFF”, and CID was “OFF” as well. The 4000 series Explorer

Software V3.6 was used to control the MS/MS measurement, and Data Explorer Software version 4.8 (Applied Biosystems, Framingham, MA) was used to process data. The matrixes for MALDI-TOF-TOF MS/MS were 5 mg/ml α-CHCA dissolved in 50% CAN containing 0.1% (v/v) TFA.

2.3 Results

2.3.1 Data Acquisition

It was a manual labor subjected by W.Y. First, the MS/MS was carried out according to *m/z* of precursor between 500 and 4000 by the 4000 series Explorer Software. Second, spectra data was convert to show on Data Explorer Software. On the spectrum, the metastable ion can be shown as a broad peak with low resolution, which could be easily differentiated from other ions. The corresponding fragment in-source ion was usually accompanied with a metastable ion peak, the *m/z* of which was around 3~4 Da smaller than that of the metastable ion. There were two type of metastable ions related which the C-terminal fragmentation: [b_{n-1}] and [$b_{n-1}+H_2O$] ions. If there was a C-terminal amino acid rearrangement occurred truly on a peptide, the C-terminal amino acid could be inferred from the difference of m/z between a precursor ion and in-source fragment ion (Figure. 14).

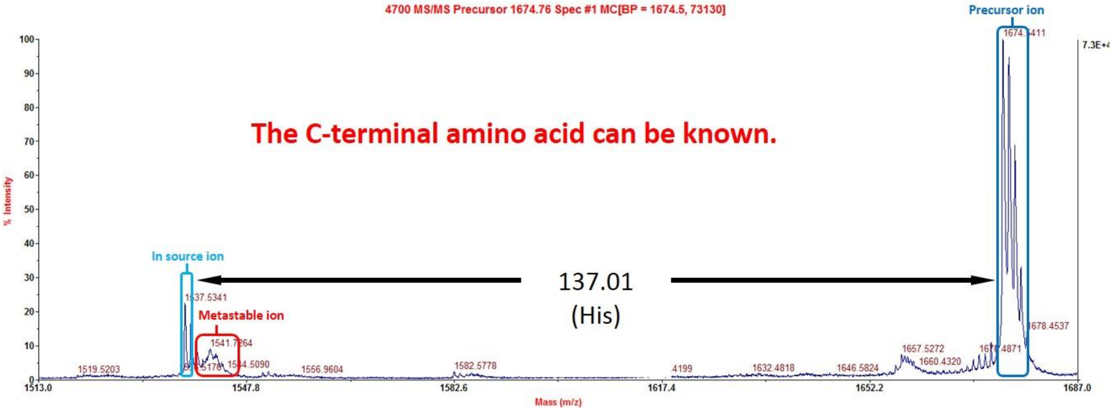


Figure. 14 C-terminal amino acid information of a peptide acquired with MS/MS.

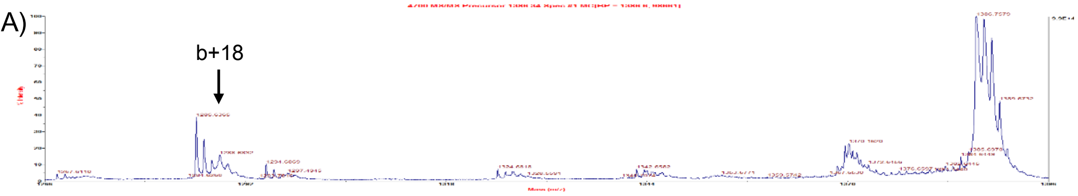
2.3.2 Effect of a Basic Amino Acid on the C-terminal Fragmentation

As described above, a salt bridge forms between the guanidino group of Arg and the C-terminal carboxylate in one peptide, then the C-terminal rearrangement occurs and the remained sequence can be shown as a broad metastable ion [b_{n-1}+H_2O] peak on the spectrum. Thereby it deduces the basic amino acid containing peptide to possess the capability to generate such C-terminal amino acid-truncated [b_{n-1}+H_2O] ion in MS/MS measurement. In order to demonstrate the contribution of basic amino acid to the C-terminal rearrangement of peptides, we compared the spectra from several synthetic peptides and natural peptides from protein digests (Table 1). The spectra indicate the generation of [b_{n-1}] (abbreviates to [b]) ions are general for all peptide sequences in the PSD spectra, except for [b_{n-1}+H_2O] (abbreviates to [b]) ions which could only be generated by Arg-containing peptide (not at the C-terminus). The Arg-containing peptides can produce [b+18] ions, as in the conclusion of the previous report, otherwise only [b] ions can be observed. Moreover, through replacing the Lys to Ala, both of the spectra of one basic amino acid (Lys) contained peptide and no basic amino acid contained peptides show [b] ion inferred to C-terminal fragmentation. These results indicate that the C-terminal fragmentation relative [b] metastable ions occur generally in any peptide sequences, and the [b+18] ions only occur in the Arg-containing peptides.

Table 1. Types of metastable ions relative to the truncation of C-terminal amino acid generated from different sequences of peptides.

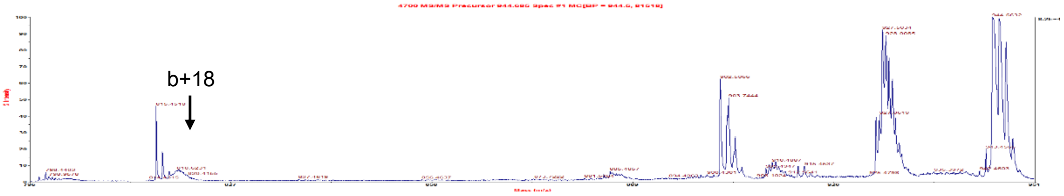
basic amino acid	R (+)	R (-)
metastable ion	b+18	b
sequences	KWCmcAIGHQERT AWCAIGHQERT YSRRHPE AQEPERNECFLSH	KWCAIGHQEAT KQEPEANECFLSH AAEFVEVT

(A)

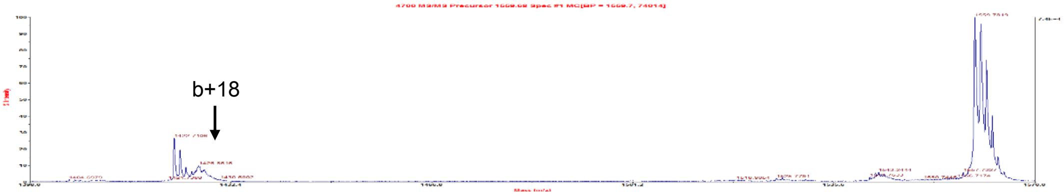


KWCmcAIGHQERT 1386.34 b+18/b

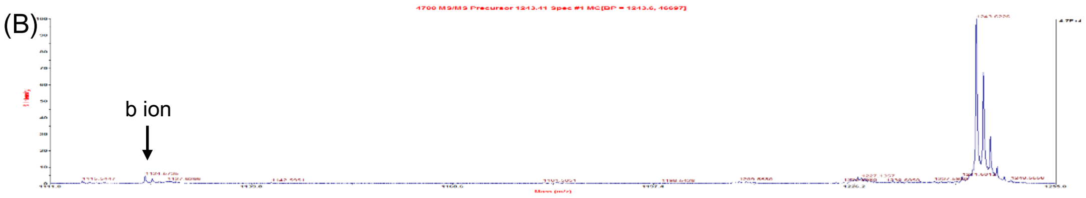
AWCAIGHQERT 1271.42 b+18/b



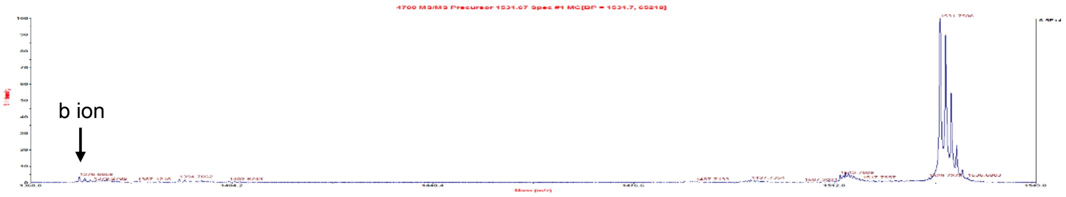
YSRRHPE 944.69 b+18/b



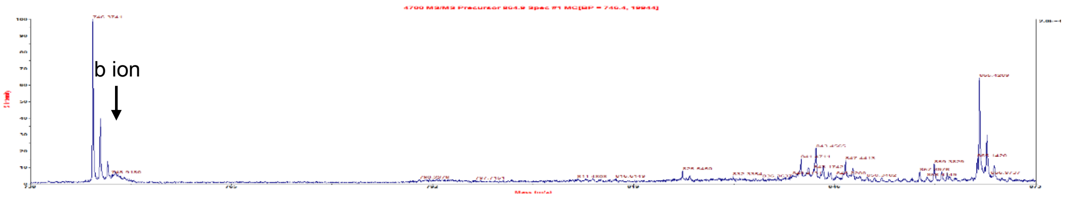
AQEPERNECFLSH 1559.68 b+18/b



KWCAIGHQEAT 1243.41 b



KQEPEANECFLSH 1531.67 b



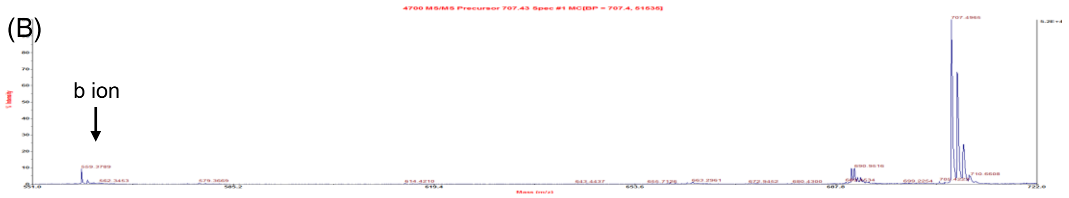
AAEFVEVT 864.90 b

Figure. 15 (A) C-terminal amino acid sequences relative b+18 metastable ions in the spectra of peptides Containing internal Arg. (B) C-terminal amino acid sequences relative b metastable ions in the spectra of peptides containing non-internal Arg.

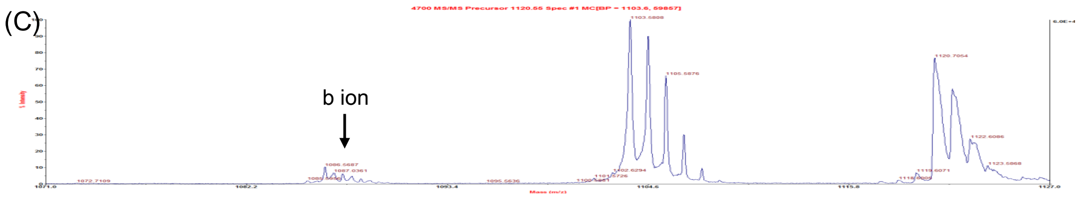
2.3.3 Effect of the C-terminal Carboxylate on the C-terminal Fragmentation

On the other hand, according to the previous report, the C-terminal carboxylate contributes to the rearrangement of C-terminus incorporation to Arg. Several peptides synthesized by replacing the C-terminal carboxyl group with an amide group were measured by MS/MS. Figure. 16 shows the spectra of peptides containing no internal Arg and peptide containing Arg with a C-terminal amide group, respectively. It indicates that the generation of [b] ions is not affected by C-terminal amide group, but the existence of Arg may suppress the generation of [b] ion under the condition without the existence of C-terminal amide group of a peptide.

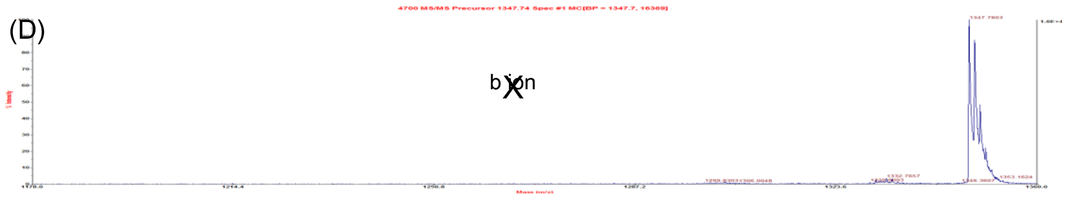
KHGTVVLTALGGIL-NH_2 1377.71 b



KFIGLM-NH_2 (Eledoisin related peptide) 707.43 b



GNHWAVGHLM-NH_2 (Neuromedin C) 1120.55 b



RPKPQQFFGLM-NH_2 (Substance P) 1347.74 no b

Figure. 16 MS/MS spectra of peptide with C-terminal amide group.

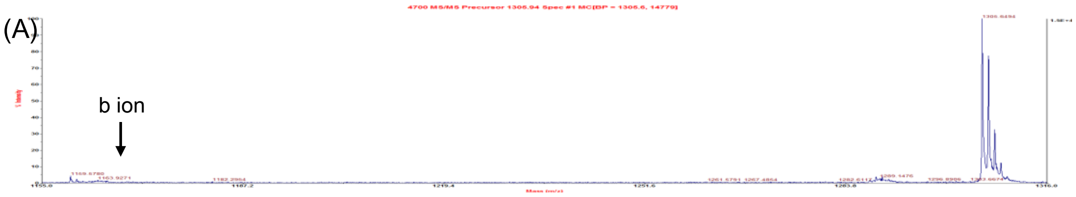
2.3.4 Protein Digest by Various Proteases

Table 2 shows C-terminal relative metastable ions in several peptides derived from BSA digested by trypsin, AspN, GluC and LysN respectively. In order to examine the dependency of the sequence on the C-terminal fragmentation, I prepared peptides with various sequences digested by different proteases, and applied to this analysis. According to the PSD spectra of those peptides (Figure. 15), [b] ions can be observed on most spectra except the peptides contain Arg (not at C-terminus), which show [b+18] ions on their spectra. It indicates that [b] ions occur generally among all the peptide sequences measured by MALDI-TOF/TOF MS, and only the Arg contained peptides (not at C-terminus) can generate [b+18] ions rather than b ions. In the spectra of peptides from trypsin and GluC digest, the known Lys, Arg or Glu can be observed. However, in the spectra of peptides from AspN and LysN digest, the C-terminal rearrangement can be measured and the amino acid at C-terminus can be inferred as well. On the other hand, the Lys or Asp at N-termini of the peptides from LysN or AspN digest are certain. Therefore, as to the peptides from proteins digested by LysN or AspN, three information of the sequences can be known after MALDI-TOF/TOF measurement: molecular mass, N-terminal amino acid and C-terminal amino acid. These findings can provide a new idea for protein identification by PMF, thus through introducing amino acids positioned at both N- and C-termini of peptides from LysN or ApsN digest, rather than the conventional utilizing of C-terminal Lys or Arg of peptides from trypsin digest only, can improve the reliability of PMF. Moreover, according to the characteristic of N-terminal rearrangement of AspN digested peptides can be observed on the PSD spectra (Figure. 15), tandem digestion by LysN and AspN are proposed to apply to this newly enhanced PMF method for increasing the reliability as well.

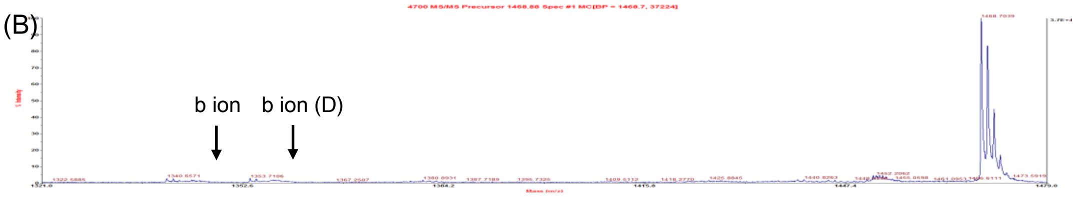
Table 2. Types of metastable ions generated from protein digested by different proteases.

protein	protease	miss	sequence	mass[M+H]	ion type
BSA	Trypsin	1	RHPYFYAPELLYYANK	2044.71	b+18
		1	QEPERNECmcFLSHK	1674.75	b+18
		1	LKECmcCmcDKPLLEK	1534.83	b
		0	HLVDEPQNLIK	1305.94	b
	AspN	0	DKPLLEKSHCmcIAEVEK	1896.95	b/D
		0	DESHAGCmcEKSLHTLFG	1788.72	b/D
		1	DKDVCmcKNYQEAK	1498.82	b/D
		0	DTHKSEIAHRFK	1468.88	b+18/D
		0	DVCmcKNYQEAK	1255.83	b/D
		1	DFAEDK	724.86	b/D
	GluC	1	RALKAWSVARLSQKFPKAEFVE	2560.40	b+18
		0	KKFWGKYLYE	1361.61	b
		1	IAHRFKDLGEE	1314.59	b+18
		0	KQIKKQTALVE	1285.76	b
		0	LCmcKVASLRE	1076.70	b+18
		0	YSRRHPE	944.69	b+18
	LysN	0	KVASLRETYGDMADCmcCmcE	2006.48	b+18
		1	KCmcCmcAADDKEACmcFAVEGP	1930.7	b

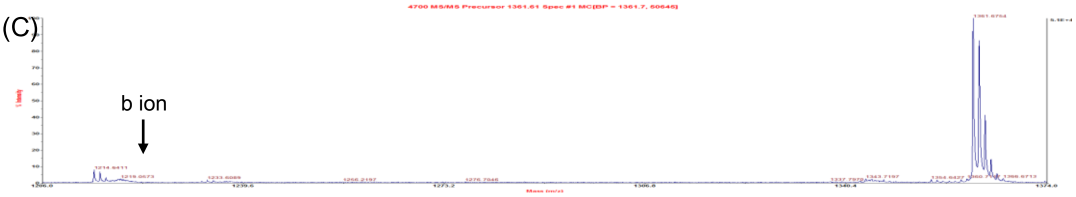
		1	KPDPNTLCmcDEFKADE	1779.63	b
		0	KQEPERNECmcFLSH	1764.61	b+18
		0	KTCmcVADESHAGCmcE	1465.52	b
		0	KYICmcDNQDTISS	1443.67	b
		1	KECmcCmcDKPLLE	1293.65	b
		0	KAWSVARLSQ	1145.67	b+18
		0	KLVVSTQTALA	1130.87	b
		0	KSHCmcIAEVE	1073.66	b
		0	KQNCmcDQFE	1069.64	b
		0	KAEFVEVT	922.75	b
		0	KATEEQL	818.78	b



HLVDEPQNLIK 1305.94 b



DTHKSEIAHRFK 1468.88 b/D



KKFWGKYLYE 1361.61 b

KQEPERNECmcFLSH 1673.74 b+18

Figure. 16 MS/MS spectra of four specific peptides from proteins digested by trypsin, AspN, GluC and LysN.

2.4 Discussion

2.4.1 b Ion and b+18 Ion

Previous report elucidated the mechanism of the rearrangement on C-terminal of Arg-containing peptides described above, and also verified by us in this project, which act as the basic theoretical fundament of this research. Since the rearrangement can induce the generation of metastable ions, which can be observed as a broad peak in the TOF-MS spectra, derived from the truncated peptide missing the C-terminal amino acid, this observation could be used as a new condition for protein identification by PMF. As we know, for the conventional PMF, trypsin is used commonly for protein digestion, and peptides with a C-terminal Lys or Arg can be generated. According to the mechanism previously reported, only peptides with internal Arg can rearrange at C-termini, trypsin digested peptides cannot. If there is a kind of protease which can cleave the N-terminus of Arg in proteins, the C-terminal amino acids of all peptides digested by such protease could be revealed via the rearrangement of the C-termini. Unfortunately, there is not such proteases at present, but another protease named LysN exists, which can create N-terminal Lys-containing peptides by protein digestion. According to the previously published report, we proposed the basic amino acid-containing peptide could produce the similar phenomenon, thus Lys may work as well as Arg for the C-terminal rearrangement of peptides. However, by our experiments, not all N-terminal Lys-containing peptides can produce [b+18] metastable ion on the TOF-MS spectra derived from C-terminal amino acid truncated peptides, except the internal Arg-containing ones. After a series of MS measurements of several synthetic peptides, I found that beside the [b+18] metastable ion generated by internal Arg-containing peptide, another kind of ion relative to C-terminal amino acid truncated peptide named [b] metastable ion can be observed from non-internal Arg-containing peptides, even in the peptides without any basic amino acid. On the other hand, we also synthesized peptides by replacing the carboxylate to amide group on the C-terminal amino acid of peptides. The MS spectra show that under the condition of there is not Arg in the peptide, [b] ion can be generated; but under the condition of there is Arg in the peptide, either [b] or [b+18] ion cannot be generated. Those results indicate Arg is not the necessary condition for the generation of b ion, as well as carboxylate; but Arg seems to suppress the generation of b ion if there is not a carboxylate.

2.4.2 Formation of a b ion adjacent to the C terminus

The most common PSD/CID fragment ion types are b and y ions (Spengler 1997). According to the mobile proton model, the proton(s) is usually localized at the most basic site in the peptide before activation, meanwhile it could be adducted to any carbonyl oxygen when there is no basic amino acid. (Wu, Lebrilla 1995, Paizs, Suhai 2004). The fragmentation mechanism involves carbonyl oxygen attack by its nucleophilicity to the neighboring amide carbonyl from the N- or C-terminal site; the carbonyl oxygen attack from the N-terminal site eventually cleaves the amide bond, leading to the proton transfer to the nitrogen atom (Fig. 18a), while the carbonyl oxygen attack from the C-terminal site does not (Fig. 18b) (Freitas, Hendrickson & Marshall 1999, Wyttenbach, Bowers 1999). Through the former pathway, the b ion, apart from the C-terminal peptide portion, could be formed as an N-protonated oxazolone (Chen, Turecek 2005). In case of a singly protonated molecule, the site of such fragmentation is highly dependent on the location of a proton over the amino acid sequence, which could be, most probably, N -amino group or any carbonyl oxygen of amide bond in the absence of basic amino acids. (Figure. 18).

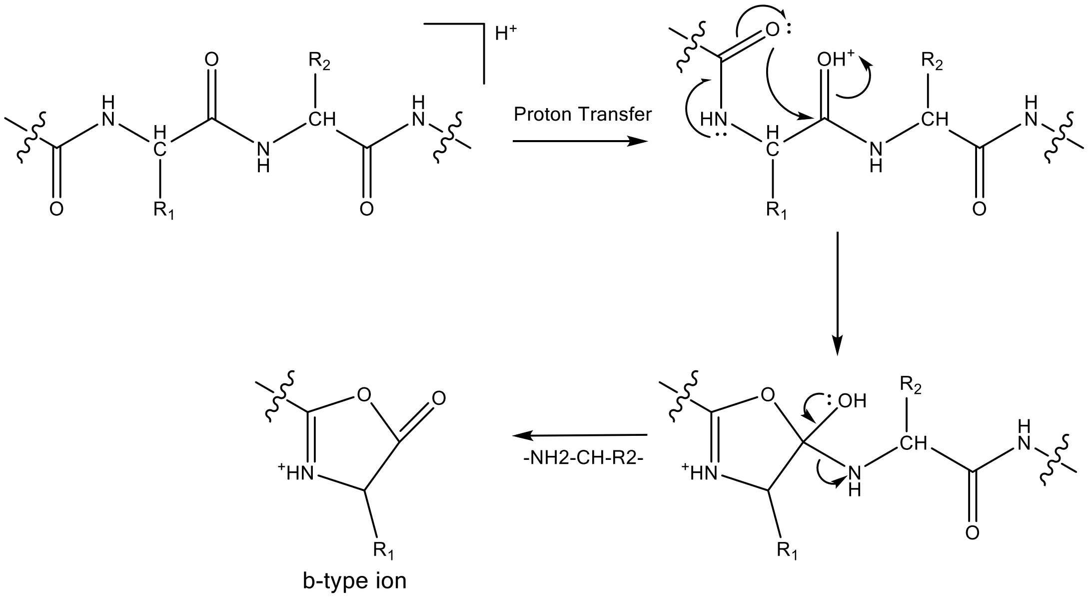


Figure. 18a Generation of b ion by common PSD/CID fragmentation.

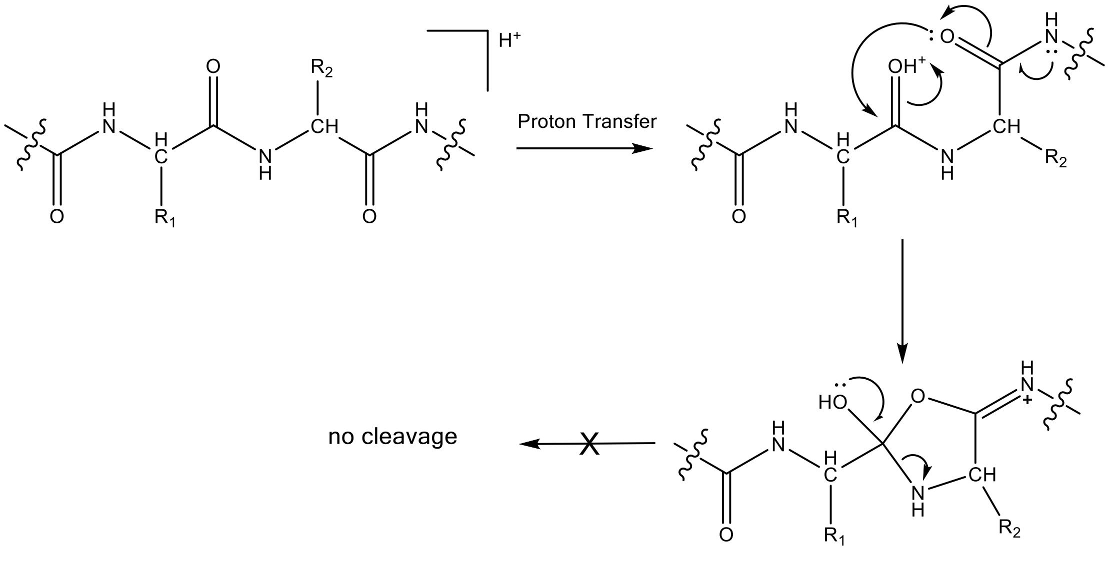


Figure. 18b No generation of b ion by proton transfer.

According to the PSD spectra obtained from MALDI-TOF/TOF MS/MS in the experiments described in this Chapter, the peptides without Arg in the sequence, but for the C terminus, gave the b-type metastable ions, which corresponded to the C-terminally-truncated peptides and were predominant among the metastable ions. The fragmentation at the last amide bond in gaseous phase could be accounted for by the two steps: the first step is decarboxylation at the C terminus; the second step is elimination of the terminal alkyl amine, followed by the formation of the b ion similar to the above fragmentation (Figure. 19). Unlike the formation of b ion at the internal amide bond in Fig. 18, there is a single pathway where the adjacent carbonyl oxygen anion attacks the carbonyl carbon, followed by elimination of the terminal $-NHCH_2(R_n)$ to form bn-1 ion. Thus, the b_{n-1} ion (C-terminally truncated peptide) could be preferentially generated than ordinary b ions in Fig. 18 during the flight in gaseous phase.

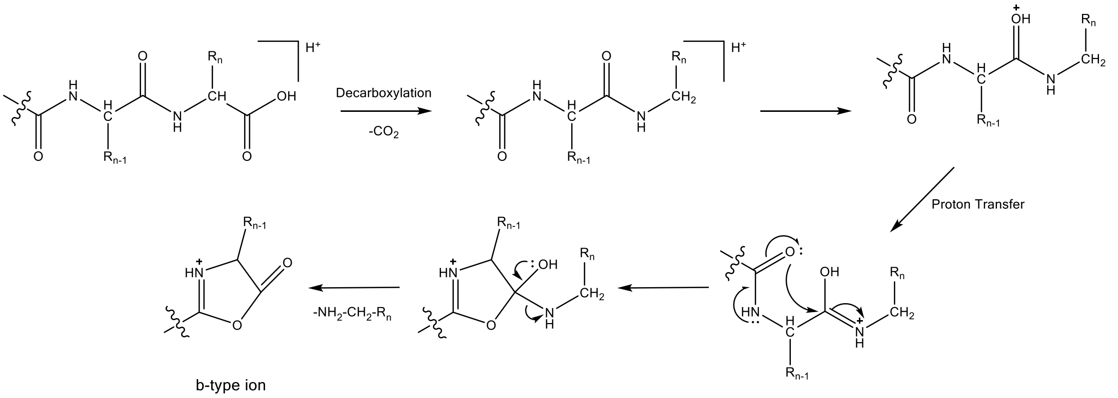


Figure. 19 Generation of C-terminally truncated bn-1 ion by PSD fragmentation.

3 Chapter III: Probability-Based Protein Identification by Searching Sequence Database Using Mass Spectrometry Data.

3.1 Introduction

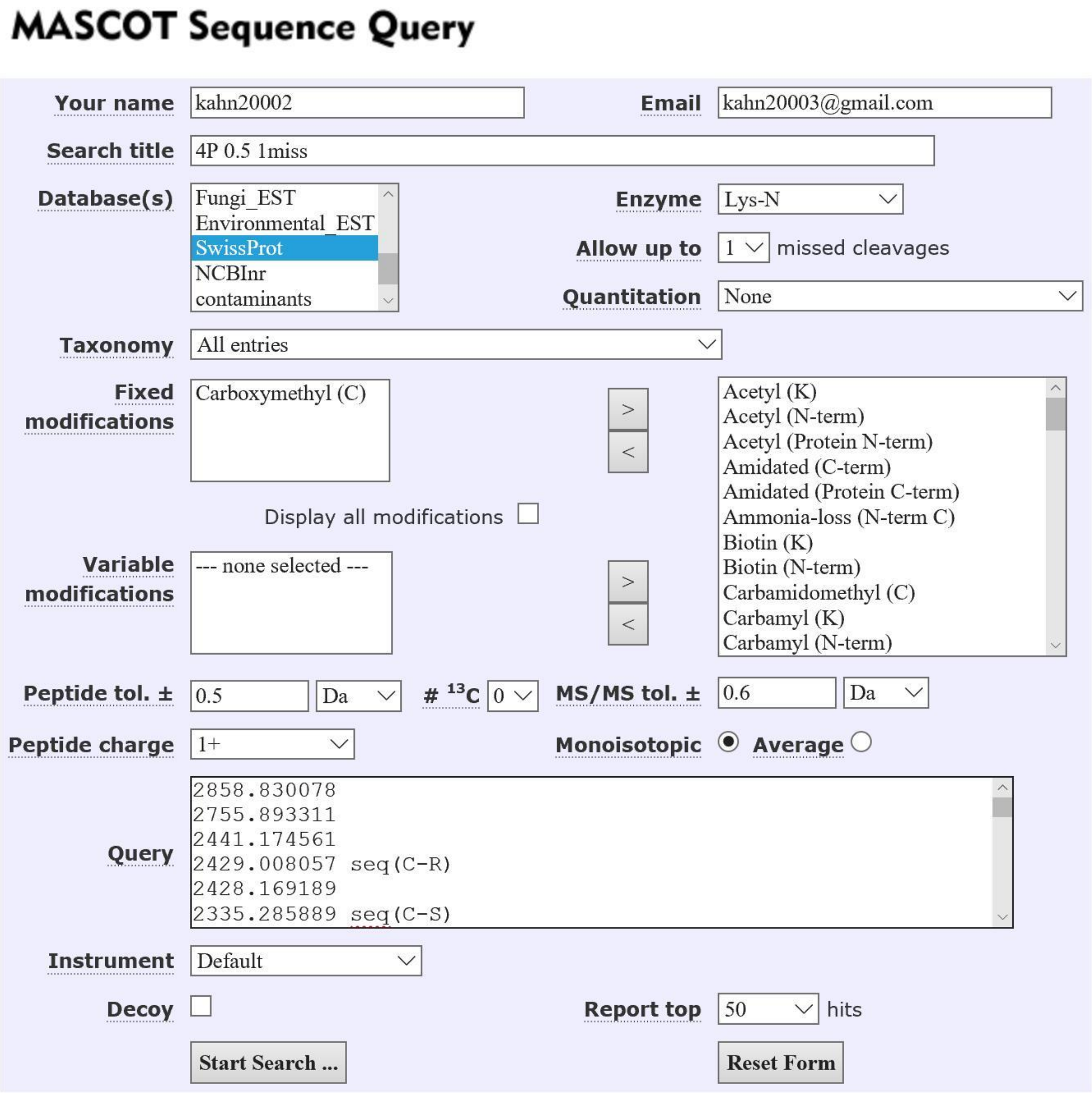
Protein identification is a necessary part in proteomics. A wide range of accurate, effective and efficient algorithms and computer programs for protein identification by searching a sequence database using MS data are developed in the last two decades. There are two main methods for computational identification: Mass-based approaches and Tag-based approaches. The former one uses molecular masses of peptides from a protein digestion by an enzyme, which is represented by PMF; on the other hand, MS/MS data from one or more peptides from the digestion of a protein are used by the latter approaches, just like *de novo* searching; moreover, still other approaches combine the data of mass and amino acid sequence (sequence query). Among all protein identification methods, the scoring system is the key. Experimental mass data obtained from the MS measurement of unknown protein is compared with theoretical mass data of known proteins in the database, and the score is used to evaluated how well the experimental and theoretical data match to each other. A threshold significance is set in the scoring system. A score above the threshold can be counted as a "hit", and the highest hit usually represents the identification result of the unknown protein. Moreover, “no hits” means there are no scores above the threshold and the protein cannot be identified. Shared-peak count is a basic scoring system for comparison, which the number of MS peaks shared by the experimental and theoretical spectra is counted. The experimental spectrum with the highest shared-peak number can be treated as the closest match to the theoretical one. Another basic scoring function is termed “coverage”. The coverage is the proportion of the unknown protein covered by matched peptides, which represent peaks in the MS spectrum. In addition, several general statistic methods such as cross correlation (Eng, McCormack & Yates 1994), Bayesian probability (Zhang, Chait 2000), expectation maximization (Nesvizhskii et al. 2003) and machine learning (Gay et al. 2002, Arnold et al. 2006) are applied to the scoring systems. One of the common computer programs named “Mascot” for protein identification will be introduced and applied as follow.

3.1.1 Mascot: a commercially available searching engine

Mascot is a search engine for protein identification involves probability-based scoring, which issued by Matrix Science Ltd., in 1999. It contains three types of search, such as peptide mass fingerprinting, sequence query, and MS/MS ion search. Mascot calculates the distribution of the lengths of tryptic peptide through the whole sequence database by using a scoring system based on the MOWSE algorithm, and the probability for every peak on the spectrum can be calculated. The probability-based scoring system was described to explain how Mascot interpret data generated by itself (Perkins et al. 1999): “*The fundamental approach is to calculate the probability that the observed match between the experimental data set and each sequence database entry is a chance event. The match with the lowest probability is reported as the best match. Whether this match is also a significant match depends on the size of the database. To take a simple example, the calculated probability of matching six out of ten peptide masses to a particular sequence might be* 10^{-5}*. This may sound like a promising result but, if the real database contains* 10^6 *sequences, several scores of this magnitude may be expected by chance. A widely used significance threshold is that the probability of the observed event occurring by chance is less than one in twenty (*$p<0.05$*). For a database of* 10^6 *entries, this would mean that significant matches were those with probabilities of less than* 5×10^{-8}*. The probability for a good match is usually a very small number, which* *must be expressed in scientific notation. This can be inconvenient, so we have adopted a convention often used in sequence similarity searches, and report a score which is -10Log*$_{10}$*(P), where P is the probability. This means that the best match is the one with the highest score, and a significant match is typically a score of the order of 70."* It indicates the smaller a protein database is, the lower scores for confidential level. This engine is available for users of the Matrix Science website.

3.2 Methods

Peptide masses data and C-terminal amino acid sequence data are acquired from the spectra according to Chapter II. Examples of the input window and output window are shown as Figure. 20 and Figure. 21.



MASCOT Sequence Query

Your name kahn20002

Email kahn20003@gmail.com

Search title 4P 0.5 1miss

Database(s) Fungi_EST, Environmental_EST, SwissProt, NCBInr, contaminants

Enzyme Lys-N

Allow up to 1 missed cleavages

Quantitation None

Taxonomy All entries

Fixed modifications Carboxymethyl (C)

Display all modifications ☐

Variable modifications --- none selected ---

Peptide tol. ± 0.5 Da

 # ^{13}C 0

MS/MS tol. ± 0.6 Da

Peptide charge 1+

Monoisotopic ● **Average** ○

Query

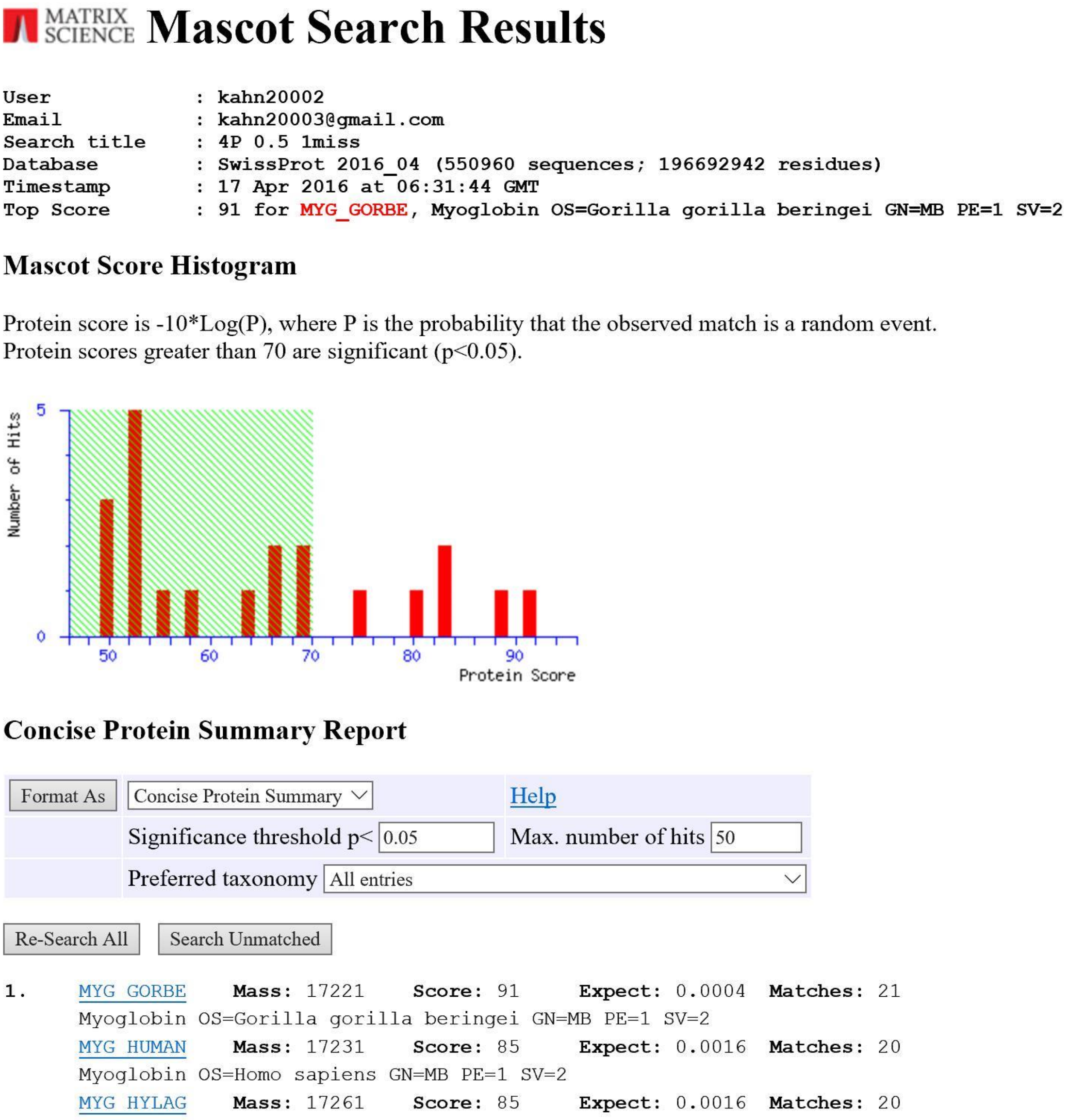
```
2858.830078
2755.893311
2441.174561
2429.008057 seq(C-R)
2428.169189
2335.285889 seq(C-S)
```

Instrument Default

Decoy ☐

Report top 50 hits

Figure. 20 Mascot input window.



Mascot Search Results

```
User           : kahn20002
Email          : kahn20003@gmail.com
Search title   : 4P 0.5 1miss
Database       : SwissProt 2016_04 (550960 sequences; 196692942 residues)
Timestamp      : 17 Apr 2016 at 06:31:44 GMT
Top Score      : 91 for MYG_GORBE, Myoglobin OS=Gorilla gorilla beringei GN=MB PE=1 SV=2
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 50
Preferred taxonomy All entries

```
1.    MYG_GORBE     Mass: 17221     Score: 91     Expect: 0.0004   Matches: 21
      Myoglobin OS=Gorilla gorilla beringei GN=MB PE=1 SV=2
      MYG_HUMAN     Mass: 17231     Score: 85     Expect: 0.0016   Matches: 20
      Myoglobin OS=Homo sapiens GN=MB PE=1 SV=2
      MYG_HYLAG     Mass: 17261     Score: 85     Expect: 0.0016   Matches: 20
```

Figure. 21 Mascot output window.

3.3 Results

3.3.1 Protein identification (Mascot) based on masses (PMF).

Table. 3 shows the Mascot protein identification by peptide masses obtained for seven single proteins. According to the output, the scores of BSA is 46, CYC is 43, MB is 29, CA is 48, and there are no scores of TF, MYH and PYGM. Input data and output data refer to the supplement.

Table. 3 Results of Mascot single protein identification by masses data of all peptides.

	BSA	TF	CYC	MB	MYH	PYGM	CA
Score	46	/	43	29	/	/	48
Number of false matched proteins	22	45	43	40	40	37	35

Table. 4 shows the Mascot protein identification by peptide masses obtained for a mixture of four proteins. According to the output, the scores of MB is 27 and there are no scores of BSA, TF and CYC. Input data and output data refer to the supplement.

Table. 4 Results of Mascot protein mixture identification by masses data of all peptides.

	BSA	TF	CYC	MB
Score	/	/	/	27
Number of false matched proteins	42			

3.3.2 Protein identification (Mascot) based on masses and C-terminal amino acid sequences of partial detected peptides.

Table. 5 shows the Mascot protein identification by masses of all detected peptides, which are the seme as those used in 3.3.1 and C-terminal amino acid sequences of partial peptides derived from seven single proteins. According to the output, the scores of BSA is 293, TF is 138, CYC is 137, MB is 133, MYH is 134, PYGM is 124 and CA is 126. Input data and output data refer to the supplement.

Table. 5 Results of Mascot single protein identification by masses data of all peptides and C-terminal amino acid sequences of partial peptides.

	BSA	TF	CYC	MB	MYH	PYGM	CA
Score	293	138	137	133	134	124	126
Number of false matched proteins	45	28	31	15	46	37	44

Table. 6 shows the Mascot protein identification by masses data of all peptides and C-terminal amino acid sequences of partial peptides derived from a mixture of four proteins. According to the output, the scores of BSA is 64, TF is 48, CYC is 55, MB is 65 Input data and output data refer to the supplement.

Table. 6 Results of Mascot protein mixture identification by masses data of all peptides and C-terminal amino acid sequences of partial peptides.

	BSA	TF	CYC	MB
Score	64	48	55	65
Number of false matched proteins	16			

3.3.3 Protein identification (Mascot) by masses and C-terminal amino acid sequences of partial detected peptides (excluding the peptides with mass values only).

Table. 7 shows the Mascot protein identification by masses and C-terminal amino acids of partial detected peptides (excluding the peptides with mass values only) derived from seven single proteins. According to the output, the scores of BSA is 364, TF is 175, CYC is 176, MB is 201, MYH is 142, PYGM is 157 and CA is 142. Input data and output data refer to the supplement.

Table. 7 Results of Mascot single protein identification by masses data and C-terminal amino acids of partial detected peptides (excluding the peptides with mass values only)

	BSA	TF	CYC	MB	MYH	PYGM	CA
Score	364	175	176	201	142	157	142
Number of false matched proteins	36	31	11	2	26	38	37

Table. 8 shows the Mascot protein identification by masses data and C-terminal amino acids of partial detected peptides (excluding the peptides with mass values only) derived from a mixture of four proteins. According to the output, the scores of BSA is 61, CYC is 66 and MB is 84, but no score of TF. Input data and output data refer to the supplement.

Table. 8 Results of Mascot protein mixture identification by masses data and C-terminal amino acids of partial detected peptides (excluding the peptides with mass values only).

	BSA	TF	CYC	MB
Score	61	/	66	84
Number of false matched proteins	1			

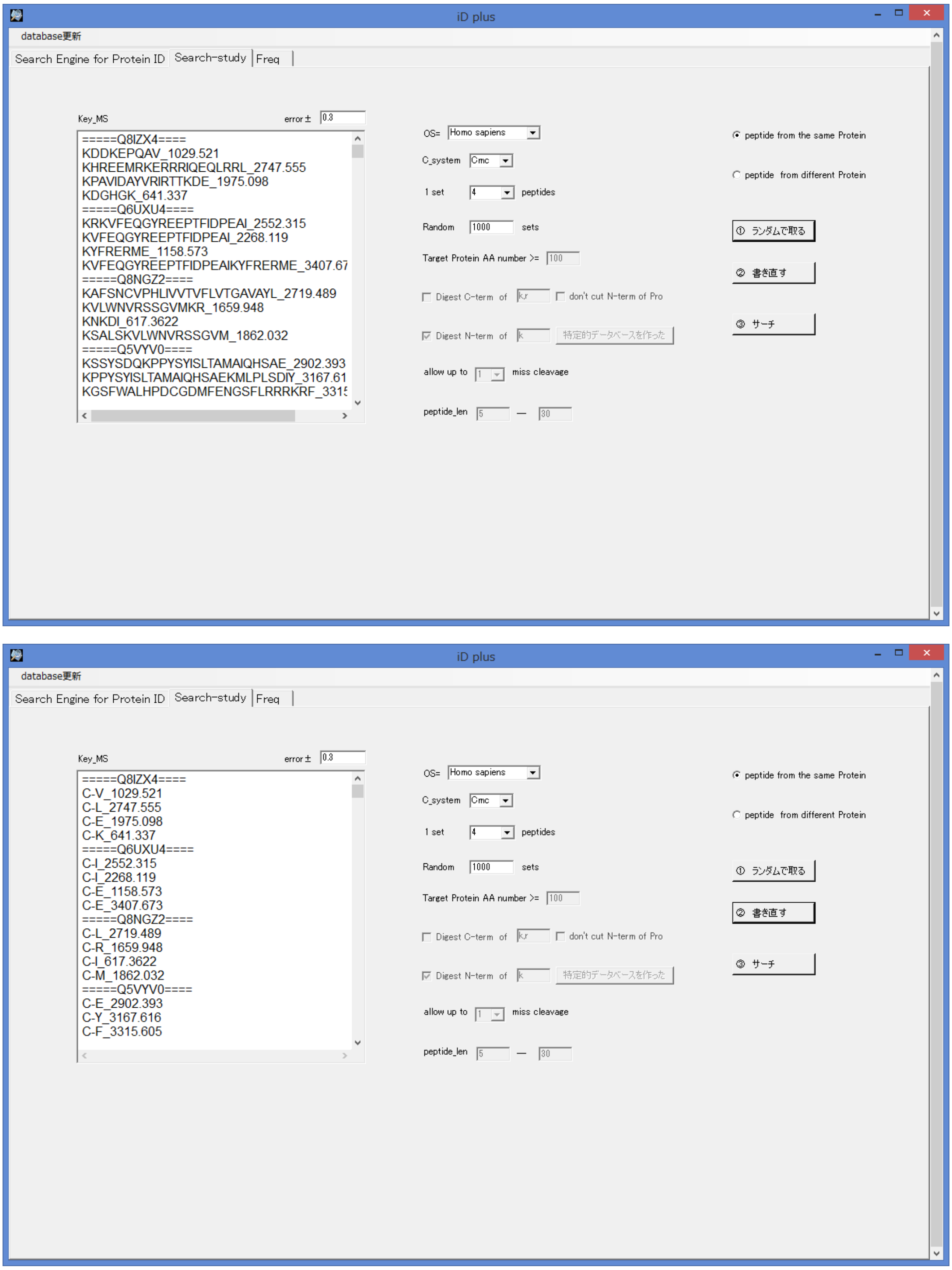
3.4 Discussion

According to the above results of single protein identification, the conventional PMF by searching peptide masses could hardly be in use for protein identification. As to the single protein identification, only four proteins can be identified with scores lower than confidential threshold (for example at 70 when the size of a database is 10^6 sequences) of seven proteins, and other three ones cannot be identified. However, after inputting the additional C-terminal amino acid sequences of the peptides partially observed together with masses, all of the seven proteins can be identified with significant scores above 70, which indicates the additional C-terminal amino acid sequence can increase the overall probability for a correct match. Moreover, after removing the peptides with masses data only from the input data, the scores of all seven identified protein increase higher than the second searches, it means that reducing the size of input data can decrease the opportunity of random matches to the theoretical data in the database. As well as the single protein identification, the experiments for protein mixture case also demonstrated the effectiveness of additional C-terminal amino acid sequences information in improving the conventional PMF: all of the four proteins in a mixture could be identified by peptides with masses and C-terminal amino acid sequences, meanwhile almost no protein could be identified by conventional PMF. Especially, after adding the C-terminal amino acid sequences of partially observed peptides into the input data, three proteins in a mixture contained four proteins except TF. However, TF could be identified by searching the masses and C-terminal amino acid sequences of partial observed peptides only. It is a significant improvement than the above mentioned case for the single protein digestion, not only reducing the random match by decreasing the amount of input data, but also the unknown protein can be identified. According to the comparison of the number of matches between the search of protein mixture by masses of all detected peptides and C-terminal amino acid sequences of partially detected peptides, and the search of protein mixture by masses and C-terminal amino acid sequences of partially detected peptides (shown in Supplement “4P mass/mass+c” and “4P mass +c”), the number of matches for MYG_HORSE, ALBU_BOVIN and CYC_HORSE decreases from 23 to 7, 31 to 8 and 20 to 5, respectively. Meanwhile, the kinds of protein identified also decreases from 21 to 7. Such a reduction of matched peptides to each protein and the species of identified protein offers a possibility of reducing the interference by noisy random matches, and increasing the accuracy of identification. It indicates an advantage of dealing with a mixture of proteins by introducing C-terminal amino acid sequences in conventional PMF.

4 Chapter IV: Non-Probability-Based Protein Identification by Searching Sequence Database Using Mass Spectrometry Data I: Computer Simulation.

4.1 Introduction of the algorithm for computer simulation of iD plus.

A non-probability-based sequence database searching software named “iD plus” was developed by our lab. There are two parts in the computational simulation: peptides from “Same Protein” (SP) and “Different Proteins” (DP) (Figure. 22). In the workflow of “SP” (Figure. 23 (A)), a protein database of LysN digested peptides was established. 1000 proteins were selected from the database randomly, and then four peptides were selected from each protein randomly in the 1000 proteins. The mass of a peptide, and both of the N-terminal (K) and C-terminal amino acids in the sequence of peptide were defined as a “set”, and every four sets from a same protein were arranged as a “combination”, 1000 combinations among all combinations were selected randomly and input into the database searching. In the results of database searching, if there was only one hit corresponded to a combination (4 peptides set), the result was denoted as “true”; if there was more than one hit corresponded to a combination, the result was denoted as “false”. All of the results were merged then output as a form with protein IDs for analysis. Figure 23. (B) shows the workflow of “DP”. 1000 proteins were generated randomly selected from the protein database and the LysN digested peptides were randomly generated, and then one peptide was selected from each of 1000 proteins in a random manner. *1000!/4!(1000-4)!* combinations were subjected to the database searching, if there was no hit corresponding to any of the combinations, the result was denoted as “true”; if there were hits corresponding to any of the combinations, the result was denoted as “false”. All of the results were merged then output as a form with protein IDs for analysis.



Key_MS error ± 0.3

```
=====Q8IZX4====
KDDKEPQAV_1029.521
KHREEMRKERRRIQEQLRRL_2747.555
KPAVIDAYVRIRTTKDE_1975.098
KDGHGK_641.337
=====Q6UXU4====
KRKVFEQGYREEPTFIDPEAI_2552.315
KVFEQGYREEPTFIDPEAI_2268.119
KYFRERME_1158.573
KVFEQGYREEPTFIDPEAIKYFRERME_3407.67
=====Q8NGZ2====
KAFSNCVPHLIVVTVFLVTGAVAYL_2719.489
KVLWNVRSSGVMKR_1659.948
KNKDI_617.3622
KSALSKVLWNVRSSGVM_1862.032
=====Q5VYV0====
KSSYSDQKPPYSYISLTAMAIQHSAE_2902.393
KPPYSYISLTAMAIQHSAEKMLPLSDIY_3167.61
KGSFWALHPDCGDMFENGSFLRRRKRF_331
```

OS= Homo sapiens
C_system Cmc
1 set 4 peptides
Random 1000 sets
Target Protein AA number >= 100
☐ Digest C-term of k,r ☐ don't cut N-term of Pro
☑ Digest N-term of k
allow up to 1 miss cleavage
peptide_len 5 — 30
● peptide from the same Protein
○ peptide from different Protein

Key_MS error ± 0.3

```
=====Q8IZX4====
C-V_1029.521
C-L_2747.555
C-E_1975.098
C-K_641.337
=====Q6UXU4====
C-I_2552.315
C-I_2268.119
C-E_1158.573
C-E_3407.673
=====Q8NGZ2====
C-L_2719.489
C-R_1659.948
C-I_617.3622
C-M_1862.032
=====Q5VYV0====
C-E_2902.393
C-Y_3167.616
C-F_3315.605
```

OS= Homo sapiens
C_system Cmc
1 set 4 peptides
Random 1000 sets
Target Protein AA number >= 100
☐ Digest C-term of k,r ☐ don't cut N-term of Pro
☑ Digest N-term of k
allow up to 1 miss cleavage
peptide_len 5 — 30
● peptide from the same Protein
○ peptide from different Protein

Figure. 22 Windows of iD plus for comuplational simulation. Above: random selection. Bottom: format change.

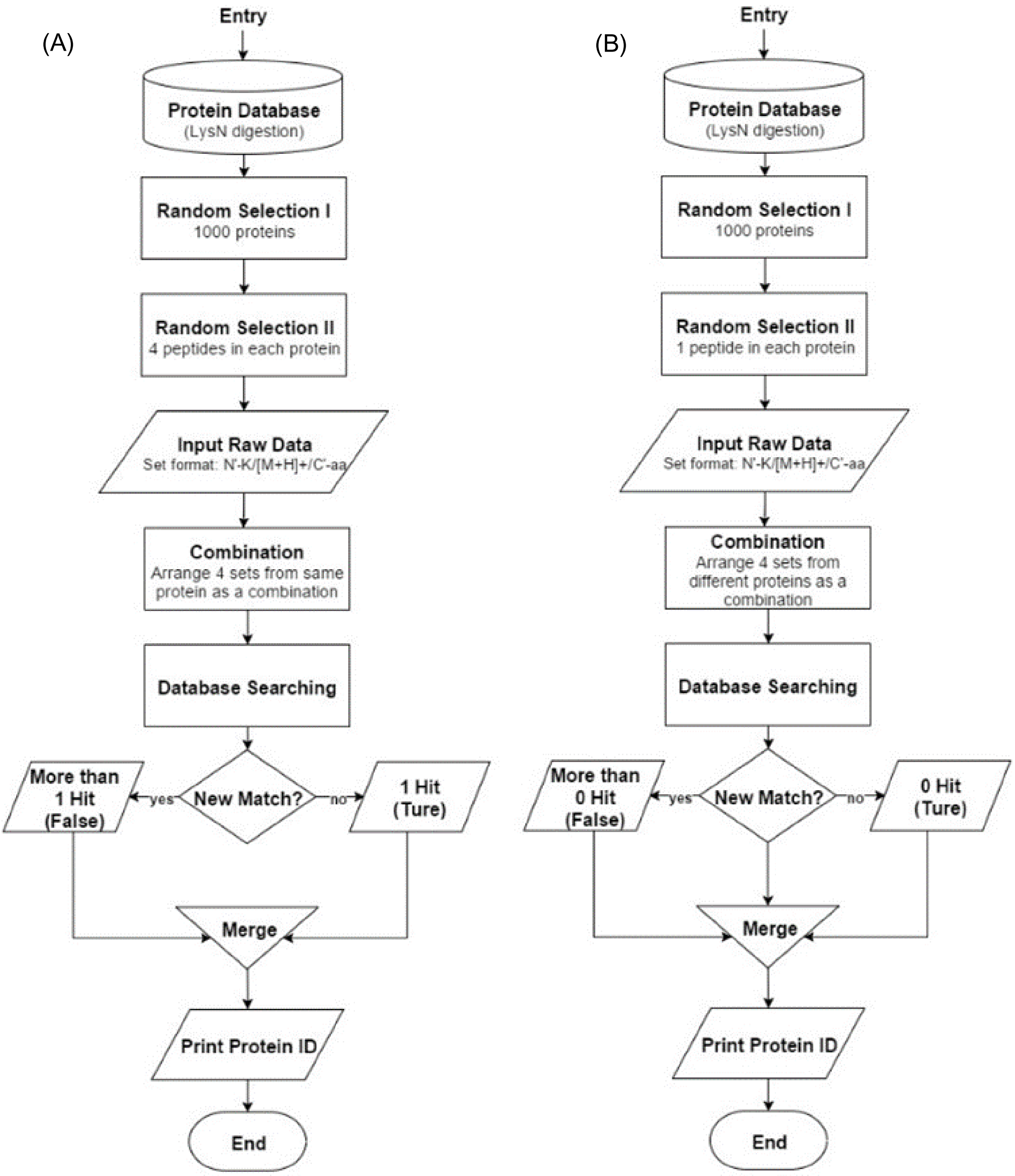


Figure. 23 Workflows of virtual searching of 1000 combinations of four sets of peptide masses and C-terminal amino acid sequences. (A) module for searching based on four sets from “Same Protein”. (B) module for searching based on four sets, each from “Different Proteins”.

4.2 Methods

Peptide masses data and C-terminal amino acid sequence data are acquired from the spectra according to Chapter II. Examples of the input data format and results format are shown in the part of supplement.

4.3 Results

Figure. 24 (A-1 & A-2) shows the comparison of searching reliability of peptides with or without C-terminal amino acid in both of “SP” and “DP” digested by LysN *in silico*. In the “SP”, there is around 60% of 11,000 combinations (three peptides) show false positive hits without C-terminal amino acid, whereas, only 0.09% of 11,000 combinations (three peptides) show false positive hits with C-terminal amino acid. “DP” shows a similar result as well. There is around 50% of 10,000 combinations (three peptides) show false positive hits, whereas only 0.04% of 10,000 combinations (three peptides). Figure. 24 (B-1 & B-2) shows the comparison of searching reliability by a combination contains different numbers of peptides in both of “SP” and “DP” digested by LysN *in silico*. In the “SP”, the rates of false positive hits resulted from searching by over 10,000 combinations contain one, two, three and four peptides are around 100%, 17-20%, 0.09% and 0%, respectively. In the “DP”, the rates of false positive hits resulted from searching by over 10,000 combinations contain one, two, three and four peptides are around 100%, 3-6%, 0.04% and 0%, respectively. These results clearly indicate the introduction of an additional C-terminal amino acid sequence is critically useful for increasing the reliability of PMF than using only one terminal amino acid information, which is deduced by the enzyme used. The reliability increases over around 100% by setting double-termini for searching than single-terminus for searching.

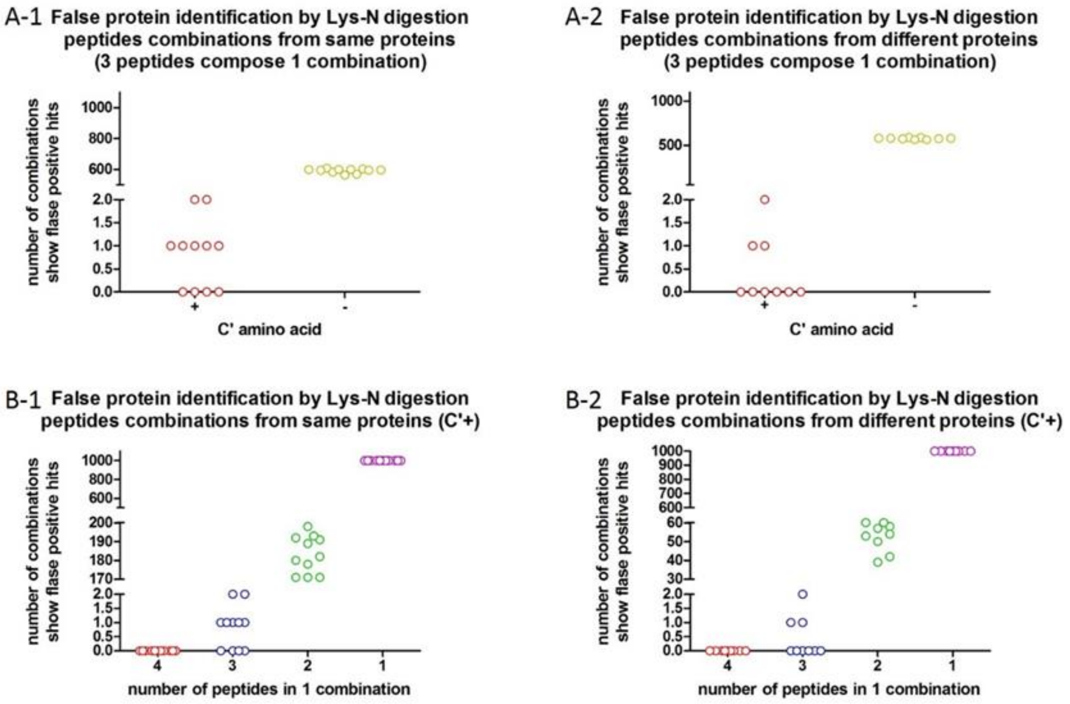


Figure. 24 Results of database searching using virtual LysN peptides. A-1: The number of false hits by LysN peptides derived from same proteins under the conditions with or without C-terminal amino acid; A-2: The number of false hits by LysN peptides derived from different proteins under the conditions with or without C-terminal amino acid; B-1: The number of false hits by different numbers of LysN peptides derived from same proteins with C-terminal amino acid sequences; B-2: The number of false hits by peptides derived from different proteins with C-terminal amino acid sequence. Each circle represents the result of 1000 combinations of database searching of peptide sets of which are generated in a random selection.

4.4 Discussion

Almost all of the protein identification performed by MS are based on computational database search software, for example the Mascot I described in Chapter III above. Mascot is a probability-based protein identification computer program by searching sequence database using MS data. The probability-based algorithm involves calculating the theoretical fragments for all the peptides in the database. The disadvantages of probability-based identification are relative low reliability and it is difficult to identify proteins mixture. Different with Mascot, the in-house computer program “iD plus” developed in our lab is not a probability based database search software. According to the results of computational simulation, we believe that if four C-terminal truncated peptides from a protein digested by LysN protease, the confident level is at about almost 100%. Such a high confident level can be shown in both of identification for single protein and protein mixture. Moreover, even one peptide is enough to use for protein identification by MS/MS sequencing, while PMF usually requires a dozen of peptide masses for using. The number of observed peptides in mass spectra relative to the amount of protein sample, the larger amount of unknown protein, the more number of peptides derived from the digest can be obtained. So the ability to use fewer peptides for protein identification is important for the sensitivity. The simulation verify at least four peptides are enough for a positive identification, the number is much less than conventional PMF. The examples of experimental practice by iD plus searching would be described in the next chapter and be discussed further.

5 Chapter V: Non-Probability-Based Protein Identification by Searching Sequence Database Using Mass Spectrometry Data II: Experimental Practice.

5.1 Introduction of the algorithm for experimental practice of iD plus.

Fig. 25 shows the workflow of protein identification based on the peptide masses and C-terminal amino acid sequences of the observed peptides as well as the N-terminal Lys, which was set as the default from the enzyme specificity. The analysis process as divided into four parts: “Combination I” (C1), “Database Searching I” (DS1), “Combination II” (C2) and “Database Searching II” (DS2). In the progress of “C1”, based on the total number *n* of peptides, the combinations of four sets of peptide masses and their C-terminal amino acid sequences were generated $n!/4!(n-4)!$ combinations. Proteins matched to any combinations of four sets were searched in the progress of “DS1”. If any combination could match to one protein, the result was denoted as “true”, then saved in a tile. If any combination could not match to any protein, the sets in these combinations, *m* (where *m* refers to the number of remaining peptides after “DS1”) sets were arranged to generate $m!/3!(m-3)!$ Combinations (“C2”), which comprised of three sets of peptide masses and C-terminal amino acid sequences. Proteins matched to any combinations were searched in the progress of “DS2”. If any combination could match to one protein, the result was denoted as “semi-true”, then added into the “true” results of “DS1”; the remaining combinations of three sets that could not match with any proteins were discarded. Regarding to the merged “true” and “semi-true” results, sequence coverages, which are the percentage of the number of amino acids of the peptides occupied the total number of amino acids of the identified protein were calculated, and used for judgement of the reliability of merged results, in case of multiple hits being obtained for a combination. Finally, protein IDs were output as identification result with the matched peptide masses and C-terminal amino acid sequences.

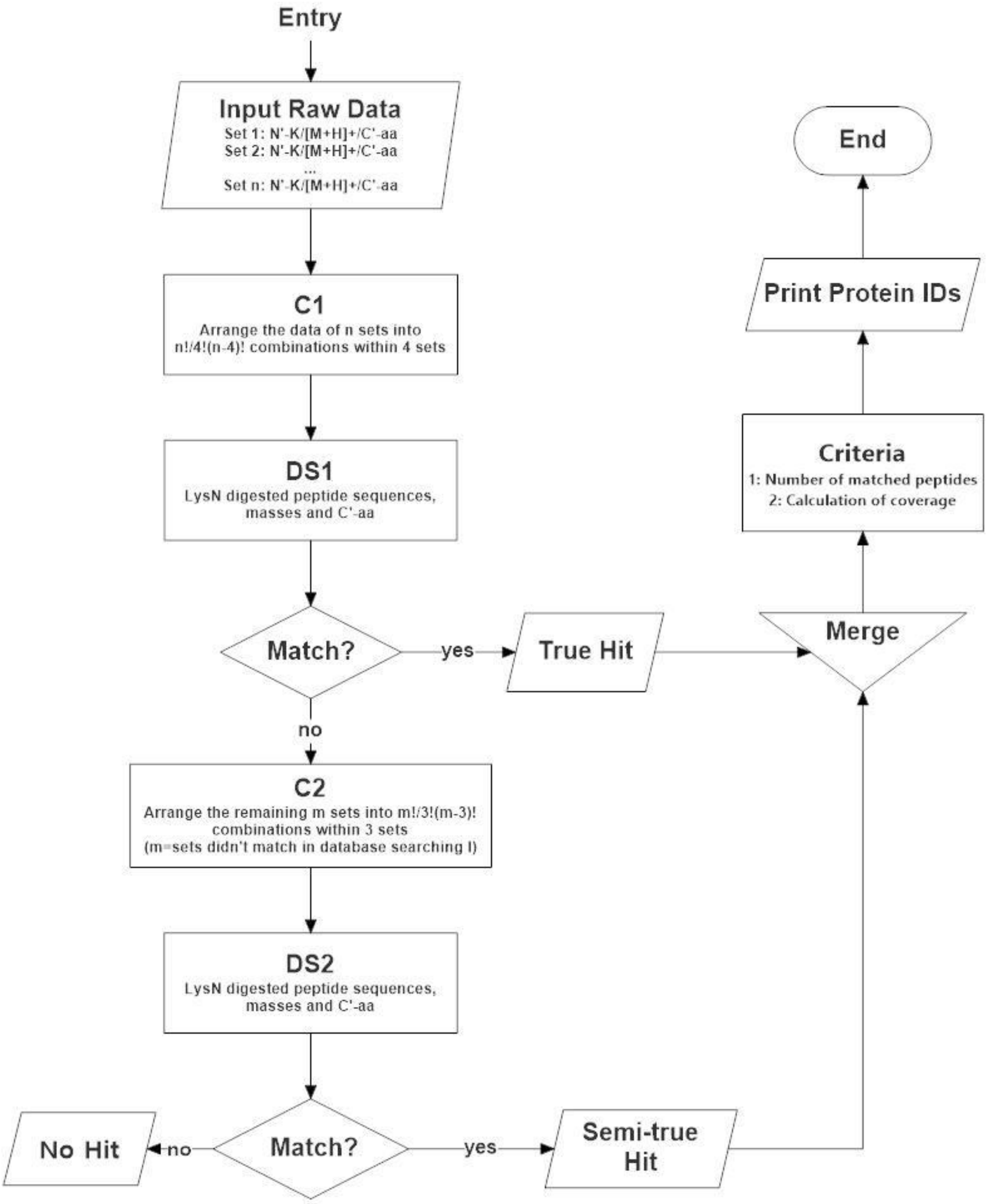
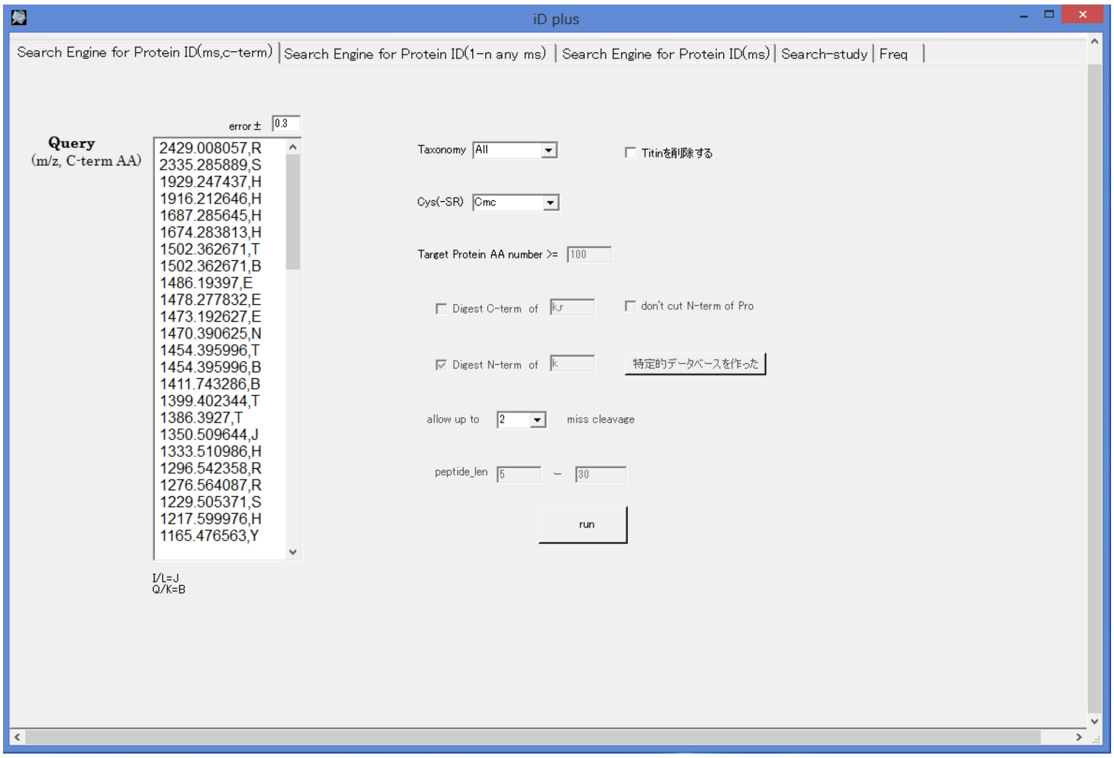


Fig. 25 Database searching workflow for protein identification by using in-house software “iD plus”.

5.2 Methods

Peptide masses data (above Da) and C-terminal amino acid sequence data are acquired from the spectra according to Chapter II. Fig. 26 show the input window of iD plus. Examples of the input data format and results format are shown in the part of supplement. The database searching by iD plus was under the condition of allowing 0.5 Da error and 1 missed cleavage in the matched peptide sequences, and removing the noisy proteins titin (protein IDs of A2ASS6 and Q8WZ42).



error ± 0.3

Query
(m/z, C-term AA)

```
2429.008057,R
2335.285889,S
1929.247437,H
1916.212646,H
1687.285645,H
1674.283813,H
1502.362671,T
1502.362671,B
1486.19397,E
1478.277832,E
1473.192627,E
1470.390625,N
1454.395996,T
1454.395996,B
1411.743286,B
1399.402344,T
1386.3927,T
1350.509644,J
1333.510986,H
1296.542358,R
1276.564087,R
1229.505371,S
1217.599976,H
1165.476563,Y
```

I/L=J
Q/K=B

Taxonomy All

☐ Titinを削除する

Cys(-SR) Cmc

Target Protein AA number >= 100

☐ Digest C-term of k,r

☐ don't cut N-term of Pro

☑ Digest N-term of k

allow up to 2 miss cleavage

peptide_len 5 ~ 30

Figure. 26 Window of iD plus for experimental protein identification (searching engine). The range of allowing mass error is multiple, the tolerance for the number of missed cleavages is optional from 0 to 2, and the titin (A2ASS6 and Q8WZ42) can be chosen to keep or remove.

5.3 Results

5.3.1 Verification of algorithm

Before applying the experimental data to iD plus, the verification of algorithm is necessary. The theoretical mass values [$M+H^+$] and the C-terminal of the peptides derived from BSA, TF, CYC and MB were input into the iD plus. According to Figure. 23, only the peptides with amino acid length between five and 30 in a database would be picked up by iD plus for database search. Table. 9 summarizes the results of algorithm verification by peptides from digest of four single proteins. All the peptide lengths between 5-30 amino acids could match to the derived proteins entirely, no additional proteins were identified.

Table. 9 Results of algorithm verification by theoretical digestion data.

Identified protein(s)	number of peptides from digest	number of peptides matched (5-30 aa)	sequence coverage
BSA	58	46	93.90%
TF	58	46	84.80%
CYC	17	11	86.70%
MB	18	11	87.70%

5.3.2 Single protein identification

Based on such a database searching workflow, various authentic proteins were measured and analyzed. BSA, TF, CYC, MB, MYH, PYGM and CA were addressed as the methodology part described in Chapter II, and the masses and C-terminal amino acid sequences of detected peptides were acquired from MALDI-TOF/TOF MS spectra. All the data was input into iD plus for sequence database searching. Table. 9-15 show the identification results by iD plus for BSA, TF, CYC, MB, MYH, PYGM and CA, respectively. For BSA identification, 73 peptide sets were input into iD plus, and 57 sets could assign to 17 proteins.

The protein with the most hits (21) and top coverage (35.10%) is BSA (P02769) (Table. 10). For TF identification, 47 peptide sets were input into iD plus, and 24 sets could assign to 5 proteins. The protein with the most hits (11) and top coverage (16.20%) is TF (Q29443) (Table.11). For CYC identification, 34 peptide sets were input into iD plus, and 27 sets could assign to 5 proteins. The protein with the most hits (8) and top coverage (89.50%) is CYC (P00004) (Table.12). For MB identification, 30 peptide sets were input into iD plus, and 17 sets could assign to 6 proteins. The protein with the most hits (8) and top coverage (61.00%) is MB (P68082/P68083) (Table.13). For MYH identification, 75 peptide sets were input into iD plus, and 59 sets could assign to 22 proteins. The protein with the most hits (14) and top coverage (7.40%) is MYH (Q28641) (Table.14). For PYGM identification, 53 peptide sets were input into iD plus, and 36 sets could assign to 9 proteins. The protein with the most hits (9) and top coverage (12.50%) is PYGM (P00489) (Table.15). For CA identification, 29 peptide sets were input into iD plus, and 18 sets could assign to 6 proteins. The protein with the most hits (6) and top coverage (28.10%) is CA (P00921) (Table.16). It indicates for single protein identification, the candidate with the most number of matched peptides and/or highest coverage can be confirmed as target protein.

Table. 10 Identification Results for BSA by iD plus.

<table>
<tr><td colspan="3">BSA</td></tr>
<tr><td>Number of sets</td><td colspan="2">73</td></tr>
<tr><td>Number of matched sets</td><td colspan="2">57</td></tr>
<tr><td>Number of matched proteins by more than three sets</td><td colspan="2">17</td></tr>
<tr><td colspan="3"></td></tr>
<tr><td>Coverage</td><td>Number of matched peptides</td><td>ID</td></tr>
<tr><td>35.10%</td><td>21</td><td>P02769</td></tr>
<tr><td>2.20%</td><td>5</td><td>Q6A078</td></tr>
<tr><td>0.20%</td><td>4</td><td>Q9I7U4</td></tr>
<tr><td>1.00%</td><td>4</td><td>Q8IDX6</td></tr>
<tr><td>0.80%</td><td>4</td><td>Q69Z23</td></tr>
<tr><td>0.80%</td><td>4</td><td>Q9UFH2</td></tr>
<tr><td>0.70%</td><td>4</td><td>Q555C6</td></tr>
<tr><td>1.70%</td><td>4</td><td>Q96RL7</td></tr>
<tr><td>1.40%</td><td>3</td><td>Q6YXP5</td></tr>
<tr><td>2.60%</td><td>3</td><td>Q9PQ74</td></tr>
<tr><td>0.80%</td><td>3</td><td>Q71LX6</td></tr>
<tr><td>2.70%</td><td>3</td><td>P21421</td></tr>
<tr><td>1.60%</td><td>3</td><td>P23466</td></tr>
<tr><td>1.50%</td><td>3</td><td>P0C568</td></tr>
<tr><td>3.30%</td><td>3</td><td>Q88VX7</td></tr>
<tr><td>0.90%</td><td>3</td><td>Q4U4S6</td></tr>
<tr><td>5.60%</td><td>3</td><td>Q46LV3</td></tr>
</table>

Table. 11 Identification Results for TF by iD plus.

TF		
Number of sets	47	
Number of matched sets	24	
Number of matched proteins by more than three sets	5	
Coverage	Number of matched peptides	ID
16.20%	11	**Q29443**
0.80%	4	Q9QYX7
1.00%	3	P09814
1.80%	3	P22082
7.60%	3	Q042P8

Table. 12 Identification Results for CYC by iD plus.

CYC		
Number of sets	34	
Number of matched sets	27	
Number of matched proteins by more than three sets	5	
Coverage	Number of matched peptides	ID
89.50%	8	**P00004**
0.40%	4	Q8I3Z1
0.40%	3	P0C6Y0
0.40%	3	C6KTB7
1.40%	3	Q6ZQQ6

Table. 13 Identification Results for MB by iD plus.

MB		
Number of sets	30	
Number of matched sets	17	
Number of matched proteins by more than three sets	6	
Coverage	Number of matched peptides	ID
61.00%	8	**P68082**
61.00%	8	**P68083**
3.70%	3	P14606
4.50%	3	Q6FKT1
0.50%	3	O68008
0.70%	3	Q4G0P3

Table. 14 Identification Results for MYH by iD plus.

MYH		
Number of sets	75	
Number of matched sets	59	
Number of matched proteins by more than three sets	22	
Coverage	Number of matched peptides	ID
7.40%	14	**Q28641**
7.40%	14	homology
7.60%	14	
7.60%	14	
7.60%	14	

7.80%	14	
1.30%	5	P62286
1.00%	5	P0C6U9
0.60%	5	P0C6X8
2.00%	4	A4QKG5
3.90%	4	P02562
1.80%	4	Q6EVK6
3.00%	3	Q54YD8
1.30%	3	Q68RU8
6.10%	3	A4XPN6
1.10%	3	Q8I4R2
6.20%	3	Q3YT49
0.20%	3	Q9I7U4
4.50%	3	P23316
4.90%	3	Q9LS42
3.70%	3	Q8ILT5
1.50%	3	Q00963

Table. 15 Identification Results for PYGM by iD plus.

PYGM		
Number of sets	53	
Number of matched sets	36	
Number of matched proteins by more than three sets	9	
Coverage	Number of matched peptides	ID
12.50%	9	**P00489**
0.40%	4	Q8I3Z1

3.80%	4	Q553B1
4.30%	3	Q5UR75
1.70%	3	Q6FSD2
2.50%	3	P37609
0.90%	3	Q8TDW7
2.30%	3	P49466
7.20%	3	Q5UPN0

Table. 16 Identification Results for CA by iD plus.

CA		
Number of sets	29	
Number of matched sets	18	
Number of matched proteins by more than three sets	6	
Coverage	Number of matched peptides	ID
28.10%	6	**P00921**
6.80%	4	P17480
6.80%	4	P25976
6.80%	4	P25977
0.50%	3	Q8IID4
0.50%	3	Q09666

5.3.3 Protein mixture identification

As to the protein identification from protein mixture by MS/MS based enhanced PMF, an artificial protein mixture of BSA, CYC and MB is measured by MALDI-TOF/TOF MS. Table.16 shows the identification results by iD plus for Table. 16 shows the searching result of the protein mixture digested LysN, proteins of a protein mixture of BSA, TF, CYC and MB. For the mixture of four proteins identification, 59 peptide sets were input into iD plus, and 39 sets could assign to 10 proteins. The protein with the 6 hits and 8.1% of coverage can be identified as BSA (P02769), the protein with the 5 hits and 33.8% of coverage can be identified as MB (68082), the protein with the 4 hits and 6.7% of coverage can be identified as TF (Q29443), and the protein with the 4 hits and 31.40% of coverage can be identified as CYC (P00004) (Table.16).

Table. 16 Identification Results for Protein Mixture by iD plus.

Protien Mixture		
Number of sets	59	
Number of matched sets	39	
Number of matched proteins by more than three sets	4	
Coverage	Number of matched peptides	ID
0.081	6	P02769
0.338	5	P68082
0.067	4	Q29443
>0.314	4	P00004

5.4 Discussion

The algorithm of iD plus is a non-probability based sequence database search, which is different with Mascot, a probability based sequence database search. The results of Mascot search in Chapter II already demonstrate the significance of C-terminal amino acid sequences deduced from metastable ions in the spectra of peptides derived from LysN digested proteins for protein identification. In this session, the algorithm of iD plus has been verified firstly by theoretical peptide masses from the protein digest list. The result indicates the validity of the algorithm of iD plus. The tolerance conditions of iD plus search allow error of ± 0.5 Da, up to 1 missed cleavage in a peptide sequence and removing the sequence of titin (A2ASS6 and Q8WZ42) from the sequence database. The first two conditions of allowing error of ± 0.5 Da and up to 1 missed cleavage correspond with the conditions of Mascot search described in Chapter III, and the reason of removing A2ASS6 and Q8WZ42 from the sequence database is that the length of the sequence of these two proteins are very long (35,213 aa and 34,350 aa). As we know, the random matches would increase with the increasing of the length of protein. In the single protein identification by iD plus using an entire sequence database, A2ASS6 and Q8WZ42 were always identified as false proteins, and a few of peptide sequence derived from protein digest could be matched to titin because in the LysN digest of titin, many sequences contain the similar masses and C-terminal amino acid sequences to other peptides derived from LysN digest of BSA/TF/CYC/MB. Therefore, A2ASS6 and Q8WZ42 were removed from sequence database to improve the algorithm of iD plus. On another hand, only the peptides with masses above 800 kDa were used for protein identification due to in the mass range below 800 kDa, matrix and other background could be measured by MS and such false input data may increase the false identification by iD plus. Under these conditions, iD plus was run for protein identification by experimental peptides data acquired from MS spectra. For the single protein identification, all the seven proteins can be matched with the most number of peptides input than the number of peptides matched to other false proteins. In addition, the coverage of the identified proteins which the most number of peptides matched to are the highest among all proteins output by iD plus. Those results are similar as Mascot search for single protein using the same input data, but iD plus does not adopt the probability-based algorithm. Whereas, the iD plus can identify protein mixture with fewer number of peptides better than Mascot does. Chapter III described that Mascot cannot identify all of the four proteins by partial peptides with masses and C-terminal amino acid sequences, though the four proteins can be identified in a mixture by adding more peptide masses without deduced C-terminal amino acid sequence. It indicates that the probability-based algorithm of Mascot cannot address the identification in the situation of fewer number of peptides can be used. However, iD plus can identify all the four proteins using the same data of peptides masses and C-terminal amino acid sequences as Mascot used and a non-probability based algorithm. Only the four proteins of BSA/MB/TF/CYC can be identified by 6, 5, 4, 4 peptides, other few number of false proteins can only match to less than 3 peptides. As to the simulation result described in Chapter IV, the reliability of protein identified by more than 4 peptides is the highest. It means the identification of protein mixture by iD plus is better than Mascot.

Conclusion

In this study, I have investigated the generation of [b] ions and [b+18] ions relative to C-terminal truncated of peptides measured by MALDI-TOF/TOF MS and proposed a new strategy of applying those metastable ions on conventional PMF to increase the reliability for protein identification. We experimentally demonstrated that the [b] ions can be generated by any peptide sequence, besides Arg-contained peptides which can produce [b+18] ions. A scheme of enhanced PMF based on the protein digest by LysN was established and the effect of C-terminal amino acid sequences were verified by Mascot. Furthermore, a non-probability based database searching algorithm “iD plus” was developed in-house. The performance of the experimental workflow and computational database searching was validated using single protein digest and protein mixture digest. The application of iD plus on addressing the identification of protein mixture was better and more reliable than the algorithm of PMF or query search of Mascot. We foresee more promising applications of the iD plus for protein identification.

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Supplement

Mascot input data format and iD plus input data (mass+c)

BSA
mass

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CA
mass+c

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4 Protein Mixture (BSA/TF/CYC/MB)
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4 Protein Mixture (BSA/TF/CYC/MB)
mass/mass+c

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4 Protein Mixture (BSA/TF/CYC/MB)
mass+c

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1048.594482 seq(C-I)
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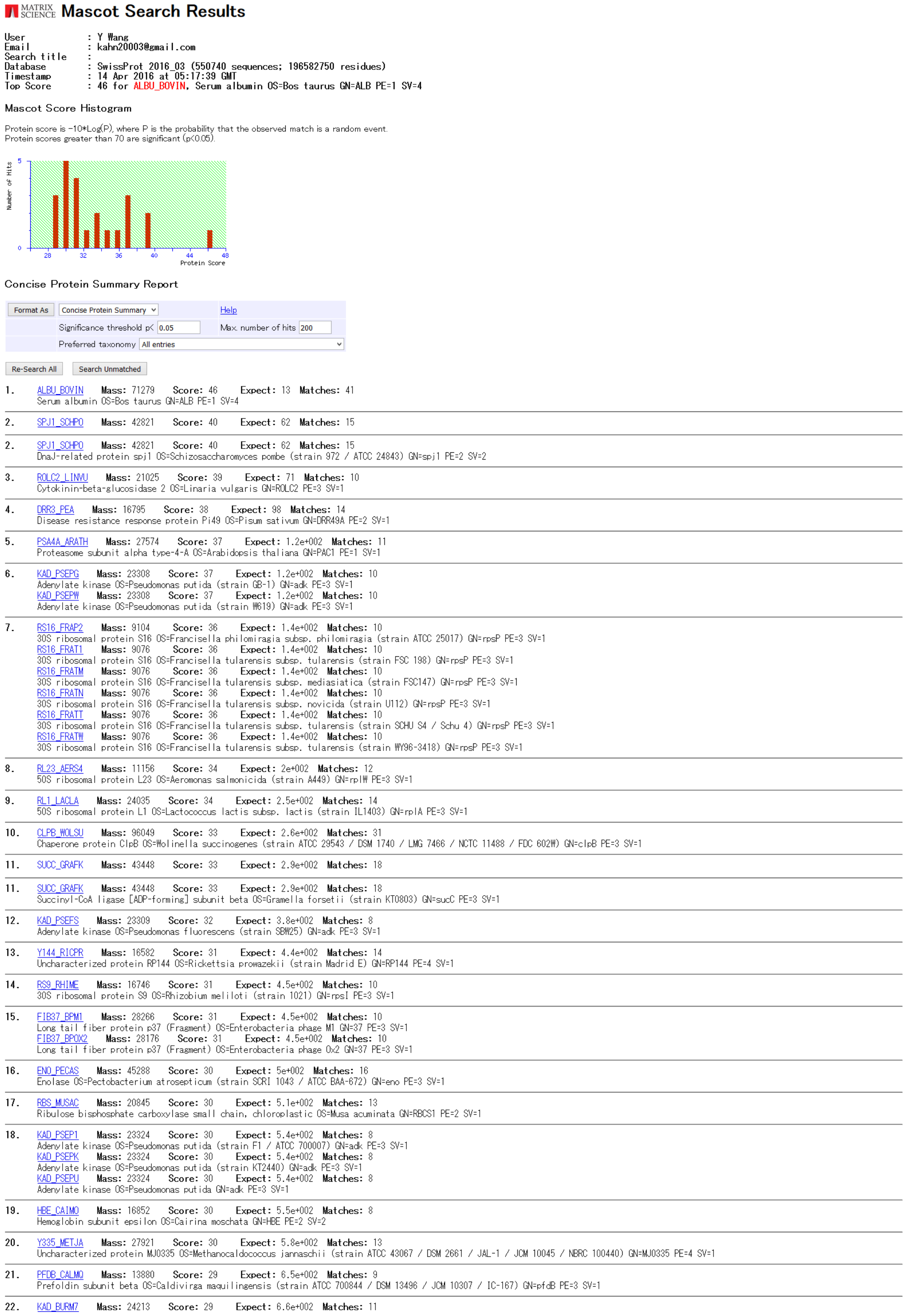
1019.489746 seq(C-Y)
1017.631958 seq(C-S)
1017.631958 seq(C-K)
1017.631958 seq(C-Q)
1016.501648 seq(C-F)
1009.585449 seq(C-T)
1006.469543 seq(C-Y)
1002.55072 seq(C-S)
996.5701294 seq(C-T)
989.6818237 seq(C-K)
989.6818237 seq(C-Q)
987.6281738 seq(C-F)
952.6633301 seq(C-T)
936.6383667 seq(C-S)
920.7224121 seq(C-R)
920.5969238 seq(C-I)
920.5969238 seq(C-L)
911.6362915 seq(C-T)
906.6486206 seq(C-E)
906.6211548 seq(C-E)
879.6194458 seq(C-E)
861.6519775 seq(C-K)
861.6519775 seq(C-Q)
858.6847534 seq(C-E)
857.6257324 seq(C-Y)
837.5790405 seq(C-G)
832.6393433 seq(C-N)
829.7144165 seq(C-G)
820.7059937 seq(C-I)
820.7059937 seq(C-L)
818.651062 seq(C-I)
818.651062 seq(C-L)
806.7282715 seq(C-T)
790.697998 seq(C-K)
790.697998 seq(C-Q)
774.6795654 seq(C-K)
774.6795654 seq(C-Q)
764.7042236 seq(C-H)
760.713562 seq(C-K)
760.713562 seq(C-Q)
752.6557007 seq(C-A)
748.6766968 seq(C-K)
748.6766968 seq(C-Q)
730.5621338 seq(C-M)
721.6623535 seq(C-Y)
708.6532593 seq(C-M)
690.7011719 seq(C-E)
689.727356 seq(C-S)
668.6647339 seq(C-F)
661.6223145 seq(C-P)
660.7211304 seq(C-K)
660.7211304 seq(C-Q)
659.7144165 seq(C-A)
648.6061401 seq(C-P)
630.7432251 seq(C-I)
630.7432251 seq(C-L)
625.6652222 seq(C-F)
609.6723633 seq(C-E)
593.7172241 seq(C-H)
585.7533569 seq(C-I)
585.7533569 seq(C-L)
572.786499 seq(C-I)
572.786499 seq(C-L)
546.5980835 seq(C-P)

Mascot Output Format

BSA mass



Mascot Search Results

```
User          : Y Wang
Email         : kahn20003@gmail.com
Search title  :
Database      : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp     : 14 Apr 2016 at 05:17:39 GMT
Top Score     : 46 for ALBU_BOVIN, Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Concise Protein Summary
Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

```
1.    ALBU_BOVIN    Mass: 71279    Score: 46    Expect: 13  Matches: 41
      Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4

2.    SPJ1_SCHPO    Mass: 42821    Score: 40    Expect: 62  Matches: 15

2.    SPJ1_SCHPO    Mass: 42821    Score: 40    Expect: 62  Matches: 15
      DnaJ-related protein spj1 OS=Schizosaccharomyces pombe (strain 972 / ATCC 24843) GN=spj1 PE=2 SV=2

3.    ROLC2_LINVU    Mass: 21025    Score: 39    Expect: 71  Matches: 10
      Cytokinin-beta-glucosidase 2 OS=Linaria vulgaris GN=ROLC2 PE=3 SV=1

4.    DRR3_PEA    Mass: 16795    Score: 38    Expect: 98  Matches: 14
      Disease resistance response protein Pi49 OS=Pisum sativum GN=DRR49A PE=2 SV=1

5.    PSA4A_ARATH    Mass: 27574    Score: 37    Expect: 1.2e+002  Matches: 11
      Proteasome subunit alpha type-4-A OS=Arabidopsis thaliana GN=PAC1 PE=1 SV=1

6.    KAD_PSEPG    Mass: 23308    Score: 37    Expect: 1.2e+002  Matches: 10
      Adenylate kinase OS=Pseudomonas putida (strain GB-1) GN=adk PE=3 SV=1
      KAD_PSEPW    Mass: 23308    Score: 37    Expect: 1.2e+002  Matches: 10
      Adenylate kinase OS=Pseudomonas putida (strain W619) GN=adk PE=3 SV=1

7.    RS16_FRAP2    Mass: 9104    Score: 36    Expect: 1.4e+002  Matches: 10
      30S ribosomal protein S16 OS=Francisella philomiragia subsp. philomiragia (strain ATCC 25017) GN=rpsP PE=3 SV=1
      RS16_FRAT1    Mass: 9076    Score: 36    Expect: 1.4e+002  Matches: 10
      30S ribosomal protein S16 OS=Francisella tularensis subsp. tularensis (strain FSC 198) GN=rpsP PE=3 SV=1
      RS16_FRATM    Mass: 9076    Score: 36    Expect: 1.4e+002  Matches: 10
      30S ribosomal protein S16 OS=Francisella tularensis subsp. mediasiatica (strain FSC147) GN=rpsP PE=3 SV=1
      RS16_FRATN    Mass: 9076    Score: 36    Expect: 1.4e+002  Matches: 10
      30S ribosomal protein S16 OS=Francisella tularensis subsp. novicida (strain U112) GN=rpsP PE=3 SV=1
      RS16_FRATT    Mass: 9076    Score: 36    Expect: 1.4e+002  Matches: 10
      30S ribosomal protein S16 OS=Francisella tularensis subsp. tularensis (strain SCHU S4 / Schu 4) GN=rpsP PE=3 SV=1
      RS16_FRATW    Mass: 9076    Score: 36    Expect: 1.4e+002  Matches: 10
      30S ribosomal protein S16 OS=Francisella tularensis subsp. tularensis (strain WY96-3418) GN=rpsP PE=3 SV=1

8.    RL23_AERS4    Mass: 11156    Score: 34    Expect: 2e+002  Matches: 12
      50S ribosomal protein L23 OS=Aeromonas salmonicida (strain A449) GN=rplW PE=3 SV=1

9.    RL1_LACLA    Mass: 24035    Score: 34    Expect: 2.5e+002  Matches: 14
      50S ribosomal protein L1 OS=Lactococcus lactis subsp. lactis (strain IL1403) GN=rplA PE=3 SV=1

10.   CLPB_WOLSU    Mass: 96049    Score: 33    Expect: 2.6e+002  Matches: 31
      Chaperone protein ClpB OS=Wolinella succinogenes (strain ATCC 29543 / DSM 1740 / LMG 7466 / NCTC 11488 / FDC 602W) GN=clpB PE=3 SV=1

11.   SUCC_GRAFK    Mass: 43448    Score: 33    Expect: 2.9e+002  Matches: 18

11.   SUCC_GRAFK    Mass: 43448    Score: 33    Expect: 2.9e+002  Matches: 18
      Succinyl-CoA ligase [ADP-forming] subunit beta OS=Gramella forsetii (strain KT0803) GN=sucC PE=3 SV=1

12.   KAD_PSEFS    Mass: 23309    Score: 32    Expect: 3.8e+002  Matches: 8
      Adenylate kinase OS=Pseudomonas fluorescens (strain SBW25) GN=adk PE=3 SV=1

13.   Y144_RICPR    Mass: 16582    Score: 31    Expect: 4.4e+002  Matches: 14
      Uncharacterized protein RP144 OS=Rickettsia prowazekii (strain Madrid E) GN=RP144 PE=4 SV=1

14.   RS9_RHIME    Mass: 16746    Score: 31    Expect: 4.5e+002  Matches: 10
      30S ribosomal protein S9 OS=Rhizobium meliloti (strain 1021) GN=rpsI PE=3 SV=1

15.   FIB37_BPM1    Mass: 28266    Score: 31    Expect: 4.5e+002  Matches: 10
      Long tail fiber protein p37 (Fragment) OS=Enterobacteria phage M1 GN=37 PE=3 SV=1
      FIB37_BPOX2    Mass: 28176    Score: 31    Expect: 4.5e+002  Matches: 10
      Long tail fiber protein p37 (Fragment) OS=Enterobacteria phage Ox2 GN=37 PE=3 SV=1

16.   ENO_PECAS    Mass: 45288    Score: 30    Expect: 5e+002  Matches: 16
      Enolase OS=Pectobacterium atrosepticum (strain SCRI 1043 / ATCC BAA-672) GN=eno PE=3 SV=1

17.   RBS_MUSAC    Mass: 20845    Score: 30    Expect: 5.1e+002  Matches: 13
      Ribulose bisphosphate carboxylase small chain, chloroplastic OS=Musa acuminata GN=RBCS1 PE=2 SV=1

18.   KAD_PSEP1    Mass: 23324    Score: 30    Expect: 5.4e+002  Matches: 8
      Adenylate kinase OS=Pseudomonas putida (strain F1 / ATCC 700007) GN=adk PE=3 SV=1
      KAD_PSEPK    Mass: 23324    Score: 30    Expect: 5.4e+002  Matches: 8
      Adenylate kinase OS=Pseudomonas putida (strain KT2440) GN=adk PE=3 SV=1
      KAD_PSEPU    Mass: 23324    Score: 30    Expect: 5.4e+002  Matches: 8
      Adenylate kinase OS=Pseudomonas putida GN=adk PE=3 SV=1

19.   HBE_CAIMO    Mass: 16852    Score: 30    Expect: 5.5e+002  Matches: 8
      Hemoglobin subunit epsilon OS=Cairina moschata GN=HBE PE=2 SV=2

20.   Y335_METJA    Mass: 27921    Score: 30    Expect: 5.8e+002  Matches: 13
      Uncharacterized protein MJ0335 OS=Methanocaldococcus jannaschii (strain ATCC 43067 / DSM 2661 / JAL-1 / JCM 10045 / NBRC 100440) GN=MJ0335 PE=4 SV=1

21.   PFDB_CALMQ    Mass: 13880    Score: 29    Expect: 6.5e+002  Matches: 9
      Prefoldin subunit beta OS=Caldivirga maquilingensis (strain ATCC 700844 / DSM 13496 / JCM 10307 / IC-167) GN=pfdB PE=3 SV=1

22.   KAD_BURM7    Mass: 24213    Score: 29    Expect: 6.6e+002  Matches: 11
```

```
22.   KAD_BURM7    Mass: 24213   Score: 29    Expect: 6.6e+002  Matches: 11
      Adenylate kinase OS=Burkholderia mallei (strain NCTC 10247) GN=adk PE=3 SV=1
      KAD_BURM9    Mass: 24213   Score: 29    Expect: 6.6e+002  Matches: 11
      Adenylate kinase OS=Burkholderia mallei (strain NCTC 10229) GN=adk PE=3 SV=1
      KAD_BURMA    Mass: 24213   Score: 29    Expect: 6.6e+002  Matches: 11
      Adenylate kinase OS=Burkholderia mallei (strain ATCC 23344) GN=adk PE=3 SV=1
      KAD_BURMS    Mass: 24213   Score: 29    Expect: 6.6e+002  Matches: 11
      Adenylate kinase OS=Burkholderia mallei (strain SAVP1) GN=adk PE=3 SV=1
      KAD_BURP0    Mass: 24213   Score: 29    Expect: 6.6e+002  Matches: 11
      Adenylate kinase OS=Burkholderia pseudomallei (strain 1106a) GN=adk PE=3 SV=1
      KAD_BURP1    Mass: 24213   Score: 29    Expect: 6.6e+002  Matches: 11
      Adenylate kinase OS=Burkholderia pseudomallei (strain 1710b) GN=adk PE=1 SV=1
      KAD_BURP6    Mass: 24213   Score: 29    Expect: 6.6e+002  Matches: 11
      Adenylate kinase OS=Burkholderia pseudomallei (strain 668) GN=adk PE=3 SV=1
      KAD_BURPS    Mass: 24213   Score: 29    Expect: 6.6e+002  Matches: 11
      Adenylate kinase OS=Burkholderia pseudomallei (strain K96243) GN=adk PE=3 SV=1
```

```
23.   HSLV_STAA1    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain Mu3 / ATCC 700698) GN=hslV PE=3 SV=1
      HSLV_STAA2    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain JH1) GN=hslV PE=3 SV=1
      HSLV_STAA3    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain USA300) GN=hslV PE=3 SV=1
      HSLV_STAA8    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain NCTC 8325) GN=hslV PE=3 SV=1
      HSLV_STAA9    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain JH9) GN=hslV PE=3 SV=1
      HSLV_STAAB    Mass: 19599   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain bovine RF122 / ET3-1) GN=hslV PE=3 SV=1
      HSLV_STAAC    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain COL) GN=hslV PE=3 SV=1
      HSLV_STAAE    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain Newman) GN=hslV PE=3 SV=1
      HSLV_STAAM    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain Mu50 / ATCC 700699) GN=hslV PE=3 SV=1
      HSLV_STAAN    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain N315) GN=hslV PE=1 SV=1
      HSLV_STAAR    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain MRSA252) GN=hslV PE=3 SV=1
      HSLV_STAAS    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain MRSA252) GN=hslV PE=3 SV=1
      HSLV_STAAS    Mass: 19618   Score: 29    Expect: 7.1e+002  Matches: 8
      ATP-dependent protease subunit HslV OS=Staphylococcus aureus (strain MSSA476) GN=hslV PE=3 SV=1
```

```
Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (815.7810,1+): <no title>
   Query2 (816.6786,1+): <no title>
   Query3 (818.4763,1+): <no title>
   Query4 (820.5340,1+): <no title>
   Query5 (832.5363,1+): <no title>
   Query6 (835.5478,1+): <no title>
   Query7 (835.5504,1+): <no title>
   Query8 (835.5682,1+): <no title>
   Query9 (836.3428,1+): <no title>
  Query10 (840.5561,1+): <no title>
  Query11 (840.5872,1+): <no title>
  Query12 (840.5909,1+): <no title>
  Query13 (840.9949,1+): <no title>
  Query14 (841.3505,1+): <no title>
  Query15 (841.5867,1+): <no title>
  Query16 (844.4614,1+): <no title>
  Query17 (854.3765,1+): <no title>
  Query18 (857.5164,1+): <no title>
  Query19 (858.5507,1+): <no title>
  Query20 (860.3229,1+): <no title>
  Query21 (860.6417,1+): <no title>
  Query22 (861.5379,1+): <no title>
  Query23 (869.5292,1+): <no title>
  Query24 (871.5303,1+): <no title>
  Query25 (872.5607,1+): <no title>
  Query26 (876.5069,1+): <no title>
  Query27 (877.1004,1+): <no title>
  Query28 (877.1182,1+): <no title>
  Query29 (878.5225,1+): <no title>
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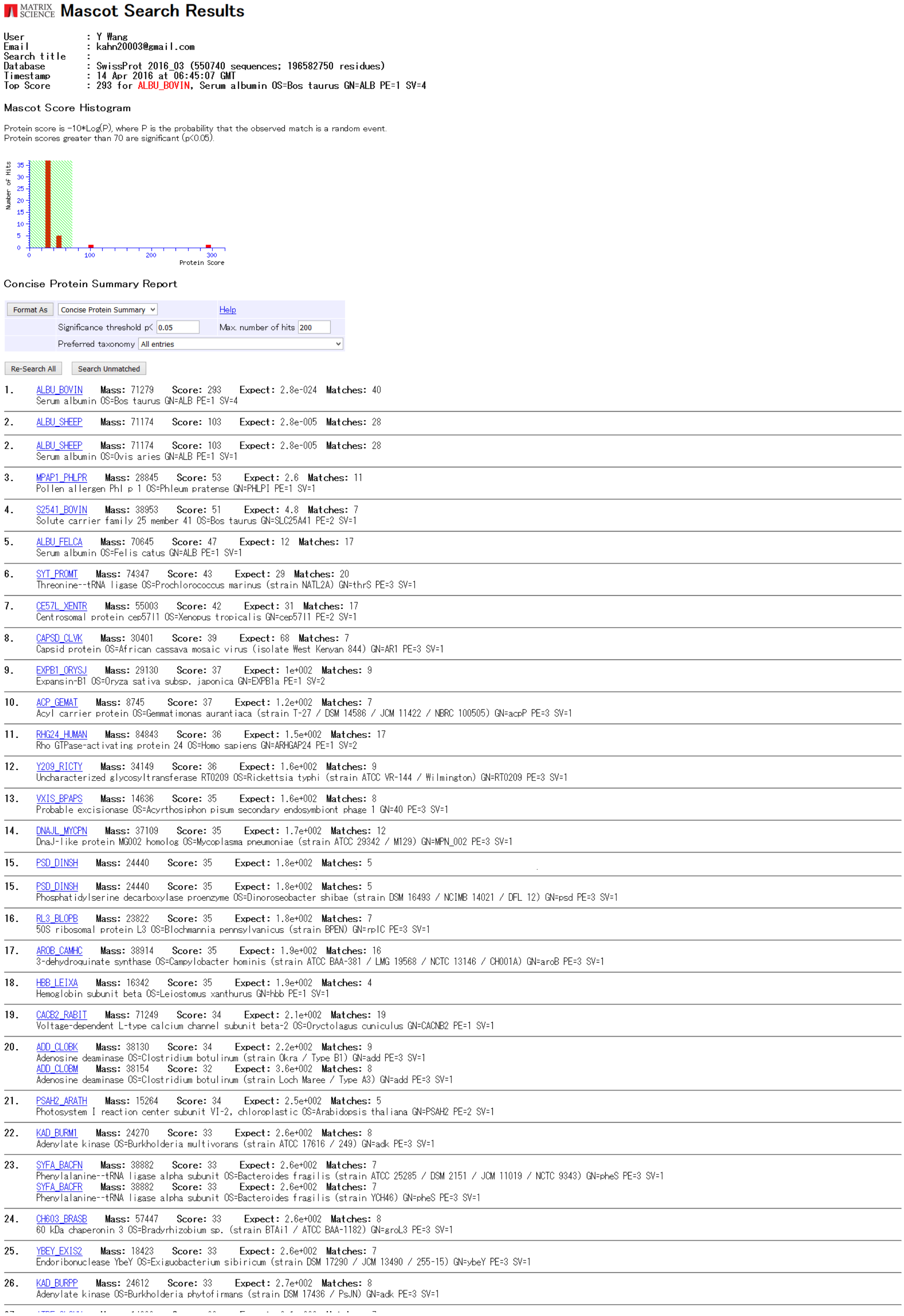
BSA mass/mass+c



Mascot Search Results

User : Y Wang
Email : kahn20003@gmail.com
Search title :
Database : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp : 14 Apr 2016 at 06:45:07 GMT
Top Score : 293 for ALBU_BOVIN, Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

1. ALBU_BOVIN **Mass:** 71279 **Score:** 293 **Expect:** 2.8e-024 **Matches:** 40
Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4

2. ALBU_SHEEP **Mass:** 71174 **Score:** 103 **Expect:** 2.8e-005 **Matches:** 28

2. ALBU_SHEEP **Mass:** 71174 **Score:** 103 **Expect:** 2.8e-005 **Matches:** 28
Serum albumin OS=Ovis aries GN=ALB PE=1 SV=1

3. MPAP1_PHLPR **Mass:** 28845 **Score:** 53 **Expect:** 2.6 **Matches:** 11
Pollen allergen Phl p 1 OS=Phleum pratense GN=PHLPI PE=1 SV=1

4. S2541_BOVIN **Mass:** 38953 **Score:** 51 **Expect:** 4.8 **Matches:** 7
Solute carrier family 25 member 41 OS=Bos taurus GN=SLC25A41 PE=2 SV=1

5. ALBU_FELCA **Mass:** 70645 **Score:** 47 **Expect:** 12 **Matches:** 17
Serum albumin OS=Felis catus GN=ALB PE=1 SV=1

6. SYT_PROMT **Mass:** 74347 **Score:** 43 **Expect:** 29 **Matches:** 20
Threonine--tRNA ligase OS=Prochlorococcus marinus (strain NATL2A) GN=thrS PE=3 SV=1

7. CE57L_XENTR **Mass:** 55003 **Score:** 42 **Expect:** 31 **Matches:** 17
Centrosomal protein cep57l1 OS=Xenopus tropicalis GN=cep57l1 PE=2 SV=1

8. CAPSD_CLVK **Mass:** 30401 **Score:** 39 **Expect:** 68 **Matches:** 7
Capsid protein OS=African cassava mosaic virus (isolate West Kenyan 844) GN=AR1 PE=3 SV=1

9. EXPB1_ORYSJ **Mass:** 29130 **Score:** 37 **Expect:** 1e+002 **Matches:** 9
Expansin-B1 OS=Oryza sativa subsp. japonica GN=EXPB1a PE=1 SV=2

10. ACP_GEMAT **Mass:** 8745 **Score:** 37 **Expect:** 1.2e+002 **Matches:** 7
Acyl carrier protein OS=Gemmatimonas aurantiaca (strain T-27 / DSM 14586 / JCM 11422 / NBRC 100505) GN=acpP PE=3 SV=1

11. RHG24_HUMAN **Mass:** 84843 **Score:** 36 **Expect:** 1.5e+002 **Matches:** 17
Rho GTPase-activating protein 24 OS=Homo sapiens GN=ARHGAP24 PE=1 SV=2

12. Y209_RICTY **Mass:** 34149 **Score:** 36 **Expect:** 1.6e+002 **Matches:** 9
Uncharacterized glycosyltransferase RT0209 OS=Rickettsia typhi (strain ATCC VR-144 / Wilmington) GN=RT0209 PE=3 SV=1

13. VXIS_BPAPS **Mass:** 14636 **Score:** 35 **Expect:** 1.6e+002 **Matches:** 8
Probable excisionase OS=Acyrthosiphon pisum secondary endosymbiont phage 1 GN=40 PE=3 SV=1

14. DNAJL_MYCPN **Mass:** 37109 **Score:** 35 **Expect:** 1.7e+002 **Matches:** 12
DnaJ-like protein MG002 homolog OS=Mycoplasma pneumoniae (strain ATCC 29342 / M129) GN=MPN_002 PE=3 SV=1

15. PSD_DINSH **Mass:** 24440 **Score:** 35 **Expect:** 1.8e+002 **Matches:** 5

15. PSD_DINSH **Mass:** 24440 **Score:** 35 **Expect:** 1.8e+002 **Matches:** 5
Phosphatidylserine decarboxylase proenzyme OS=Dinoroseobacter shibae (strain DSM 16493 / NCIMB 14021 / DFL 12) GN=psd PE=3 SV=1

16. RL3_BLOPB **Mass:** 23822 **Score:** 35 **Expect:** 1.8e+002 **Matches:** 7
50S ribosomal protein L3 OS=Blochmannia pennsylvanicus (strain BPEN) GN=rplC PE=3 SV=1

17. AROB_CAMHC **Mass:** 38914 **Score:** 35 **Expect:** 1.9e+002 **Matches:** 16
3-dehydroquinate synthase OS=Campylobacter hominis (strain ATCC BAA-381 / LMG 19568 / NCTC 13146 / CH001A) GN=aroB PE=3 SV=1

18. HBB_LEIXA **Mass:** 16342 **Score:** 35 **Expect:** 1.9e+002 **Matches:** 4
Hemoglobin subunit beta OS=Leiostomus xanthurus GN=hbb PE=1 SV=1

19. CACB2_RABIT **Mass:** 71249 **Score:** 34 **Expect:** 2.1e+002 **Matches:** 19
Voltage-dependent L-type calcium channel subunit beta-2 OS=Oryctolagus cuniculus GN=CACNB2 PE=1 SV=1

20. ADD_CLOBK **Mass:** 38130 **Score:** 34 **Expect:** 2.2e+002 **Matches:** 9
Adenosine deaminase OS=Clostridium botulinum (strain Okra / Type B1) GN=add PE=3 SV=1
ADD_CLOBM **Mass:** 38154 **Score:** 32 **Expect:** 3.6e+002 **Matches:** 8
Adenosine deaminase OS=Clostridium botulinum (strain Loch Maree / Type A3) GN=add PE=3 SV=1

21. PSAH2_ARATH **Mass:** 15264 **Score:** 34 **Expect:** 2.5e+002 **Matches:** 5
Photosystem I reaction center subunit VI-2, chloroplastic OS=Arabidopsis thaliana GN=PSAH2 PE=2 SV=1

22. KAD_BURM1 **Mass:** 24270 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 8
Adenylate kinase OS=Burkholderia multivorans (strain ATCC 17616 / 249) GN=adk PE=3 SV=1

23. SYFA_BACFN **Mass:** 38882 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 7
Phenylalanine--tRNA ligase alpha subunit OS=Bacteroides fragilis (strain ATCC 25285 / DSM 2151 / JCM 11019 / NCTC 9343) GN=pheS PE=3 SV=1
SYFA_BACFR **Mass:** 38882 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 7
Phenylalanine--tRNA ligase alpha subunit OS=Bacteroides fragilis (strain YCH46) GN=pheS PE=3 SV=1

24. CH603_BRASB **Mass:** 57447 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 8
60 kDa chaperonin 3 OS=Bradyrhizobium sp. (strain BTAi1 / ATCC BAA-1182) GN=groL3 PE=3 SV=1

25. YBEY_EXIS2 **Mass:** 18423 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 7
Endoribonuclease YbeY OS=Exiguobacterium sibiricum (strain DSM 17290 / JCM 13490 / 255-15) GN=ybeY PE=3 SV=1

26. KAD_BURPP **Mass:** 24612 **Score:** 33 **Expect:** 2.7e+002 **Matches:** 8
Adenylate kinase OS=Burkholderia phytofirmans (strain DSM 17436 / PsJN) GN=adk PE=3 SV=1

```
     Adenylate kinase OS=Burkholderia phytofirmans (strain DSM 17436 / PsJN) GN=adk PE=3 SV=1

27.  ATPE_CLONN    Mass: 14963    Score: 32    Expect: 3.1e+002  Matches: 7
     ATP synthase epsilon chain OS=Clostridium novyi (strain NT) GN=atpC PE=3 SV=1

28.  RHG24_MOUSE    Mass: 84685    Score: 32    Expect: 3.2e+002  Matches: 14
     Rho GTPase-activating protein 24 OS=Mus musculus GN=Arhgap24 PE=1 SV=2

29.  Y218_RICPR    Mass: 34113    Score: 32    Expect: 3.2e+002  Matches: 8
     Uncharacterized glycosyltransferase RP128 OS=Rickettsia prowazekii (strain Madrid E) GN=RP218 PE=3 SV=1

30.  RS17_BACCR    Mass: 10198    Score: 32    Expect: 3.3e+002  Matches: 6
     30S ribosomal protein S17 OS=Bacillus cereus (strain ATCC 14579 / DSM 31 / JCM 2152 / NBRC 15305 / NCIMB 9373 / NRRL B-3711) GN=rpsQ PE=3 SV=1

31.  TIG_WOLTR    Mass: 51462    Score: 32    Expect: 3.4e+002  Matches: 9
     Trigger factor OS=Wolbachia sp. subsp. Brugia malayi (strain TRS) GN=tig PE=3 SV=1

32.  KAD_BURA4    Mass: 24200    Score: 32    Expect: 3.4e+002  Matches: 8
     Adenylate kinase OS=Burkholderia ambifaria (strain MC40-6) GN=adk PE=3 SV=1
     KAD_BURCM    Mass: 24200    Score: 32    Expect: 3.7e+002  Matches: 8
     Adenylate kinase OS=Burkholderia ambifaria (strain ATCC BAA-244 / AMMD) GN=adk PE=3 SV=1

33.  NADM_METJA    Mass: 19597    Score: 32    Expect: 3.4e+002  Matches: 5
     Nicotinamide-nucleotide adenylyltransferase OS=Methanocaldococcus jannaschii (strain ATCC 43067 / DSM 2661 / JAL-1 / JCM 10045 / NBRC 100440) GN=MJ0541 PE=1 SV=3

34.  RS14_EXIS2    Mass: 10114    Score: 32    Expect: 3.6e+002  Matches: 2
     30S ribosomal protein S14 OS=Exiguobacterium sibiricum (strain DSM 17290 / JCM 13490 / 255-15) GN=rpsN PE=3 SV=1

35.  NS5_LDCPR    Mass: 36011    Score: 32    Expect: 3.7e+002  Matches: 8
     Non-structural protein 5 OS=Lymantria dispar cypovirus 1 (isolate Rao) GN=S9 PE=4 SV=1

36.  CRL_ECO27    Mass: 15763    Score: 32    Expect: 3.7e+002  Matches: 4
     Sigma factor-binding protein Crl OS=Escherichia coli O127:H6 (strain E2348/69 / EPEC) GN=crl PE=3 SV=1
     CRL_ECO7I    Mass: 15763    Score: 32    Expect: 3.7e+002  Matches: 4
     Sigma factor-binding protein Crl OS=Escherichia coli O7:K1 (strain IAI39 / ExPEC) GN=crl PE=3 SV=1
     CRL_ECOL6    Mass: 15806    Score: 32    Expect: 3.7e+002  Matches: 4
     Sigma factor-binding protein Crl OS=Escherichia coli O6:H1 (strain CFT073 / ATCC 700928 / UPEC) GN=crl PE=3 SV=3
     CRL_ECOLU    Mass: 15778    Score: 32    Expect: 3.7e+002  Matches: 4
     Sigma factor-binding protein Crl OS=Escherichia coli O17:K52:H18 (strain UMN026 / ExPEC) GN=crl PE=3 SV=1

37.  UL24_ELHVK    Mass: 26729    Score: 31    Expect: 4e+002  Matches: 11

37.  UL24_ELHVK    Mass: 26729    Score: 31    Expect: 4e+002  Matches: 11
     Protein UL24 homolog OS=Elephantid herpesvirus 1 (isolate Asian elephant/Berlin/Kiba/1998) PE=2 SV=1

38.  RANL_DROME    Mass: 24939    Score: 31    Expect: 4e+002  Matches: 11
     GTP-binding nuclear protein Ran-like OS=Drosophila melanogaster GN=Ran-like PE=1 SV=1

39.  MSH3_MOUSE    Mass: 124331    Score: 31    Expect: 4.2e+002  Matches: 17
     DNA mismatch repair protein Msh3 OS=Mus musculus GN=Msh3 PE=2 SV=3

40.  BRE5_YEAST    Mass: 57813    Score: 31    Expect: 4.2e+002  Matches: 15
     UBP3-associated protein BRE5 OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=BRE5 PE=1 SV=1

41.  SFSA_PROM5    Mass: 28564    Score: 31    Expect: 4.3e+002  Matches: 12
     Sugar fermentation stimulation protein homolog OS=Prochlorococcus marinus (strain MIT 9515) GN=sfsA PE=3 SV=1

42.  RL26_BRACM    Mass: 16885    Score: 31    Expect: 4.5e+002  Matches: 6
     60S ribosomal protein L26 OS=Brassica campestris GN=RPL26 PE=2 SV=1

43.  MURC_BACCN    Mass: 49574    Score: 31    Expect: 4.6e+002  Matches: 8
     UDP-N-acetylmuramate--L-alanine ligase OS=Bacillus cytotoxicus (strain DSM 22905 / CIP 110041 / 391-98 / NVH 391-98) GN=murC PE=3 SV=1

44.  RS17_BACSU    Mass: 10193    Score: 31    Expect: 4.7e+002  Matches: 4
     30S ribosomal protein S17 OS=Bacillus subtilis (strain 168) GN=rpsQ PE=1 SV=3

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
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   Query2 (816.6786,1+): <no title>
   Query3 (818.4763,1+): <no title>
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   Query4 (818.4763,1+): <no title>
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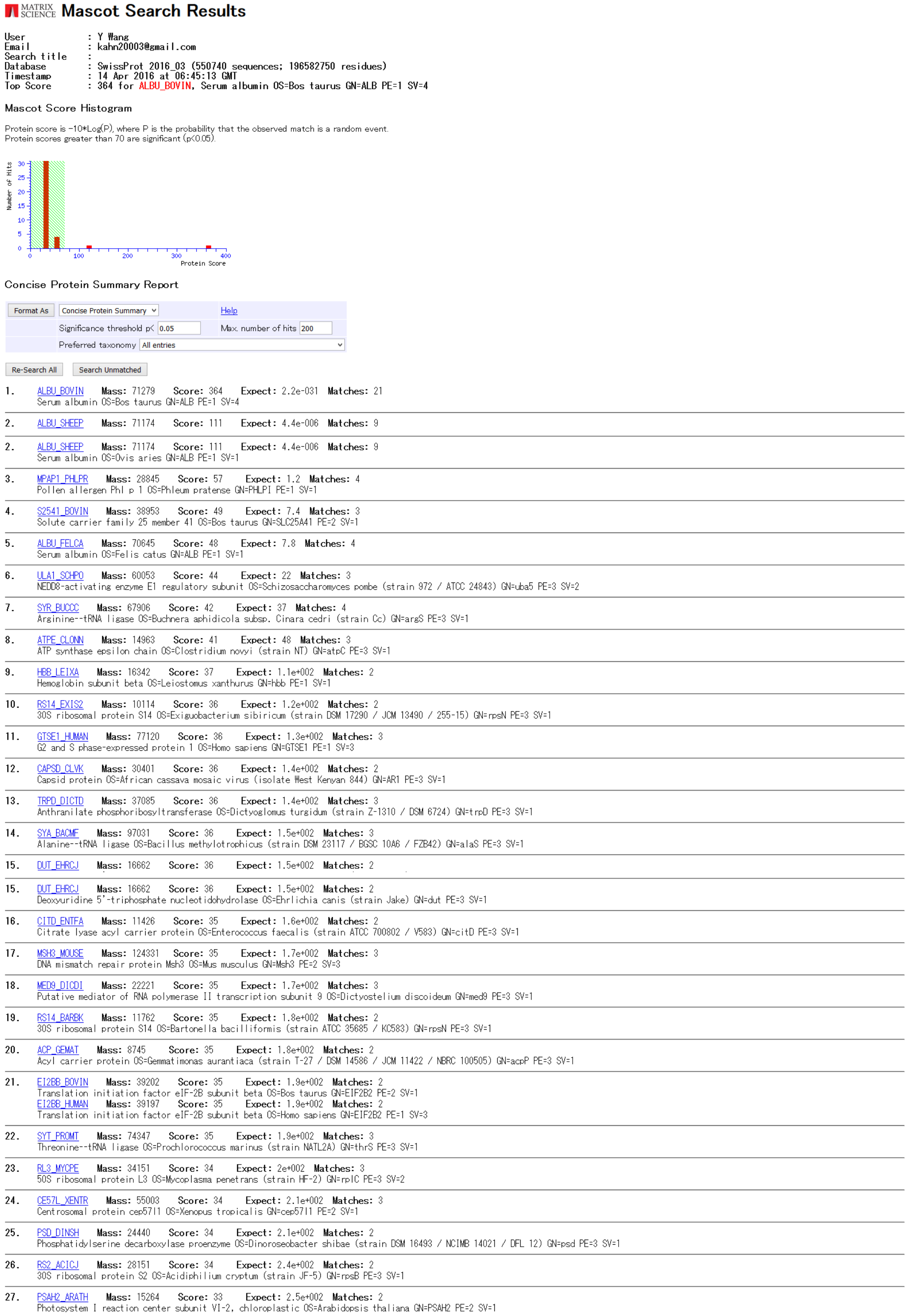
BSA mass+c



Mascot Search Results

```
User          : Y Wang
Email         : kahn20003@gmail.com
Search title  :
Database      : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp     : 14 Apr 2016 at 06:45:13 GMT
Top Score     : 364 for ALBU_BOVIN, Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

```
1.   ALBU_BOVIN     Mass: 71279    Score: 364    Expect: 2.2e-031  Matches: 21
     Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4

2.   ALBU_SHEEP     Mass: 71174    Score: 111    Expect: 4.4e-006  Matches: 9

2.   ALBU_SHEEP     Mass: 71174    Score: 111    Expect: 4.4e-006  Matches: 9
     Serum albumin OS=Ovis aries GN=ALB PE=1 SV=1

3.   MPAP1_PHLPR    Mass: 28845    Score: 57     Expect: 1.2  Matches: 4
     Pollen allergen Phl p 1 OS=Phleum pratense GN=PHLPI PE=1 SV=1

4.   S2541_BOVIN    Mass: 38953    Score: 49     Expect: 7.4  Matches: 3
     Solute carrier family 25 member 41 OS=Bos taurus GN=SLC25A41 PE=2 SV=1

5.   ALBU_FELCA     Mass: 70645    Score: 48     Expect: 7.8  Matches: 4
     Serum albumin OS=Felis catus GN=ALB PE=1 SV=1

6.   ULA1_SCHPO     Mass: 60053    Score: 44     Expect: 22  Matches: 3
     NEDD8-activating enzyme E1 regulatory subunit OS=Schizosaccharomyces pombe (strain 972 / ATCC 24843) GN=uba5 PE=3 SV=2

7.   SYR_BUCCC     Mass: 67906    Score: 42     Expect: 37  Matches: 4
     Arginine--tRNA ligase OS=Buchnera aphidicola subsp. Cinara cedri (strain Cc) GN=argS PE=3 SV=1

8.   ATPE_CLONN     Mass: 14963    Score: 41     Expect: 48  Matches: 3
     ATP synthase epsilon chain OS=Clostridium novyi (strain NT) GN=atpC PE=3 SV=1

9.   HBB_LEIXA     Mass: 16342    Score: 37     Expect: 1.1e+002  Matches: 2
     Hemoglobin subunit beta OS=Leiostomus xanthurus GN=hbb PE=1 SV=1

10.  RS14_EXIS2     Mass: 10114    Score: 36     Expect: 1.2e+002  Matches: 2
     30S ribosomal protein S14 OS=Exiguobacterium sibiricum (strain DSM 17290 / JCM 13490 / 255-15) GN=rpsN PE=3 SV=1

11.  GTSE1_HUMAN    Mass: 77120    Score: 36     Expect: 1.3e+002  Matches: 3
     G2 and S phase-expressed protein 1 OS=Homo sapiens GN=GTSE1 PE=1 SV=3

12.  CAPSD_CLVK     Mass: 30401    Score: 36     Expect: 1.4e+002  Matches: 2
     Capsid protein OS=African cassava mosaic virus (isolate West Kenyan 844) GN=AR1 PE=3 SV=1

13.  TRPD_DICTD     Mass: 37085    Score: 36     Expect: 1.4e+002  Matches: 3
     Anthranilate phosphoribosyltransferase OS=Dictyoglomus turgidum (strain Z-1310 / DSM 6724) GN=trpD PE=3 SV=1

14.  SYA_BACMF     Mass: 97031    Score: 36     Expect: 1.5e+002  Matches: 3
     Alanine--tRNA ligase OS=Bacillus methylotrophicus (strain DSM 23117 / BGSC 10A6 / FZB42) GN=alaS PE=3 SV=1

15.  DUT_EHRCJ     Mass: 16662    Score: 36     Expect: 1.5e+002  Matches: 2

15.  DUT_EHRCJ     Mass: 16662    Score: 36     Expect: 1.5e+002  Matches: 2
     Deoxyuridine 5'-triphosphate nucleotidohydrolase OS=Ehrlichia canis (strain Jake) GN=dut PE=3 SV=1

16.  CITD_ENTFA     Mass: 11426    Score: 35     Expect: 1.6e+002  Matches: 2
     Citrate lyase acyl carrier protein OS=Enterococcus faecalis (strain ATCC 700802 / V583) GN=citD PE=3 SV=1

17.  MSH3_MOUSE     Mass: 124331   Score: 35     Expect: 1.7e+002  Matches: 3
     DNA mismatch repair protein Msh3 OS=Mus musculus GN=Msh3 PE=2 SV=3

18.  MED9_DICDI     Mass: 22221    Score: 35     Expect: 1.7e+002  Matches: 3
     Putative mediator of RNA polymerase II transcription subunit 9 OS=Dictyostelium discoideum GN=med9 PE=3 SV=1

19.  RS14_BARBK     Mass: 11762    Score: 35     Expect: 1.8e+002  Matches: 2
     30S ribosomal protein S14 OS=Bartonella bacilliformis (strain ATCC 35685 / KC583) GN=rpsN PE=3 SV=1

20.  ACP_GEMAT     Mass: 8745     Score: 35     Expect: 1.8e+002  Matches: 2
     Acyl carrier protein OS=Gemmatimonas aurantiaca (strain T-27 / DSM 14586 / JCM 11422 / NBRC 100505) GN=acpP PE=3 SV=1

21.  EI2BB_BOVIN    Mass: 39202    Score: 35     Expect: 1.9e+002  Matches: 2
     Translation initiation factor eIF-2B subunit beta OS=Bos taurus GN=EIF2B2 PE=2 SV=1
     EI2BB_HUMAN    Mass: 39197    Score: 35     Expect: 1.9e+002  Matches: 2
     Translation initiation factor eIF-2B subunit beta OS=Homo sapiens GN=EIF2B2 PE=1 SV=3

22.  SYT_PROMT     Mass: 74347    Score: 35     Expect: 1.9e+002  Matches: 3
     Threonine--tRNA ligase OS=Prochlorococcus marinus (strain NATL2A) GN=thrS PE=3 SV=1

23.  RL3_MYCPE     Mass: 34151    Score: 34     Expect: 2e+002  Matches: 3
     50S ribosomal protein L3 OS=Mycoplasma penetrans (strain HF-2) GN=rplC PE=3 SV=2

24.  CE57L_XENTR    Mass: 55003    Score: 34     Expect: 2.1e+002  Matches: 3
     Centrosomal protein cep57l1 OS=Xenopus tropicalis GN=cep57l1 PE=2 SV=1

25.  PSD_DINSH     Mass: 24440    Score: 34     Expect: 2.1e+002  Matches: 2
     Phosphatidylserine decarboxylase proenzyme OS=Dinoroseobacter shibae (strain DSM 16493 / NCIMB 14021 / DFL 12) GN=psd PE=3 SV=1

26.  RS2_ACICJ     Mass: 28151    Score: 34     Expect: 2.4e+002  Matches: 2
     30S ribosomal protein S2 OS=Acidiphilium cryptum (strain JF-5) GN=rpsB PE=3 SV=1

27.  PSAH2_ARATH    Mass: 15264    Score: 33     Expect: 2.5e+002  Matches: 2
     Photosystem I reaction center subunit VI-2, chloroplastic OS=Arabidopsis thaliana GN=PSAH2 PE=2 SV=1
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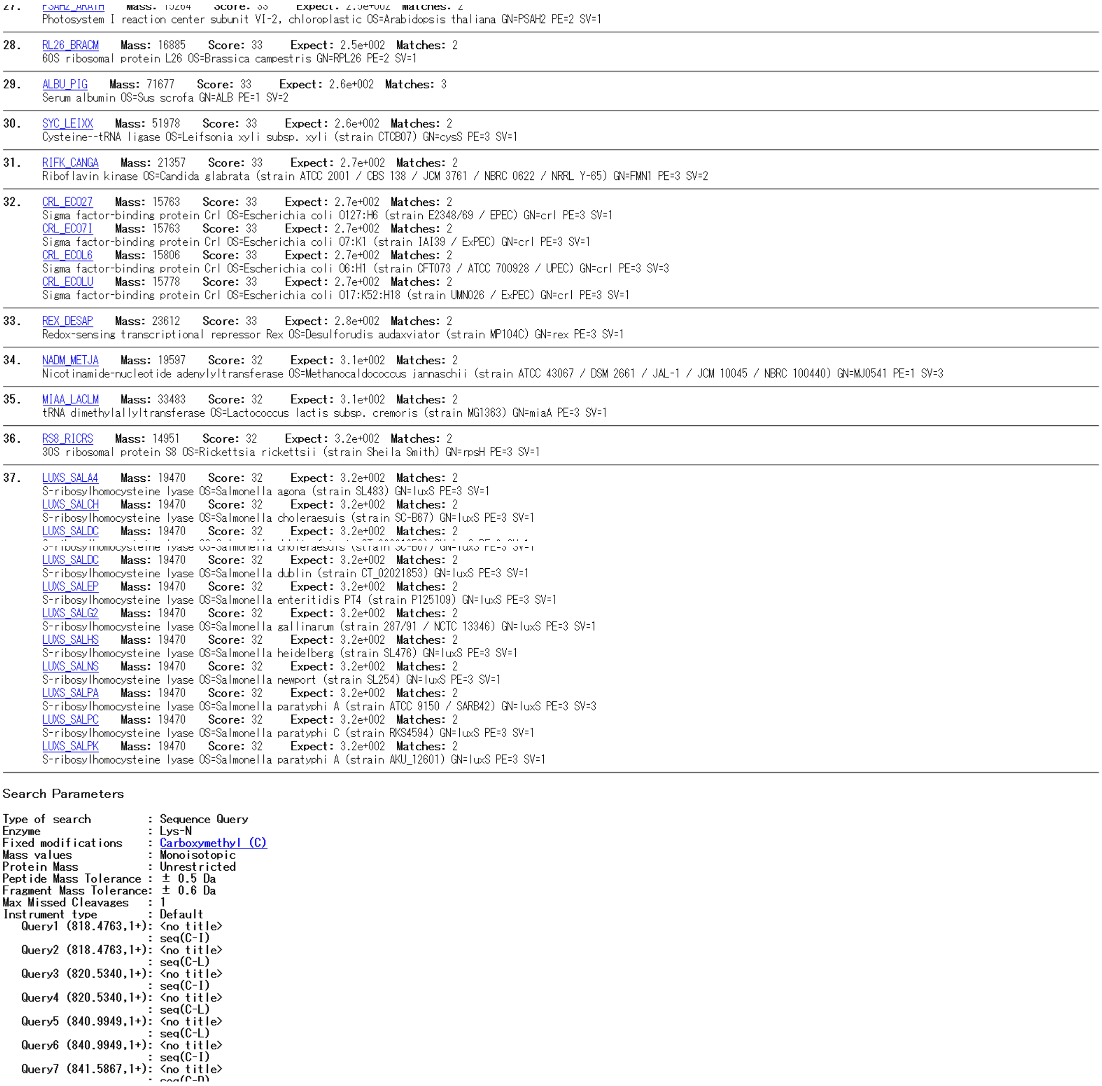

```
27.  PSAH2_ARATH   Mass: 15264   Score: 33    Expect: 2.5e+002  Matches: 2
     Photosystem I reaction center subunit VI-2, chloroplastic OS=Arabidopsis thaliana GN=PSAH2 PE=2 SV=1

28.  RL26_BRACM    Mass: 16885   Score: 33    Expect: 2.5e+002  Matches: 2
     60S ribosomal protein L26 OS=Brassica campestris GN=RPL26 PE=2 SV=1

29.  ALBU_PIG    Mass: 71677   Score: 33    Expect: 2.6e+002  Matches: 3
     Serum albumin OS=Sus scrofa GN=ALB PE=1 SV=2

30.  SYC_LEIXX    Mass: 51978   Score: 33    Expect: 2.6e+002  Matches: 2
     Cysteine--tRNA ligase OS=Leifsonia xyli subsp. xyli (strain CTCB07) GN=cysS PE=3 SV=1

31.  RIFK_CANGA    Mass: 21357   Score: 33    Expect: 2.7e+002  Matches: 2
     Riboflavin kinase OS=Candida glabrata (strain ATCC 2001 / CBS 138 / JCM 3761 / NBRC 0622 / NRRL Y-65) GN=FMN1 PE=3 SV=2

32.  CRL_ECO27    Mass: 15763   Score: 33    Expect: 2.7e+002  Matches: 2
     Sigma factor-binding protein Crl OS=Escherichia coli O127:H6 (strain E2348/69 / EPEC) GN=crl PE=3 SV=1
     CRL_ECO7I    Mass: 15763   Score: 33    Expect: 2.7e+002  Matches: 2
     Sigma factor-binding protein Crl OS=Escherichia coli O7:K1 (strain IAI39 / ExPEC) GN=crl PE=3 SV=1
     CRL_ECOL6    Mass: 15806   Score: 33    Expect: 2.7e+002  Matches: 2
     Sigma factor-binding protein Crl OS=Escherichia coli O6:H1 (strain CFT073 / ATCC 700928 / UPEC) GN=crl PE=3 SV=3
     CRL_ECOLU    Mass: 15778   Score: 33    Expect: 2.7e+002  Matches: 2
     Sigma factor-binding protein Crl OS=Escherichia coli O17:K52:H18 (strain UMN026 / ExPEC) GN=crl PE=3 SV=1

33.  REX_DESAP    Mass: 23612   Score: 33    Expect: 2.8e+002  Matches: 2
     Redox-sensing transcriptional repressor Rex OS=Desulforudis audaxviator (strain MP104C) GN=rex PE=3 SV=1

34.  NADM_METJA    Mass: 19597   Score: 32    Expect: 3.1e+002  Matches: 2
     Nicotinamide-nucleotide adenylyltransferase OS=Methanocaldococcus jannaschii (strain ATCC 43067 / DSM 2661 / JAL-1 / JCM 10045 / NBRC 100440) GN=MJ0541 PE=1 SV=3

35.  MIAA_LACLM    Mass: 33483   Score: 32    Expect: 3.1e+002  Matches: 2
     tRNA dimethylallyltransferase OS=Lactococcus lactis subsp. cremoris (strain MG1363) GN=miaA PE=3 SV=1

36.  RS8_RICRS    Mass: 14951   Score: 32    Expect: 3.2e+002  Matches: 2
     30S ribosomal protein S8 OS=Rickettsia rickettsii (strain Sheila Smith) GN=rpsH PE=3 SV=1

37.  LUXS_SALA4    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella agona (strain SL483) GN=luxS PE=3 SV=1
     LUXS_SALCH    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella choleraesuis (strain SC-B67) GN=luxS PE=3 SV=1
     LUXS_SALDC    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella choleraesuis (strain SC-B67) GN=luxS PE=3 SV=1
     LUXS_SALDC    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella dublin (strain CT_02021853) GN=luxS PE=3 SV=1
     LUXS_SALEP    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella enteritidis PT4 (strain P125109) GN=luxS PE=3 SV=1
     LUXS_SALG2    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella gallinarum (strain 287/91 / NCTC 13346) GN=luxS PE=3 SV=1
     LUXS_SALHS    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella heidelberg (strain SL476) GN=luxS PE=3 SV=1
     LUXS_SALNS    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella newport (strain SL254) GN=luxS PE=3 SV=1
     LUXS_SALPA    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella paratyphi A (strain ATCC 9150 / SARB42) GN=luxS PE=3 SV=3
     LUXS_SALPC    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella paratyphi C (strain RKS4594) GN=luxS PE=3 SV=1
     LUXS_SALPK    Mass: 19470   Score: 32    Expect: 3.2e+002  Matches: 2
     S-ribosylhomocysteine lyase OS=Salmonella paratyphi A (strain AKU_12601) GN=luxS PE=3 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
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   Query7 (841.5867,1+): <no title>
                       : seq(C-D)
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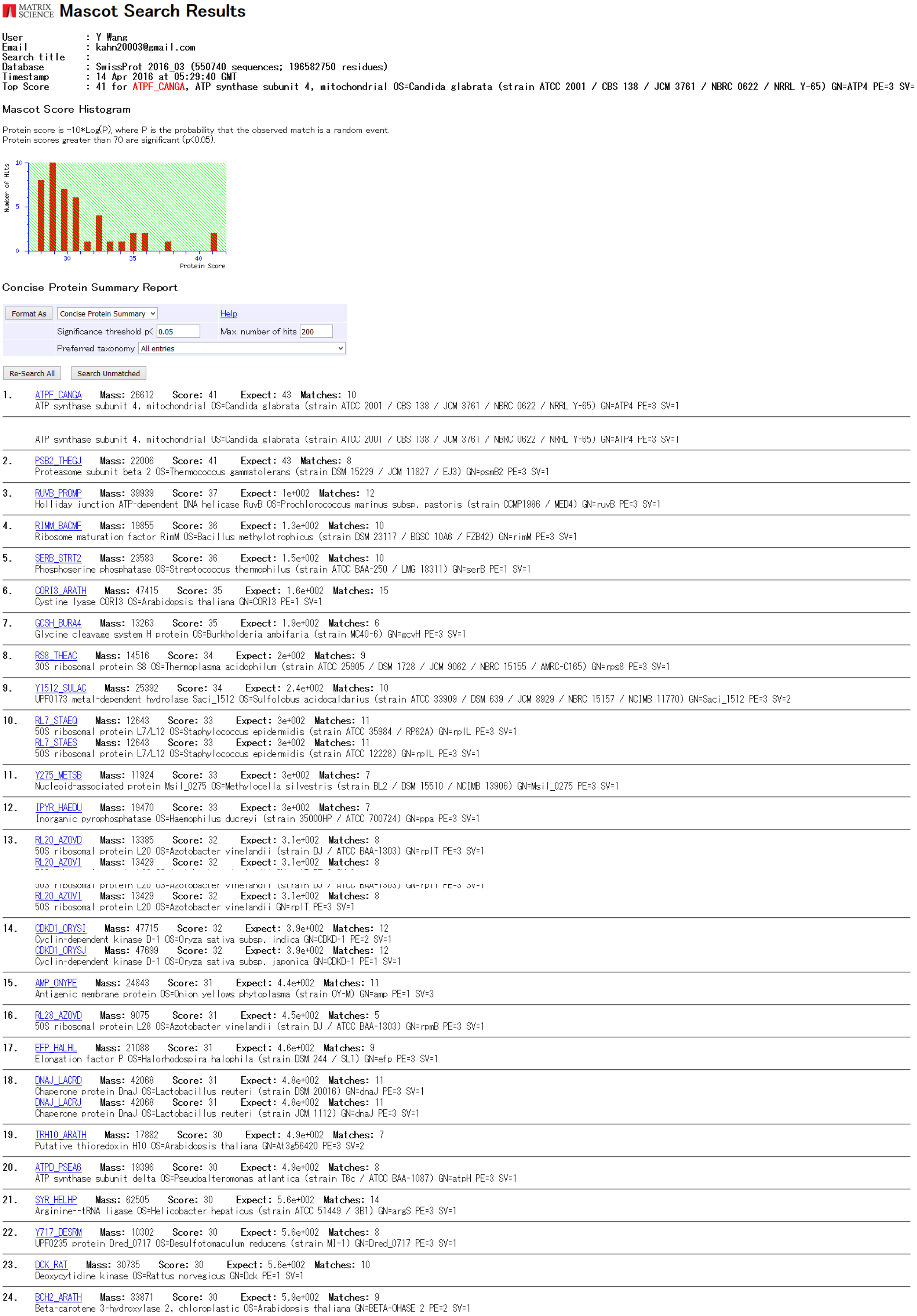
TF mass



Mascot Search Results

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User          : Y Wang
Email         : kahn20003@gmail.com
Search title  :
Database      : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp     : 14 Apr 2016 at 05:29:40 GMT
Top Score     : 41 for ATPF_CANGA, ATP synthase subunit 4, mitochondrial OS=Candida glabrata (strain ATCC 2001 / CBS 138 / JCM 3761 / NBRC 0622 / NRRL Y-65) GN=ATP4 PE=3 SV=
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Concise Protein Summary

Significance threshold p< 0.05

Max. number of hits 200

Preferred taxonomy All entries

```
1.   ATPF_CANGA   Mass: 26612   Score: 41   Expect: 43  Matches: 10
     ATP synthase subunit 4, mitochondrial OS=Candida glabrata (strain ATCC 2001 / CBS 138 / JCM 3761 / NBRC 0622 / NRRL Y-65) GN=ATP4 PE=3 SV=1
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ATP synthase subunit 4, mitochondrial OS=Candida glabrata (strain ATCC 2001 / CBS 138 / JCM 3761 / NBRC 0622 / NRRL Y-65) GN=ATP4 PE=3 SV=1

```
2.   PSB2_THEGJ   Mass: 22006   Score: 41   Expect: 43  Matches: 8
     Proteasome subunit beta 2 OS=Thermococcus gammatolerans (strain DSM 15229 / JCM 11827 / EJ3) GN=psmB2 PE=3 SV=1

3.   RUVB_PROMP   Mass: 39939   Score: 37   Expect: 1e+002  Matches: 12
     Holliday junction ATP-dependent DNA helicase RuvB OS=Prochlorococcus marinus subsp. pastoris (strain CCMP1986 / MED4) GN=ruvB PE=3 SV=1

4.   RIMM_BACMF   Mass: 19855   Score: 36   Expect: 1.3e+002  Matches: 10
     Ribosome maturation factor RimM OS=Bacillus methylotrophicus (strain DSM 23117 / BGSC 10A6 / FZB42) GN=rimM PE=3 SV=1

5.   SERB_STRT2   Mass: 23583   Score: 36   Expect: 1.5e+002  Matches: 10
     Phosphoserine phosphatase OS=Streptococcus thermophilus (strain ATCC BAA-250 / LMG 18311) GN=serB PE=1 SV=1

6.   CORI3_ARATH   Mass: 47415   Score: 35   Expect: 1.6e+002  Matches: 15
     Cystine lyase CORI3 OS=Arabidopsis thaliana GN=CORI3 PE=1 SV=1

7.   GCSH_BURA4   Mass: 13263   Score: 35   Expect: 1.9e+002  Matches: 6
     Glycine cleavage system H protein OS=Burkholderia ambifaria (strain MC40-6) GN=gcvH PE=3 SV=1

8.   RS8_THEAC   Mass: 14516   Score: 34   Expect: 2e+002  Matches: 9
     30S ribosomal protein S8 OS=Thermoplasma acidophilum (strain ATCC 25905 / DSM 1728 / JCM 9062 / NBRC 15155 / AMRC-C165) GN=rps8 PE=3 SV=1

9.   Y1512_SULAC   Mass: 25392   Score: 34   Expect: 2.4e+002  Matches: 10
     UPF0173 metal-dependent hydrolase Saci_1512 OS=Sulfolobus acidocaldarius (strain ATCC 33909 / DSM 639 / JCM 8929 / NBRC 15157 / NCIMB 11770) GN=Saci_1512 PE=3 SV=2

10.  RL7_STAEQ   Mass: 12643   Score: 33   Expect: 3e+002  Matches: 11
     50S ribosomal protein L7/L12 OS=Staphylococcus epidermidis (strain ATCC 35984 / RP62A) GN=rplL PE=3 SV=1
     RL7_STAES   Mass: 12643   Score: 33   Expect: 3e+002  Matches: 11
     50S ribosomal protein L7/L12 OS=Staphylococcus epidermidis (strain ATCC 12228) GN=rplL PE=3 SV=1

11.  Y275_METSB   Mass: 11924   Score: 33   Expect: 3e+002  Matches: 7
     Nucleoid-associated protein Msil_0275 OS=Methylocella silvestris (strain BL2 / DSM 15510 / NCIMB 13906) GN=Msil_0275 PE=3 SV=1

12.  IPYR_HAEDU   Mass: 19470   Score: 33   Expect: 3e+002  Matches: 7
     Inorganic pyrophosphatase OS=Haemophilus ducreyi (strain 35000HP / ATCC 700724) GN=ppa PE=3 SV=1

13.  RL20_AZOVD   Mass: 13385   Score: 32   Expect: 3.1e+002  Matches: 8
     50S ribosomal protein L20 OS=Azotobacter vinelandii (strain DJ / ATCC BAA-1303) GN=rplT PE=3 SV=1
     RL20_AZOVI   Mass: 13429   Score: 32   Expect: 3.1e+002  Matches: 8
     50S ribosomal protein L20 OS=Azotobacter vinelandii (strain DJ / ATCC BAA-1303) GN=rplT PE=3 SV=1
     RL20_AZOVI   Mass: 13429   Score: 32   Expect: 3.1e+002  Matches: 8
     50S ribosomal protein L20 OS=Azotobacter vinelandii GN=rplT PE=3 SV=1

14.  CDKD1_ORYSI   Mass: 47715   Score: 32   Expect: 3.9e+002  Matches: 12
     Cyclin-dependent kinase D-1 OS=Oryza sativa subsp. indica GN=CDKD-1 PE=2 SV=1
     CDKD1_ORYSJ   Mass: 47699   Score: 32   Expect: 3.9e+002  Matches: 12
     Cyclin-dependent kinase D-1 OS=Oryza sativa subsp. japonica GN=CDKD-1 PE=1 SV=1

15.  AMP_ONYPE   Mass: 24843   Score: 31   Expect: 4.4e+002  Matches: 11
     Antigenic membrane protein OS=Onion yellows phytoplasma (strain OY-M) GN=amp PE=1 SV=3

16.  RL28_AZOVD   Mass: 9075   Score: 31   Expect: 4.5e+002  Matches: 5
     50S ribosomal protein L28 OS=Azotobacter vinelandii (strain DJ / ATCC BAA-1303) GN=rpmB PE=3 SV=1

17.  EFP_HALHL   Mass: 21088   Score: 31   Expect: 4.6e+002  Matches: 9
     Elongation factor P OS=Halorhodospira halophila (strain DSM 244 / SL1) GN=efp PE=3 SV=1

18.  DNAJ_LACRD   Mass: 42068   Score: 31   Expect: 4.8e+002  Matches: 11
     Chaperone protein DnaJ OS=Lactobacillus reuteri (strain DSM 20016) GN=dnaJ PE=3 SV=1
     DNAJ_LACRJ   Mass: 42068   Score: 31   Expect: 4.8e+002  Matches: 11
     Chaperone protein DnaJ OS=Lactobacillus reuteri (strain JCM 1112) GN=dnaJ PE=3 SV=1

19.  TRH10_ARATH   Mass: 17882   Score: 30   Expect: 4.9e+002  Matches: 7
     Putative thioredoxin H10 OS=Arabidopsis thaliana GN=At3g56420 PE=3 SV=2

20.  ATPD_PSEA6   Mass: 19396   Score: 30   Expect: 4.9e+002  Matches: 8
     ATP synthase subunit delta OS=Pseudoalteromonas atlantica (strain T6c / ATCC BAA-1087) GN=atpH PE=3 SV=1

21.  SYR_HELHP   Mass: 62505   Score: 30   Expect: 5.6e+002  Matches: 14
     Arginine--tRNA ligase OS=Helicobacter hepaticus (strain ATCC 51449 / 3B1) GN=argS PE=3 SV=1

22.  Y717_DESRM   Mass: 10302   Score: 30   Expect: 5.6e+002  Matches: 8
     UPF0235 protein Dred_0717 OS=Desulfotomaculum reducens (strain MI-1) GN=Dred_0717 PE=3 SV=1

23.  DCK_RAT   Mass: 30735   Score: 30   Expect: 5.6e+002  Matches: 10
     Deoxycytidine kinase OS=Rattus norvegicus GN=Dck PE=1 SV=1

24.  BCH2_ARATH   Mass: 33871   Score: 30   Expect: 5.9e+002  Matches: 9
     Beta-carotene 3-hydroxylase 2, chloroplastic OS=Arabidopsis thaliana GN=BETA-OHASE 2 PE=2 SV=1
```

Beta-carotene 3-hydroxylase 2, chloroplastic OS=Arabidopsis thaliana GN=BETA-OHASE 2 PE=2 SV=1

25. RL14E_THESM **Mass:** 9004 **Score:** 30 **Expect:** 6e+002 **Matches:** 7
50S ribosomal protein L14e OS=Thermococcus sibiricus (strain MM 739 / DSM 12597) GN=rpl14e PE=3 SV=1

26. ZN727_HUMAN **Mass:** 60196 **Score:** 30 **Expect:** 6.2e+002 **Matches:** 19
Putative zinc finger protein 727 OS=Homo sapiens GN=ZNF727 PE=5 SV=3

27. PSAH_BRACM **Mass:** 15400 **Score:** 29 **Expect:** 6.3e+002 **Matches:** 7
Photosystem I reaction center subunit VI, chloroplastic OS=Brassica campestris GN=PSAH PE=2 SV=1

28. URE3_CHRSD **Mass:** 11020 **Score:** 29 **Expect:** 6.5e+002 **Matches:** 7
Urease subunit gamma OS=Chromohalobacter salexigens (strain DSM 3043 / ATCC BAA-138 / NCIMB 13768) GN=ureA PE=3 SV=1

29. RL6_MANSM **Mass:** 19027 **Score:** 29 **Expect:** 6.6e+002 **Matches:** 7
50S ribosomal protein L6 OS=Mannheimia succiniciproducens (strain MBEL55E) GN=rplF PE=3 SV=1

30. MINE_NITEU **Mass:** 11560 **Score:** 29 **Expect:** 6.9e+002 **Matches:** 6
Cell division topological specificity factor OS=Nitrosomonas europaea (strain ATCC 19718 / NBRC 14298) GN=minE PE=3 SV=1

31. HIS6_BACLD **Mass:** 27423 **Score:** 29 **Expect:** 6.9e+002 **Matches:** 8
Imidazole glycerol phosphate synthase subunit HisF OS=Bacillus licheniformis (strain ATCC 14580 / DSM 13 / JCM 2505 / NBRC 12200 / NCIMB 9375 / NRRL NRS-1264 / Gibson 46) GN=hisF PE=3 SV=1

32. Y2210_ARCFU **Mass:** 8957 **Score:** 29 **Expect:** 6.9e+002 **Matches:** 6
Uncharacterized protein AF_2210 OS=Archaeoglobus fulgidus (strain ATCC 49558 / VC-16 / DSM 4304 / JCM 9628 / NBRC 100126) GN=AF_2210 PE=4 SV=1

33. ADH3_GEOSE **Mass:** 36837 **Score:** 29 **Expect:** 7.3e+002 **Matches:** 11
Alcohol dehydrogenase OS=Geobacillus stearothermophilus PE=1 SV=1

34. URE3_TOLAT **Mass:** 11055 **Score:** 29 **Expect:** 7.6e+002 **Matches:** 8
Urease subunit gamma OS=Tolumonas auensis (strain DSM 9187 / TA4) GN=ureA PE=3 SV=1

35. ATPD_HYPNA **Mass:** 19615 **Score:** 28 **Expect:** 7.8e+002 **Matches:** 8
ATP synthase subunit delta OS=Hyphomonas neptunium (strain ATCC 15444) GN=atpH PE=3 SV=1

36. AURKB_DANRE **Mass:** 37213 **Score:** 28 **Expect:** 7.8e+002 **Matches:** 8
Aurora kinase B OS=Danio rerio GN=aurkb PE=2 SV=1

37. IHFA_PSYIN **Mass:** 11032 **Score:** 28 **Expect:** 7.8e+002 **Matches:** 8
Integration host factor subunit alpha OS=Psychromonas ingrahamii (strain 37) GN=ihfA PE=3 SV=1

37. IHFA_PSYIN **Mass:** 11032 **Score:** 28 **Expect:** 7.8e+002 **Matches:** 8
Integration host factor subunit alpha OS=Psychromonas ingrahamii (strain 37) GN=ihfA PE=3 SV=1

38. KAR_YEAST **Mass:** 35596 **Score:** 28 **Expect:** 8.1e+002 **Matches:** 11
NADPH-dependent alpha-keto amide reductase OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=YDL124W PE=1 SV=1

39. RASF4_MOUSE **Mass:** 37201 **Score:** 28 **Expect:** 8.1e+002 **Matches:** 10
Ras association domain-containing protein 4 OS=Mus musculus GN=Rassf4 PE=1 SV=1

40. MSD4_MOUSE **Mass:** 41279 **Score:** 28 **Expect:** 8.1e+002 **Matches:** 11
Myb/SANT-like DNA-binding domain-containing protein 4 OS=Mus musculus GN=Msantd4 PE=2 SV=1
MSD4_RAT **Mass:** 41265 **Score:** 28 **Expect:** 8.1e+002 **Matches:** 11
Myb/SANT-like DNA-binding domain-containing protein 4 OS=Rattus norvegicus GN=Msantd4 PE=1 SV=1

41. MUCEN_RAT **Mass:** 26616 **Score:** 28 **Expect:** 8.1e+002 **Matches:** 8
Endomucin OS=Rattus norvegicus GN=Emcn PE=2 SV=1

42. GCST_CLOBJ **Mass:** 41806 **Score:** 28 **Expect:** 8.3e+002 **Matches:** 13
Aminomethyltransferase OS=Clostridium botulinum (strain Kyoto / Type A2) GN=gcvT PE=3 SV=1

43. CWC26_KLULA **Mass:** 30726 **Score:** 28 **Expect:** 8.7e+002 **Matches:** 14
Pre-mRNA-splicing factor CWC26 OS=Kluyveromyces lactis (strain ATCC 8585 / CBS 2359 / DSM 70799 / NBRC 1267 / NRRL Y-1140 / WM37) GN=CWC26 PE=3 SV=1

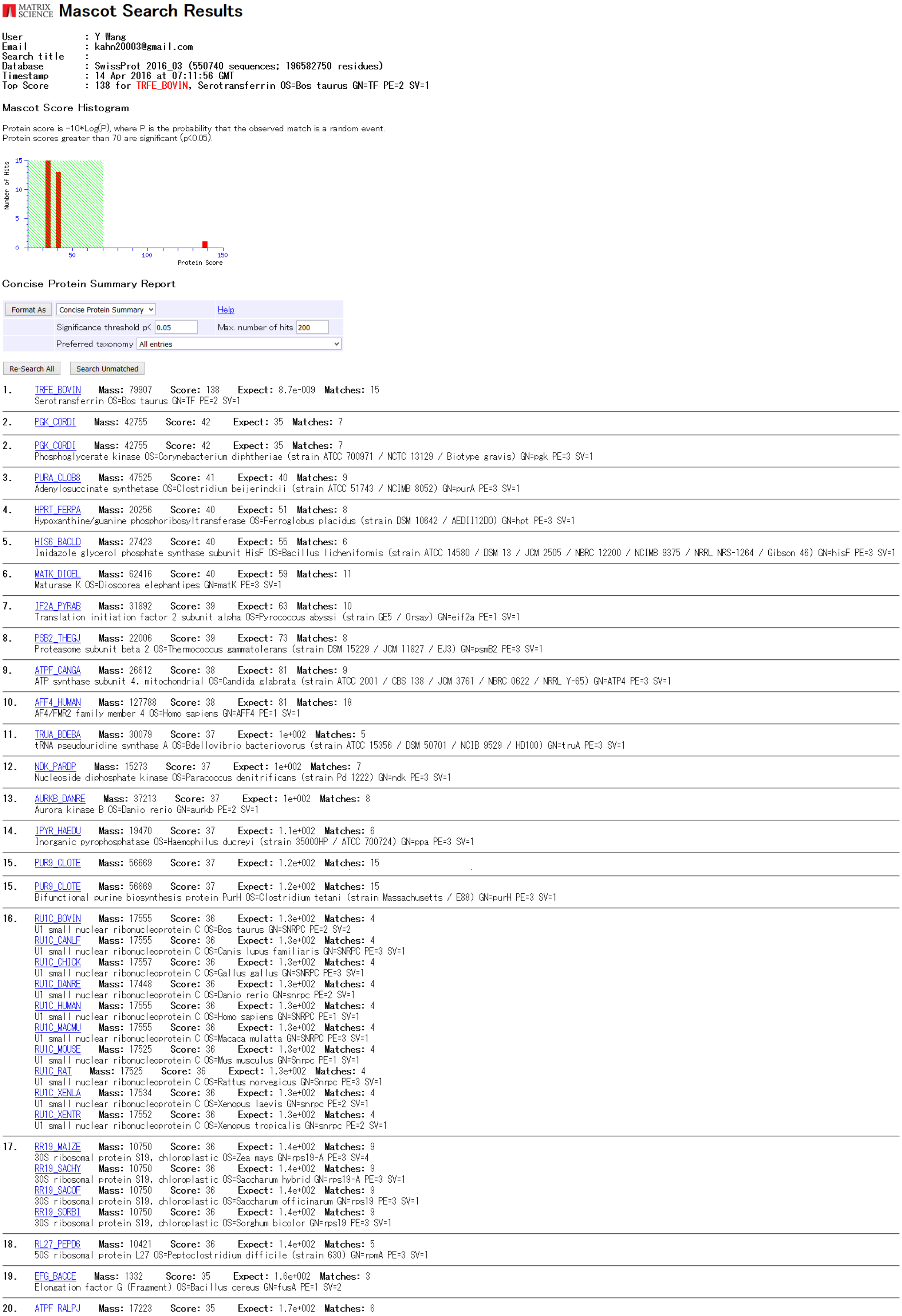
44. RRP1_DICDI **Mass:** 57004 **Score:** 28 **Expect:** 8.7e+002 **Matches:** 17
Ribosomal RNA processing protein 1 homolog OS=Dictyostelium discoideum GN=rrp1 PE=3 SV=1

45. TEX30_MOUSE **Mass:** 25595 **Score:** 28 **Expect:** 8.7e+002 **Matches:** 10
Testis-expressed sequence 30 protein OS=Mus musculus GN=Tex30 PE=1 SV=1

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
```

TF mass/mass+c



Mascot Search Results

User : Y Wang
Email : kahn20003@gmail.com
Search title :
Database : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp : 14 Apr 2016 at 07:11:56 GMT
Top Score : 138 for TRFE_BOVIN, Serotransferrin OS=Bos taurus GN=TF PE=2 SV=1

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

1. **TRFE_BOVIN** **Mass:** 79907 **Score:** 138 **Expect:** 8.7e-009 **Matches:** 15
Serotransferrin OS=Bos taurus GN=TF PE=2 SV=1

2. **PGK_CORDI** **Mass:** 42755 **Score:** 42 **Expect:** 35 **Matches:** 7

2. **PGK_CORDI** **Mass:** 42755 **Score:** 42 **Expect:** 35 **Matches:** 7
Phosphoglycerate kinase OS=Corynebacterium diphtheriae (strain ATCC 700971 / NCTC 13129 / Biotype gravis) GN=pgk PE=3 SV=1

3. **PURA_CLOB8** **Mass:** 47525 **Score:** 41 **Expect:** 40 **Matches:** 9
Adenylosuccinate synthetase OS=Clostridium beijerinckii (strain ATCC 51743 / NCIMB 8052) GN=purA PE=3 SV=1

4. **HPRT_FERPA** **Mass:** 20256 **Score:** 40 **Expect:** 51 **Matches:** 8
Hypoxanthine/guanine phosphoribosyltransferase OS=Ferroglobus placidus (strain DSM 10642 / AEDII12DO) GN=hpt PE=3 SV=1

5. **HIS6_BACLD** **Mass:** 27423 **Score:** 40 **Expect:** 55 **Matches:** 6
Imidazole glycerol phosphate synthase subunit HisF OS=Bacillus licheniformis (strain ATCC 14580 / DSM 13 / JCM 2505 / NBRC 12200 / NCIMB 9375 / NRRL NRS-1264 / Gibson 46) GN=hisF PE=3 SV=1

6. **MATK_DIOEL** **Mass:** 62416 **Score:** 40 **Expect:** 59 **Matches:** 11
Maturase K OS=Dioscorea elephantipes GN=matK PE=3 SV=1

7. **IF2A_PYRAB** **Mass:** 31892 **Score:** 39 **Expect:** 63 **Matches:** 10
Translation initiation factor 2 subunit alpha OS=Pyrococcus abyssi (strain GE5 / Orsay) GN=eif2a PE=1 SV=1

8. **PSB2_THEGJ** **Mass:** 22006 **Score:** 39 **Expect:** 73 **Matches:** 8
Proteasome subunit beta 2 OS=Thermococcus gammatolerans (strain DSM 15229 / JCM 11827 / EJ3) GN=psmB2 PE=3 SV=1

9. **ATPF_CANGA** **Mass:** 26612 **Score:** 38 **Expect:** 81 **Matches:** 9
ATP synthase subunit 4, mitochondrial OS=Candida glabrata (strain ATCC 2001 / CBS 138 / JCM 3761 / NBRC 0622 / NRRL Y-65) GN=ATP4 PE=3 SV=1

10. **AFF4_HUMAN** **Mass:** 127788 **Score:** 38 **Expect:** 81 **Matches:** 18
AF4/FMR2 family member 4 OS=Homo sapiens GN=AFF4 PE=1 SV=1

11. **TRUA_BDEBA** **Mass:** 30079 **Score:** 37 **Expect:** 1e+002 **Matches:** 5
tRNA pseudouridine synthase A OS=Bdellovibrio bacteriovorus (strain ATCC 15356 / DSM 50701 / NCIB 9529 / HD100) GN=truA PE=3 SV=1

12. **NDK_PARDP** **Mass:** 15273 **Score:** 37 **Expect:** 1e+002 **Matches:** 7
Nucleoside diphosphate kinase OS=Paracoccus denitrificans (strain Pd 1222) GN=ndk PE=3 SV=1

13. **AURKB_DANRE** **Mass:** 37213 **Score:** 37 **Expect:** 1e+002 **Matches:** 8
Aurora kinase B OS=Danio rerio GN=aurkb PE=2 SV=1

14. **IPYR_HAEDU** **Mass:** 19470 **Score:** 37 **Expect:** 1.1e+002 **Matches:** 6
Inorganic pyrophosphatase OS=Haemophilus ducreyi (strain 35000HP / ATCC 700724) GN=ppa PE=3 SV=1

15. **PUR9_CLOTE** **Mass:** 56669 **Score:** 37 **Expect:** 1.2e+002 **Matches:** 15

15. **PUR9_CLOTE** **Mass:** 56669 **Score:** 37 **Expect:** 1.2e+002 **Matches:** 15
Bifunctional purine biosynthesis protein PurH OS=Clostridium tetani (strain Massachusetts / E88) GN=purH PE=3 SV=1

16. **RU1C_BOVIN** **Mass:** 17555 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Bos taurus GN=SNRPC PE=2 SV=2
RU1C_CANLF **Mass:** 17555 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Canis lupus familiaris GN=SNRPC PE=3 SV=1
RU1C_CHICK **Mass:** 17557 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Gallus gallus GN=SNRPC PE=3 SV=1
RU1C_DANRE **Mass:** 17448 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Danio rerio GN=snrpc PE=2 SV=1
RU1C_HUMAN **Mass:** 17555 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Homo sapiens GN=SNRPC PE=1 SV=1
RU1C_MACMU **Mass:** 17555 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Macaca mulatta GN=SNRPC PE=3 SV=1
RU1C_MOUSE **Mass:** 17525 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Mus musculus GN=Snrpc PE=1 SV=1
RU1C_RAT **Mass:** 17525 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Rattus norvegicus GN=Snrpc PE=3 SV=1
RU1C_XENLA **Mass:** 17534 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Xenopus laevis GN=snrpc PE=2 SV=1
RU1C_XENTR **Mass:** 17552 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 4
U1 small nuclear ribonucleoprotein C OS=Xenopus tropicalis GN=snrpc PE=2 SV=1

17. **RR19_MAIZE** **Mass:** 10750 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 9
30S ribosomal protein S19, chloroplastic OS=Zea mays GN=rps19-A PE=3 SV=4
RR19_SACHY **Mass:** 10750 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 9
30S ribosomal protein S19, chloroplastic OS=Saccharum hybrid GN=rps19-A PE=3 SV=1
RR19_SACOF **Mass:** 10750 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 9
30S ribosomal protein S19, chloroplastic OS=Saccharum officinarum GN=rps19 PE=3 SV=1
RR19_SORBI **Mass:** 10750 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 9
30S ribosomal protein S19, chloroplastic OS=Sorghum bicolor GN=rps19 PE=3 SV=1

18. **RL27_PEPD6** **Mass:** 10421 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 5
50S ribosomal protein L27 OS=Peptoclostridium difficile (strain 630) GN=rpmA PE=3 SV=1

19. **EFG_BACCE** **Mass:** 1332 **Score:** 35 **Expect:** 1.6e+002 **Matches:** 3
Elongation factor G (Fragment) OS=Bacillus cereus GN=fusA PE=1 SV=2

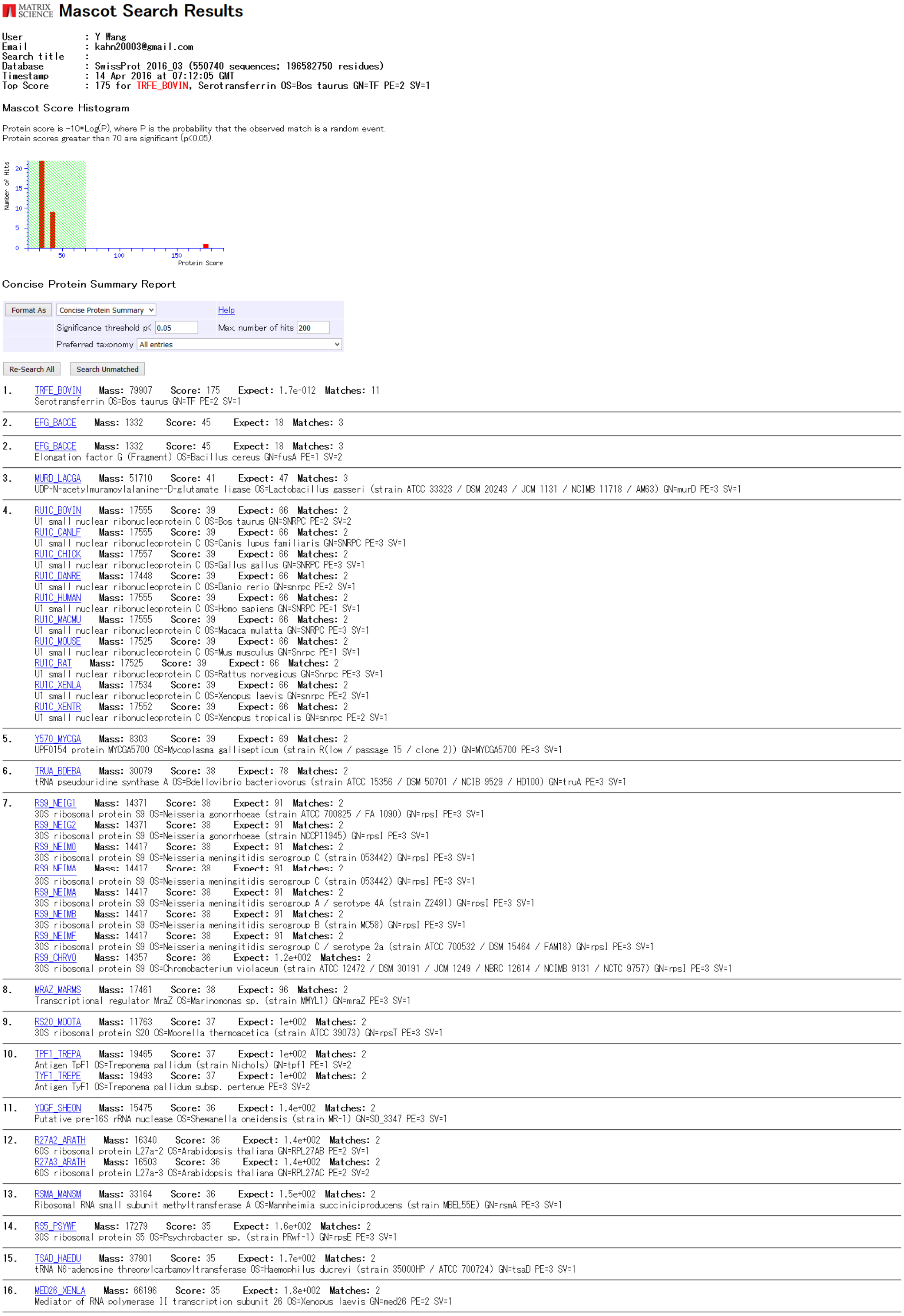
20. **ATPF RALPJ** **Mass:** 17223 **Score:** 35 **Expect:** 1.7e+002 **Matches:** 6

```
20.   ATPF_RALPJ    Mass: 17223    Score: 35     Expect: 1.7e+002  Matches: 6
      ATP synthase subunit b OS=Ralstonia pickettii (strain 12J) GN=atpF PE=3 SV=1

21.   Y010_OSHVF    Mass: 39543    Score: 35     Expect: 1.9e+002  Matches: 8
      Uncharacterized protein ORF10 OS=Ostreid herpesvirus 1 (isolate France) GN=ORF10 PE=4 SV=1

22.   RS18_CLOB1    Mass: 9513     Score: 35     Expect: 1.9e+002  Matches: 6
      30S ribosomal protein S18 OS=Clostridium botulinum (strain ATCC 19397 / Type A) GN=rpsR PE=3 SV=1
      RS18_CLOB6    Mass: 9513     Score: 35     Expect: 1.9e+002  Matches: 6
      30S ribosomal protein S18 OS=Clostridium botulinum (strain 657 / Type Ba4) GN=rpsR PE=3 SV=1
      RS18_CLOBH    Mass: 9513     Score: 35     Expect: 1.9e+002  Matches: 6
      30S ribosomal protein S18 OS=Clostridium botulinum (strain Hall / ATCC 3502 / NCTC 13319 / Type A) GN=rpsR PE=3 SV=1
      RS18_CLOBJ    Mass: 9513     Score: 35     Expect: 1.9e+002  Matches: 6
      30S ribosomal protein S18 OS=Clostridium botulinum (strain Kyoto / Type A2) GN=rpsR PE=3 SV=1
      RS18_CLOBK    Mass: 9513     Score: 35     Expect: 1.9e+002  Matches: 6
      30S ribosomal protein S18 OS=Clostridium botulinum (strain Okra / Type B1) GN=rpsR PE=3 SV=1
      RS18_CLOBL    Mass: 9513     Score: 35     Expect: 1.9e+002  Matches: 6
      30S ribosomal protein S18 OS=Clostridium botulinum (strain Langeland / NCTC 10281 / Type F) GN=rpsR PE=3 SV=1
      RS18_CLOBM    Mass: 9513     Score: 35     Expect: 1.9e+002  Matches: 6
      30S ribosomal protein S18 OS=Clostridium botulinum (strain Loch Maree / Type A3) GN=rpsR PE=3 SV=1

23.   Y570_MYCGA    Mass: 8303     Score: 34     Expect: 2e+002  Matches: 3
      UPF0154 protein MYCGA5700 OS=Mycoplasma gallisepticum (strain R(low / passage 15 / clone 2)) GN=MYCGA5700 PE=3 SV=1

24.   RS20_MOOTA    Mass: 11763    Score: 34     Expect: 2e+002  Matches: 6
      30S ribosomal protein S20 OS=Moorella thermoacetica (strain ATCC 39073) GN=rpsT PE=3 SV=1

25.   RL9_BUCA5    Mass: 16648    Score: 34     Expect: 2e+002  Matches: 8
      50S ribosomal protein L9 OS=Buchnera aphidicola subsp. Acyrthosiphon pisum (strain 5A) GN=rplI PE=3 SV=1
      RL9_BUCAI    Mass: 16648    Score: 34     Expect: 2e+002  Matches: 8
      50S ribosomal protein L9 OS=Buchnera aphidicola subsp. Acyrthosiphon pisum (strain APS) GN=rplI PE=3 SV=1
      RL9_BUCAT    Mass: 16648    Score: 34     Expect: 2e+002  Matches: 8
      50S ribosomal protein L9 OS=Buchnera aphidicola subsp. Acyrthosiphon pisum (strain Tuc7) GN=rplI PE=3 SV=1

26.   PROQ_HAEDU    Mass: 20342    Score: 34     Expect: 2.1e+002  Matches: 7
      RNA chaperone ProQ OS=Haemophilus ducreyi (strain 35000HP / ATCC 700724) GN=proQ PE=3 SV=1

27.   R27A2_ARATH    Mass: 16340    Score: 34     Expect: 2.4e+002  Matches: 4
      60S ribosomal protein L27a-2 OS=Arabidopsis thaliana GN=RPL27AB PE=2 SV=1
      R27A3_ARATH    Mass: 16503    Score: 34     Expect: 2.4e+002  Matches: 4
      60S ribosomal protein L27a-2 OS=Arabidopsis thaliana GN=RPL27AB PE=2 SV=1
      R27A3_ARATH    Mass: 16503    Score: 34     Expect: 2.4e+002  Matches: 4
      60S ribosomal protein L27a-3 OS=Arabidopsis thaliana GN=RPL27AC PE=2 SV=2

28.   HIS6_GEOSW    Mass: 27361    Score: 34     Expect: 2.4e+002  Matches: 5
      Imidazole glycerol phosphate synthase subunit HisF OS=Geobacillus sp. (strain WCH70) GN=hisF PE=3 SV=1

29.   BT5_ARATH    Mass: 43098    Score: 34     Expect: 2.5e+002  Matches: 7
      BTB/POZ and TAZ domain-containing protein 5 OS=Arabidopsis thaliana GN=BT5 PE=1 SV=1
```

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (825.6694,1+): <no title>
   Query2 (829.7115,1+): <no title>
                       : seq(C-G)
   Query3 (832.5867,1+): <no title>
   Query4 (837.5821,1+): <no title>
                       : seq(C-G)
   Query5 (846.6149,1+): <no title>
   Query6 (849.5739,1+): <no title>
   Query7 (851.6666,1+): <no title>
   Query8 (852.6093,1+): <no title>
                       : seq(C-I)
   Query9 (852.6093,1+): <no title>
                       : seq(C-L)
  Query10 (857.6361,1+): <no title>
  Query11 (857.6544,1+): <no title>
  Query12 (861.2213,1+): <no title>
  Query13 (861.2609,1+): <no title>
  Query14 (861.2654,1+): <no title>
  Query15 (869.6453,1+): <no title>
  Query16 (877.2189,1+): <no title>
  Query17 (887.6663,1+): <no title>
                       : seq(C-G)
```

TF mass+c



Mascot Search Results

```
User         : Y Wang
Email        : kahn20003@gmail.com
Search title :
Database     : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp    : 14 Apr 2016 at 07:12:05 GMT
Top Score    : 175 for TRFE_BOVIN, Serotransferrin OS=Bos taurus GN=TF PE=2 SV=1
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Concise Protein Summary Help
Significance threshold p< 0.05 Max. number of hits 200
Preferred taxonomy All entries

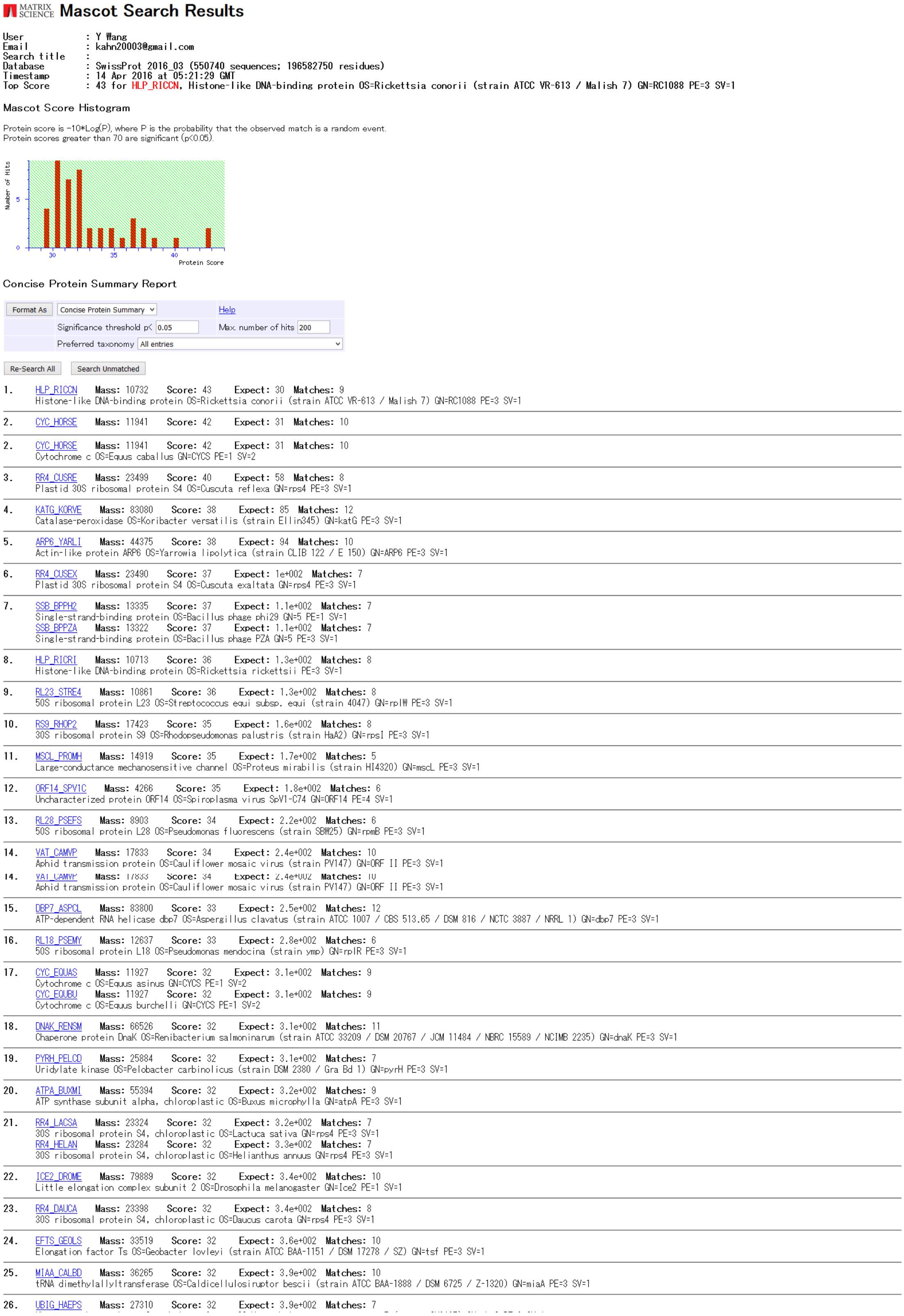
```
1.   TRFE_BOVIN    Mass: 79907   Score: 175   Expect: 1.7e-012  Matches: 11
     Serotransferrin OS=Bos taurus GN=TF PE=2 SV=1

2.   EFG_BACCE     Mass: 1332    Score: 45    Expect: 18  Matches: 3

2.   EFG_BACCE     Mass: 1332    Score: 45    Expect: 18  Matches: 3
     Elongation factor G (Fragment) OS=Bacillus cereus GN=fusA PE=1 SV=2

3.   MURD_LACGA    Mass: 51710   Score: 41    Expect: 47  Matches: 3
     UDP-N-acetylmuramoylalanine--D-glutamate ligase OS=Lactobacillus gasseri (strain ATCC 33323 / DSM 20243 / JCM 1131 / NCIMB 11718 / AM63) GN=murD PE=3 SV=1

4.   RU1C_BOVIN    Mass: 17555   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Bos taurus GN=SNRPC PE=2 SV=2
     RU1C_CANLF    Mass: 17555   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Canis lupus familiaris GN=SNRPC PE=3 SV=1
     RU1C_CHICK    Mass: 17557   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Gallus gallus GN=SNRPC PE=3 SV=1
     RU1C_DANRE    Mass: 17448   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Danio rerio GN=snrpc PE=2 SV=1
     RU1C_HUMAN    Mass: 17555   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Homo sapiens GN=SNRPC PE=1 SV=1
     RU1C_MACMU    Mass: 17555   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Macaca mulatta GN=SNRPC PE=3 SV=1
     RU1C_MOUSE    Mass: 17525   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Mus musculus GN=Snrpc PE=1 SV=1
     RU1C_RAT    Mass: 17525   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Rattus norvegicus GN=Snrpc PE=3 SV=1
     RU1C_XENLA    Mass: 17534   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Xenopus laevis GN=snrpc PE=2 SV=1
     RU1C_XENTR    Mass: 17552   Score: 39    Expect: 66  Matches: 2
     U1 small nuclear ribonucleoprotein C OS=Xenopus tropicalis GN=snrpc PE=2 SV=1

5.   Y570_MYCGA    Mass: 8303    Score: 39    Expect: 69  Matches: 2
     UPF0154 protein MYCGA5700 OS=Mycoplasma gallisepticum (strain R(low / passage 15 / clone 2)) GN=MYCGA5700 PE=3 SV=1

6.   TRUA_BDEBA    Mass: 30079   Score: 38    Expect: 78  Matches: 2
     tRNA pseudouridine synthase A OS=Bdellovibrio bacteriovorus (strain ATCC 15356 / DSM 50701 / NCIB 9529 / HD100) GN=truA PE=3 SV=1

7.   RS9_NEIG1    Mass: 14371   Score: 38    Expect: 91  Matches: 2
     30S ribosomal protein S9 OS=Neisseria gonorrhoeae (strain ATCC 700825 / FA 1090) GN=rpsI PE=3 SV=1
     RS9_NEIG2    Mass: 14371   Score: 38    Expect: 91  Matches: 2
     30S ribosomal protein S9 OS=Neisseria gonorrhoeae (strain NCCP11945) GN=rpsI PE=3 SV=1
     RS9_NEIMO    Mass: 14417   Score: 38    Expect: 91  Matches: 2
     30S ribosomal protein S9 OS=Neisseria meningitidis serogroup C (strain 053442) GN=rpsI PE=3 SV=1
     RS9_NEIMA    Mass: 14417   Score: 38    Expect: 91  Matches: 2
     30S ribosomal protein S9 OS=Neisseria meningitidis serogroup C (strain 053442) GN=rpsI PE=3 SV=1
     RS9_NEIMA    Mass: 14417   Score: 38    Expect: 91  Matches: 2
     30S ribosomal protein S9 OS=Neisseria meningitidis serogroup A / serotype 4A (strain Z2491) GN=rpsI PE=3 SV=1
     RS9_NEIMB    Mass: 14417   Score: 38    Expect: 91  Matches: 2
     30S ribosomal protein S9 OS=Neisseria meningitidis serogroup B (strain MC58) GN=rpsI PE=3 SV=1
     RS9_NEIMF    Mass: 14417   Score: 38    Expect: 91  Matches: 2
     30S ribosomal protein S9 OS=Neisseria meningitidis serogroup C / serotype 2a (strain ATCC 700532 / DSM 15464 / FAM18) GN=rpsI PE=3 SV=1
     RS9_CHRVO    Mass: 14357   Score: 36    Expect: 1.2e+002  Matches: 2
     30S ribosomal protein S9 OS=Chromobacterium violaceum (strain ATCC 12472 / DSM 30191 / JCM 1249 / NBRC 12614 / NCIMB 9131 / NCTC 9757) GN=rpsI PE=3 SV=1

8.   MRAZ_MARMS    Mass: 17461   Score: 38    Expect: 96  Matches: 2
     Transcriptional regulator MraZ OS=Marinomonas sp. (strain MWYL1) GN=mraZ PE=3 SV=1

9.   RS20_MOOTA    Mass: 11763   Score: 37    Expect: 1e+002  Matches: 2
     30S ribosomal protein S20 OS=Moorella thermoacetica (strain ATCC 39073) GN=rpsT PE=3 SV=1

10.  TPF1_TREPA    Mass: 19465   Score: 37    Expect: 1e+002  Matches: 2
     Antigen TpF1 OS=Treponema pallidum (strain Nichols) GN=tpf1 PE=1 SV=2
     TYF1_TREPE    Mass: 19493   Score: 37    Expect: 1e+002  Matches: 2
     Antigen TyF1 OS=Treponema pallidum subsp. pertenue PE=3 SV=2

11.  YQGF_SHEON    Mass: 15475   Score: 36    Expect: 1.4e+002  Matches: 2
     Putative pre-16S rRNA nuclease OS=Shewanella oneidensis (strain MR-1) GN=SO_3347 PE=3 SV=1

12.  R27A2_ARATH    Mass: 16340   Score: 36    Expect: 1.4e+002  Matches: 2
     60S ribosomal protein L27a-2 OS=Arabidopsis thaliana GN=RPL27AB PE=2 SV=1
     R27A3_ARATH    Mass: 16503   Score: 36    Expect: 1.4e+002  Matches: 2
     60S ribosomal protein L27a-3 OS=Arabidopsis thaliana GN=RPL27AC PE=2 SV=2

13.  RSMA_MANSM    Mass: 33164   Score: 36    Expect: 1.5e+002  Matches: 2
     Ribosomal RNA small subunit methyltransferase A OS=Mannheimia succiniciproducens (strain MBEL55E) GN=rsmA PE=3 SV=1

14.  RS5_PSYWF    Mass: 17279   Score: 35    Expect: 1.6e+002  Matches: 2
     30S ribosomal protein S5 OS=Psychrobacter sp. (strain PRwf-1) GN=rpsE PE=3 SV=1

15.  TSAD_HAEDU    Mass: 37901   Score: 35    Expect: 1.7e+002  Matches: 2
     tRNA N6-adenosine threonylcarbamoyltransferase OS=Haemophilus ducreyi (strain 35000HP / ATCC 700724) GN=tsaD PE=3 SV=1

16.  MED26_XENLA    Mass: 66196   Score: 35    Expect: 1.8e+002  Matches: 2
     Mediator of RNA polymerase II transcription subunit 26 OS=Xenopus laevis GN=med26 PE=2 SV=1
```

```
     Mediator of RNA polymerase II transcription subunit 26 OS=Xenopus laevis GN=med26 PE=2 SV=1

17.  AOFB_MOUSE    Mass: 59042    Score: 35    Expect: 1.9e+002  Matches: 2
     Amine oxidase [flavin-containing] B OS=Mus musculus GN=Maob PE=1 SV=4

18.  PGK_CORDI    Mass: 42755    Score: 34    Expect: 2e+002  Matches: 2
     Phosphoglycerate kinase OS=Corynebacterium diphtheriae (strain ATCC 700971 / NCTC 13129 / Biotype gravis) GN=pgk PE=3 SV=1

19.  ES1_RAT    Mass: 28503    Score: 34    Expect: 2e+002  Matches: 2
     ES1 protein homolog, mitochondrial OS=Rattus norvegicus PE=1 SV=2

20.  CP2E1_CANLF    Mass: 56853    Score: 34    Expect: 2.3e+002  Matches: 2
     Cytochrome P450 2E1 OS=Canis lupus familiaris GN=CYP2E1 PE=2 SV=1

21.  CYC2_AFIMA    Mass: 13636    Score: 34    Expect: 2.5e+002  Matches: 2
     Cytochrome c2 OS=Afifella marina PE=1 SV=1

22.  GUAA_SODGM    Mass: 58973    Score: 33    Expect: 2.6e+002  Matches: 2
     GMP synthase [glutamine-hydrolyzing] OS=Sodalis glossinidius (strain morsitans) GN=guaA PE=3 SV=1
     GUAA_CROS8    Mass: 59084    Score: 32    Expect: 3.1e+002  Matches: 2
     GMP synthase [glutamine-hydrolyzing] OS=Cronobacter sakazakii (strain ATCC BAA-894) GN=guaA PE=3 SV=1

23.  BT5_ARATH    Mass: 43098    Score: 33    Expect: 2.7e+002  Matches: 2
     BTB/POZ and TAZ domain-containing protein 5 OS=Arabidopsis thaliana GN=BT5 PE=1 SV=1

24.  KHSE_CORDI    Mass: 32551    Score: 33    Expect: 2.7e+002  Matches: 2
     Homoserine kinase OS=Corynebacterium diphtheriae (strain ATCC 700971 / NCTC 13129 / Biotype gravis) GN=thrB PE=3 SV=1

25.  MATK_ZINOF    Mass: 62479    Score: 33    Expect: 2.8e+002  Matches: 2
     Maturase K OS=Zingiber officinale GN=matK PE=3 SV=1

26.  EFP_PSYCK    Mass: 21006    Score: 33    Expect: 2.8e+002  Matches: 2
     Elongation factor P OS=Psychrobacter cryohalolentis (strain K5) GN=efp PE=3 SV=1

27.  HIS6_BACLD    Mass: 27423    Score: 33    Expect: 3e+002  Matches: 2
     Imidazole glycerol phosphate synthase subunit HisF OS=Bacillus licheniformis (strain ATCC 14580 / DSM 13 / JCM 2505 / NBRC 12200 / NCIMB 9375 / NRRL NRS-1264 / Gibson 46) GN=hisF PE=3 SV=1

28.  PFA5_KLULA    Mass: 41222    Score: 33    Expect: 3e+002  Matches: 2
     Palmitoyltransferase PFA5 OS=Kluyveromyces lactis (strain ATCC 8585 / CBS 2359 / DSM 70799 / NBRC 1267 / NRRL Y-1140 / WM37) GN=PFA5 PE=3 SV=1

29.  CD5L_MOUSE    Mass: 40345    Score: 33    Expect: 3e+002  Matches: 2

29.  CD5L_MOUSE    Mass: 40345    Score: 33    Expect: 3e+002  Matches: 2
     CD5 antigen-like OS=Mus musculus GN=Cd5l PE=1 SV=3

30.  SSB_MYCPE    Mass: 21466    Score: 32    Expect: 3.2e+002  Matches: 2
     Single-stranded DNA-binding protein OS=Mycoplasma penetrans (strain HF-2) GN=ssb PE=3 SV=1

31.  GCST_CROS8    Mass: 40112    Score: 32    Expect: 3.2e+002  Matches: 2
     Aminomethyltransferase OS=Cronobacter sakazakii (strain ATCC BAA-894) GN=gcvT PE=3 SV=1

32.  GP42_EBVB9    Mass: 25879    Score: 32    Expect: 3.4e+002  Matches: 2
     Glycoprotein 42 OS=Epstein-Barr virus (strain B95-8) GN=BZLF2 PE=1 SV=1
```

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (829.7115,1+): <no title>
                       : seq(C-G)
   Query2 (837.5821,1+): <no title>
                       : seq(C-G)
   Query3 (852.6093,1+): <no title>
                       : seq(C-I)
   Query4 (852.6093,1+): <no title>
                       : seq(C-L)
   Query5 (887.6663,1+): <no title>
                       : seq(C-G)
   Query6 (911.6262,1+): <no title>
                       : seq(C-V)
   Query7 (919.6888,1+): <no title>
                       : seq(C-V)
   Query8 (951.5214,1+): <no title>
                       : seq(C-G)
   Query9 (957.5470,1+): <no title>
                       : seq(C-K)
  Query10 (957.5470,1+): <no title>
```

CYC mass



Mascot Search Results

```
User           : Y Wang
Email          : kahn20003@gmail.com
Search title   :
Database       : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp      : 14 Apr 2016 at 05:21:29 GMT
Top Score      : 43 for HLP_RICCN, Histone-like DNA-binding protein OS=Rickettsia conorii (strain ATCC VR-613 / Malish 7) GN=RC1088 PE=3 SV=1
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Format As: Concise Protein Summary

Significance threshold p< 0.05

Max. number of hits 200

Preferred taxonomy All entries

```
1.   HLP_RICCN    Mass: 10732   Score: 43    Expect: 30  Matches: 9
     Histone-like DNA-binding protein OS=Rickettsia conorii (strain ATCC VR-613 / Malish 7) GN=RC1088 PE=3 SV=1

2.   CYC_HORSE    Mass: 11941   Score: 42    Expect: 31  Matches: 10

2.   CYC_HORSE    Mass: 11941   Score: 42    Expect: 31  Matches: 10
     Cytochrome c OS=Equus caballus GN=CYCS PE=1 SV=2

3.   RR4_CUSRE    Mass: 23499   Score: 40    Expect: 58  Matches: 8
     Plastid 30S ribosomal protein S4 OS=Cuscuta reflexa GN=rps4 PE=3 SV=1

4.   KATG_KORVE    Mass: 83080   Score: 38    Expect: 85  Matches: 12
     Catalase-peroxidase OS=Koribacter versatilis (strain Ellin345) GN=katG PE=3 SV=1

5.   ARP6_YARLI    Mass: 44375   Score: 38    Expect: 94  Matches: 10
     Actin-like protein ARP6 OS=Yarrowia lipolytica (strain CLIB 122 / E 150) GN=ARP6 PE=3 SV=1

6.   RR4_CUSEX    Mass: 23490   Score: 37    Expect: 1e+002  Matches: 7
     Plastid 30S ribosomal protein S4 OS=Cuscuta exaltata GN=rps4 PE=3 SV=1

7.   SSB_BPPH2    Mass: 13335   Score: 37    Expect: 1.1e+002  Matches: 7
     Single-strand-binding protein OS=Bacillus phage phi29 GN=5 PE=1 SV=1
     SSB_BPPZA    Mass: 13322   Score: 37    Expect: 1.1e+002  Matches: 7
     Single-strand-binding protein OS=Bacillus phage PZA GN=5 PE=3 SV=1

8.   HLP_RICRI    Mass: 10713   Score: 36    Expect: 1.3e+002  Matches: 8
     Histone-like DNA-binding protein OS=Rickettsia rickettsii PE=3 SV=1

9.   RL23_STRE4    Mass: 10861   Score: 36    Expect: 1.3e+002  Matches: 8
     50S ribosomal protein L23 OS=Streptococcus equi subsp. equi (strain 4047) GN=rplW PE=3 SV=1

10.  RS9_RHOP2    Mass: 17423   Score: 35    Expect: 1.6e+002  Matches: 8
     30S ribosomal protein S9 OS=Rhodopseudomonas palustris (strain HaA2) GN=rpsI PE=3 SV=1

11.  MSCL_PROMH    Mass: 14919   Score: 35    Expect: 1.7e+002  Matches: 5
     Large-conductance mechanosensitive channel OS=Proteus mirabilis (strain HI4320) GN=mscL PE=3 SV=1

12.  ORF14_SPV1C    Mass: 4266    Score: 35    Expect: 1.8e+002  Matches: 6
     Uncharacterized protein ORF14 OS=Spiroplasma virus SpV1-C74 GN=ORF14 PE=4 SV=1

13.  RL28_PSEFS    Mass: 8903    Score: 34    Expect: 2.2e+002  Matches: 6
     50S ribosomal protein L28 OS=Pseudomonas fluorescens (strain SBW25) GN=rpmB PE=3 SV=1

14.  VAT_CAMVP    Mass: 17833   Score: 34    Expect: 2.4e+002  Matches: 10
     Aphid transmission protein OS=Cauliflower mosaic virus (strain PV147) GN=ORF II PE=3 SV=1
14.  VAT_CAMVP    Mass: 17833   Score: 34    Expect: 2.4e+002  Matches: 10
     Aphid transmission protein OS=Cauliflower mosaic virus (strain PV147) GN=ORF II PE=3 SV=1

15.  DBP7_ASPCL    Mass: 83800   Score: 33    Expect: 2.5e+002  Matches: 12
     ATP-dependent RNA helicase dbp7 OS=Aspergillus clavatus (strain ATCC 1007 / CBS 513.65 / DSM 816 / NCTC 3887 / NRRL 1) GN=dbp7 PE=3 SV=1

16.  RL18_PSEMY    Mass: 12637   Score: 33    Expect: 2.8e+002  Matches: 6
     50S ribosomal protein L18 OS=Pseudomonas mendocina (strain ymp) GN=rplR PE=3 SV=1

17.  CYC_EQUAS    Mass: 11927   Score: 32    Expect: 3.1e+002  Matches: 9
     Cytochrome c OS=Equus asinus GN=CYCS PE=1 SV=2
     CYC_EQUBU    Mass: 11927   Score: 32    Expect: 3.1e+002  Matches: 9
     Cytochrome c OS=Equus burchelli GN=CYCS PE=1 SV=2

18.  DNAK_RENSM    Mass: 66526   Score: 32    Expect: 3.1e+002  Matches: 11
     Chaperone protein DnaK OS=Renibacterium salmoninarum (strain ATCC 33209 / DSM 20767 / JCM 11484 / NBRC 15589 / NCIMB 2235) GN=dnaK PE=3 SV=1

19.  PYRH_PELCD    Mass: 25884   Score: 32    Expect: 3.1e+002  Matches: 7
     Uridylate kinase OS=Pelobacter carbinolicus (strain DSM 2380 / Gra Bd 1) GN=pyrH PE=3 SV=1

20.  ATPA_BUXMI    Mass: 55394   Score: 32    Expect: 3.2e+002  Matches: 9
     ATP synthase subunit alpha, chloroplastic OS=Buxus microphylla GN=atpA PE=3 SV=1

21.  RR4_LACSA    Mass: 23324   Score: 32    Expect: 3.2e+002  Matches: 7
     30S ribosomal protein S4, chloroplastic OS=Lactuca sativa GN=rps4 PE=3 SV=1
     RR4_HELAN    Mass: 23284   Score: 32    Expect: 3.3e+002  Matches: 7
     30S ribosomal protein S4, chloroplastic OS=Helianthus annuus GN=rps4 PE=3 SV=1

22.  ICE2_DROME    Mass: 79889   Score: 32    Expect: 3.4e+002  Matches: 10
     Little elongation complex subunit 2 OS=Drosophila melanogaster GN=Ice2 PE=1 SV=1

23.  RR4_DAUCA    Mass: 23398   Score: 32    Expect: 3.4e+002  Matches: 8
     30S ribosomal protein S4, chloroplastic OS=Daucus carota GN=rps4 PE=3 SV=1

24.  EFTS_GEOLS    Mass: 33519   Score: 32    Expect: 3.6e+002  Matches: 10
     Elongation factor Ts OS=Geobacter lovleyi (strain ATCC BAA-1151 / DSM 17278 / SZ) GN=tsf PE=3 SV=1

25.  MIAA_CALBD    Mass: 36265   Score: 32    Expect: 3.9e+002  Matches: 10
     tRNA dimethylallyltransferase OS=Caldicellulosiruptor bescii (strain ATCC BAA-1888 / DSM 6725 / Z-1320) GN=miaA PE=3 SV=1

26.  UBIG_HAEPS    Mass: 27310   Score: 32    Expect: 3.9e+002  Matches: 7
```

26. UBIG_HAEPS **Mass:** 27310 **Score:** 32 **Expect:** 3.9e+002 **Matches:** 7
Ubiquinone biosynthesis O-methyltransferase OS=Haemophilus parasuis serovar 5 (strain SH0165) GN=ubiG PE=3 SV=1

27. RS17_CHLPN **Mass:** 9883 **Score:** 31 **Expect:** 4e+002 **Matches:** 9
30S ribosomal protein S17 OS=Chlamydia pneumoniae GN=rpsQ PE=3 SV=1

28. RR4_OROMI **Mass:** 23729 **Score:** 31 **Expect:** 4e+002 **Matches:** 8
Plastid 30S ribosomal protein S4 OS=Orobanche minor GN=rps4 PE=3 SV=1

29. TRMB_WOLPM **Mass:** 26351 **Score:** 31 **Expect:** 4.1e+002 **Matches:** 8
tRNA (guanine-N(7)-)-methyltransferase OS=Wolbachia pipientis wMel GN=trmB PE=3 SV=1

30. RL14_STRMU **Mass:** 13013 **Score:** 31 **Expect:** 4.2e+002 **Matches:** 8
50S ribosomal protein L14 OS=Streptococcus mutans serotype c (strain ATCC 700610 / UA159) GN=rplN PE=3 SV=1

31. RL28_HAHCH **Mass:** 8972 **Score:** 31 **Expect:** 4.3e+002 **Matches:** 7
50S ribosomal protein L28 OS=Hahella chejuensis (strain KCTC 2396) GN=rpmB PE=3 SV=1

32. VAPC4_PYRHO **Mass:** 17179 **Score:** 31 **Expect:** 4.6e+002 **Matches:** 7
Ribonuclease VapC4 OS=Pyrococcus horikoshii (strain ATCC 700860 / DSM 12428 / JCM 9974 / NBRC 100139 / OT-3) GN=vapC4 PE=1 SV=1

33. EFP_SODGM **Mass:** 20693 **Score:** 31 **Expect:** 4.8e+002 **Matches:** 6
Elongation factor P OS=Sodalis glossinidius (strain morsitans) GN=efp PE=3 SV=1

34. Y921_STAHJ **Mass:** 19235 **Score:** 31 **Expect:** 4.8e+002 **Matches:** 6
UPF0340 protein SH0921 OS=Staphylococcus haemolyticus (strain JCSC1435) GN=SH0921 PE=3 SV=1

35. Y2358_BACLD **Mass:** 11830 **Score:** 30 **Expect:** 4.9e+002 **Matches:** 6
Nucleoid-associated protein BLi00029/BL02358 OS=Bacillus licheniformis (strain ATCC 14580 / DSM 13 / JCM 2505 / NBRC 12200 / NCIMB 9375 / NRRL NRS-1264 / Gibson 46) GN=BLi00029 PE=3 SV=1

36. FLIJ_SALTI **Mass:** 17369 **Score:** 30 **Expect:** 4.9e+002 **Matches:** 5
Flagellar FliJ protein OS=Salmonella typhi GN=fliJ PE=3 SV=1
FLIJ_SALTY **Mass:** 17369 **Score:** 30 **Expect:** 4.9e+002 **Matches:** 5
Flagellar FliJ protein OS=Salmonella typhimurium (strain LT2 / SGSC1412 / ATCC 700720) GN=fliJ PE=1 SV=1

37. CO6A5_HUMAN **Mass:** 291832 **Score:** 30 **Expect:** 5e+002 **Matches:** 23
Collagen alpha-5(VI) chain OS=Homo sapiens GN=COL6A5 PE=1 SV=1

38. LYS4_NEUCR **Mass:** 84375 **Score:** 30 **Expect:** 5.3e+002 **Matches:** 11
Homoaconitase, mitochondrial OS=Neurospora crassa (strain ATCC 24698 / 74-OR23-1A / CBS 708.71 / DSM 1257 / FGSC 987) GN=lys-4 PE=3 SV=1
Homoaconitase, mitochondrial OS=Neurospora crassa (strain ATCC 24698 / 74-OR23-1A / CBS 708.71 / DSM 1257 / FGSC 987) GN=lys-4 PE=3 SV=1

39. ATPA_RHILO **Mass:** 55038 **Score:** 30 **Expect:** 5.4e+002 **Matches:** 8
ATP synthase subunit alpha OS=Rhizobium loti (strain MAFF303099) GN=atpA PE=3 SV=1

40. ISPE_CLOAB **Mass:** 31458 **Score:** 30 **Expect:** 5.5e+002 **Matches:** 10
4-diphosphocytidyl-2-C-methyl-D-erythritol kinase OS=Clostridium acetobutylicum (strain ATCC 824 / DSM 792 / JCM 1419 / LMG 5710 / VKM B-1787) GN=ispE PE=3 SV=1

41. RS9_RHIL3 **Mass:** 16680 **Score:** 30 **Expect:** 5.6e+002 **Matches:** 6
30S ribosomal protein S9 OS=Rhizobium leguminosarum bv. viciae (strain 3841) GN=rpsI PE=3 SV=1

42. RL34_UREP2 **Mass:** 5701 **Score:** 30 **Expect:** 5.9e+002 **Matches:** 7
50S ribosomal protein L34 OS=Ureaplasma parvum serovar 3 (strain ATCC 27815 / 27 / NCTC 11736) GN=rpmH PE=3 SV=1
RL34_UREPA **Mass:** 5701 **Score:** 30 **Expect:** 5.9e+002 **Matches:** 7
50S ribosomal protein L34 OS=Ureaplasma parvum serovar 3 (strain ATCC 700970) GN=rpmH PE=3 SV=1

43. RL28_SHEHH **Mass:** 9224 **Score:** 30 **Expect:** 5.9e+002 **Matches:** 6
50S ribosomal protein L28 OS=Shewanella halifaxensis (strain HAW-EB4) GN=rpmB PE=3 SV=1
RL28_SHEPA **Mass:** 9240 **Score:** 30 **Expect:** 5.9e+002 **Matches:** 6
50S ribosomal protein L28 OS=Shewanella pealeana (strain ATCC 700345 / ANG-SQ1) GN=rpmB PE=3 SV=1

44. RL28_PSEP1 **Mass:** 8975 **Score:** 30 **Expect:** 6.2e+002 **Matches:** 7
50S ribosomal protein L28 OS=Pseudomonas putida (strain F1 / ATCC 700007) GN=rpmB PE=3 SV=1

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (820.9055,1+): <no title>
   Query2 (828.7350,1+): <no title>
   Query3 (828.8386,1+): <no title>
   Query4 (832.8204,1+): <no title>
   Query5 (856.7492,1+): <no title>
   Query6 (859.9046,1+): <no title>
```

CYC mass/mass+c

Mascot Search Results

```
User            : Y Wang
Email           : kahn20003@gmail.com
Search title    :
Database        : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp       : 14 Apr 2016 at 07:29:14 GMT
Top Score       : 137 for CYC_HORSE, Cytochrome c OS=Equus caballus GN=CYCS PE=1 SV=2
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

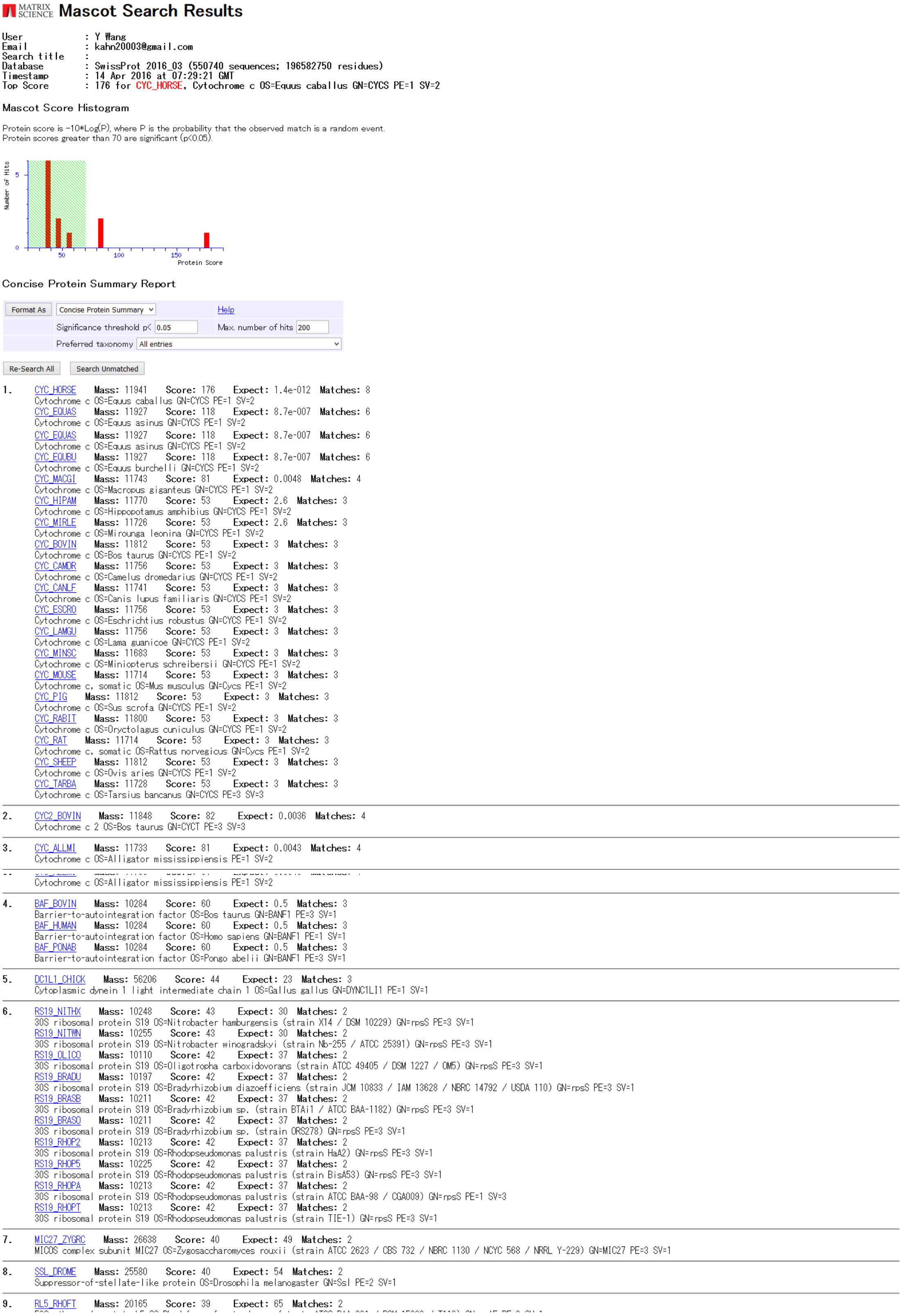
```
1.    CYC_HORSE    Mass: 11941   Score: 137    Expect: 1.1e-008  Matches: 10
      Cytochrome c OS=Equus caballus GN=CYCS PE=1 SV=2
      CYC_MACGI    Mass: 11743   Score: 61     Expect: 0.41  Matches: 5
      Cytochrome c OS=Macropus giganteus GN=CYCS PE=1 SV=2
      CYC_MACGI    Mass: 11743   Score: 61     Expect: 0.41  Matches: 5
      Cytochrome c OS=Macropus giganteus GN=CYCS PE=1 SV=2
      CYC_HIPAM    Mass: 11770   Score: 40     Expect: 50  Matches: 4
      Cytochrome c OS=Hippopotamus amphibius GN=CYCS PE=1 SV=2
      CYC_CAMDR    Mass: 11756   Score: 40     Expect: 55  Matches: 4
      Cytochrome c OS=Camelus dromedarius GN=CYCS PE=1 SV=2
      CYC_ESCRO    Mass: 11756   Score: 40     Expect: 55  Matches: 4
      Cytochrome c OS=Eschrichtius robustus GN=CYCS PE=1 SV=2
      CYC_LAMGU    Mass: 11756   Score: 40     Expect: 55  Matches: 4
      Cytochrome c OS=Lama guanicoe GN=CYCS PE=1 SV=2
      CYC_MOUSE    Mass: 11714   Score: 40     Expect: 55  Matches: 4
      Cytochrome c, somatic OS=Mus musculus GN=Cycs PE=1 SV=2
      CYC_RAT    Mass: 11714   Score: 40     Expect: 55  Matches: 4
      Cytochrome c, somatic OS=Rattus norvegicus GN=Cycs PE=1 SV=2
      CYC_TARBA    Mass: 11728   Score: 40     Expect: 55  Matches: 4
      Cytochrome c OS=Tarsius bancanus GN=CYCS PE=3 SV=3

2.    CYC_EQUAS    Mass: 11927   Score: 100    Expect: 5.8e-005  Matches: 9
      Cytochrome c OS=Equus asinus GN=CYCS PE=1 SV=2
      CYC_EQUBU    Mass: 11927   Score: 100    Expect: 5.8e-005  Matches: 9
      Cytochrome c OS=Equus burchelli GN=CYCS PE=1 SV=2
      CYC_MIRLE    Mass: 11726   Score: 45     Expect: 17  Matches: 5
      Cytochrome c OS=Mirounga leonina GN=CYCS PE=1 SV=2
      CYC_CANLF    Mass: 11741   Score: 45     Expect: 19  Matches: 5
      Cytochrome c OS=Canis lupus familiaris GN=CYCS PE=1 SV=2
      CYC_MINSC    Mass: 11683   Score: 45     Expect: 19  Matches: 5
      Cytochrome c OS=Miniopterus schreibersii GN=CYCS PE=1 SV=2

3.    CYC2_BOVIN    Mass: 11848   Score: 62     Expect: 0.35  Matches: 5
      Cytochrome c 2 OS=Bos taurus GN=CYCT PE=3 SV=3

4.    CYC_ALLMI    Mass: 11733   Score: 62     Expect: 0.38  Matches: 5
      Cytochrome c OS=Alligator mississippiensis PE=1 SV=2

5.    CYC_BOVIN    Mass: 11812   Score: 48     Expect: 8  Matches: 6
      Cytochrome c OS=Bos taurus GN=CYCS PE=1 SV=2
      CYC_PIG    Mass: 11812   Score: 48     Expect: 8  Matches: 6
      Cytochrome c OS=Sus scrofa GN=CYCS PE=1 SV=2
      CYC_SHEEP    Mass: 11812   Score: 48     Expect: 8  Matches: 6
      Cytochrome c OS=Ovis aries GN=CYCS PE=1 SV=2

      Cytochrome c OS=Ovis aries GN=CYCS PE=1 SV=2

6.    RL5_BORA1    Mass: 20185   Score: 46     Expect: 16  Matches: 7
      50S ribosomal protein L5 OS=Bordetella avium (strain 197N) GN=rplE PE=3 SV=1

7.    BAF_BOVIN    Mass: 10284   Score: 44     Expect: 20  Matches: 3
      Barrier-to-autointegration factor OS=Bos taurus GN=BANF1 PE=3 SV=1
      BAF_HUMAN    Mass: 10284   Score: 44     Expect: 20  Matches: 3
      Barrier-to-autointegration factor OS=Homo sapiens GN=BANF1 PE=1 SV=1
      BAF_PONAB    Mass: 10284   Score: 44     Expect: 20  Matches: 3
      Barrier-to-autointegration factor OS=Pongo abelii GN=BANF1 PE=3 SV=1

8.    CYC_RABIT    Mass: 11800   Score: 43     Expect: 29  Matches: 5
      Cytochrome c OS=Oryctolagus cuniculus GN=CYCS PE=1 SV=2

9.    Y989_METJA    Mass: 12743   Score: 41     Expect: 45  Matches: 4
      Uncharacterized protein MJ0989 OS=Methanocaldococcus jannaschii (strain ATCC 43067 / DSM 2661 / JAL-1 / JCM 10045 / NBRC 100440) GN=MJ0989 PE=4 SV=1

10.   RL5_BORBR    Mass: 20261   Score: 40     Expect: 51  Matches: 6
      50S ribosomal protein L5 OS=Bordetella bronchiseptica (strain ATCC BAA-588 / NCTC 13252 / RB50) GN=rplE PE=3 SV=1
      RL5_BORPA    Mass: 20261   Score: 40     Expect: 51  Matches: 6
      50S ribosomal protein L5 OS=Bordetella parapertussis (strain 12822 / ATCC BAA-587 / NCTC 13253) GN=rplE PE=3 SV=1
      RL5_BORPE    Mass: 20261   Score: 40     Expect: 51  Matches: 6
      50S ribosomal protein L5 OS=Bordetella pertussis (strain Tohama I / ATCC BAA-589 / NCTC 13251) GN=rplE PE=3 SV=1

11.   RL5_RHOFT    Mass: 20165   Score: 39     Expect: 69  Matches: 4
      50S ribosomal protein L5 OS=Rhodoferax ferrireducens (strain ATCC BAA-621 / DSM 15236 / T118) GN=rplE PE=3 SV=1

12.   ATPA_BUXMI    Mass: 55394   Score: 39     Expect: 74  Matches: 8
      ATP synthase subunit alpha, chloroplastic OS=Buxus microphylla GN=atpA PE=3 SV=1

13.   HIS5_LISW6    Mass: 23241   Score: 38     Expect: 78  Matches: 5
      Imidazole glycerol phosphate synthase subunit HisH OS=Listeria welshimeri serovar 6b (strain ATCC 35897 / DSM 20650 / SLCC5334) GN=hisH PE=3 SV=1

14.   MURA_THEYD    Mass: 45440   Score: 38     Expect: 83  Matches: 8
      UDP-N-acetylglucosamine 1-carboxyvinyltransferase OS=Thermodesulfovibrio yellowstonii (strain ATCC 51303 / DSM 11347 / YP87) GN=murA PE=3 SV=1

15.   RL5_POLSJ    Mass: 20219   Score: 38     Expect: 96  Matches: 4
      50S ribosomal protein L5 OS=Polaromonas sp. (strain JS666 / ATCC BAA-500) GN=rplE PE=3 SV=1

16.   RIMO_SOLUE    Mass: 51927   Score: 37     Expect: 1.1e+002  Matches: 6
```

```
16.   RIMO_SOLUE    Mass: 51927    Score: 37    Expect: 1.1e+002  Matches: 6
      Ribosomal protein S12 methylthiotransferase RimO OS=Solibacter usitatus (strain Ellin6076) GN=rimO PE=3 SV=1

17.   DNLJ_HELPS    Mass: 74285    Score: 37    Expect: 1.2e+002  Matches: 11
      DNA ligase OS=Helicobacter pylori (strain Shi470) GN=ligA PE=3 SV=1

18.   HRCA_MYCA5    Mass: 38351    Score: 36    Expect: 1.3e+002  Matches: 7
      Heat-inducible transcription repressor HrcA OS=Mycoplasma arthritidis (strain 158L3-1) GN=hrcA PE=3 SV=1

19.   RR4_ASPMA    Mass: 22677    Score: 36    Expect: 1.4e+002  Matches: 6
      30S ribosomal protein S4, chloroplastic (Fragment) OS=Asparagus maritimus GN=rps4 PE=3 SV=1

20.   DC1L1_CHICK    Mass: 56206    Score: 36    Expect: 1.4e+002  Matches: 6
      Cytoplasmic dynein 1 light intermediate chain 1 OS=Gallus gallus GN=DYNC1LI1 PE=1 SV=1

21.   PYRE_CELJU    Mass: 23289    Score: 36    Expect: 1.4e+002  Matches: 5
      Orotate phosphoribosyltransferase OS=Cellvibrio japonicus (strain Ueda107) GN=pyrE PE=3 SV=1

22.   AMPP3_BOTFB    Mass: 51823    Score: 36    Expect: 1.4e+002  Matches: 6
      Probable Xaa-Pro aminopeptidase pepP OS=Botryotinia fuckeliana (strain B05.10) GN=pepP PE=3 SV=1

23.   PFS2_YEAST    Mass: 53515    Score: 36    Expect: 1.4e+002  Matches: 9
      Polyadenylation factor subunit 2 OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=PFS2 PE=1 SV=1

24.   ACCO3_PETHY    Mass: 36540    Score: 36    Expect: 1.4e+002  Matches: 6
      1-aminocyclopropane-1-carboxylate oxidase 3 OS=Petunia hybrida GN=ACO3 PE=3 SV=1

25.   ARL6_MOUSE    Mass: 21120    Score: 36    Expect: 1.4e+002  Matches: 5
      ADP-ribosylation factor-like protein 6 OS=Mus musculus GN=Arl6 PE=1 SV=1

26.   RR4_CUSRE    Mass: 23499    Score: 36    Expect: 1.5e+002  Matches: 6
      Plastid 30S ribosomal protein S4 OS=Cuscuta reflexa GN=rps4 PE=3 SV=1

27.   WECG_PHOLL    Mass: 27935    Score: 35    Expect: 1.7e+002  Matches: 5
      UDP-N-acetyl-D-mannosaminuronic acid transferase OS=Photorhabdus luminescens subsp. laumondii (strain DSM 15139 / CIP 105565 / TT01) GN=wecG PE=3 SV=1

28.   SLBP_MOUSE    Mass: 31873    Score: 35    Expect: 1.9e+002  Matches: 5
      Histone RNA hairpin-binding protein OS=Mus musculus GN=Slbp PE=1 SV=1

29.   RR4 CALFG    Mass: 23504    Score: 35    Expect: 1.9e+002  Matches: 5

29.   RR4_CALFG    Mass: 23504    Score: 35    Expect: 1.9e+002  Matches: 5
      30S ribosomal protein S4, chloroplastic OS=Calycanthus floridus var. glaucus GN=rps4 PE=3 SV=1

30.   ATPA_ACUOB    Mass: 54769    Score: 34    Expect: 2e+002  Matches: 8
      ATP synthase subunit alpha, chloroplastic OS=Acutodesmus obliquus GN=atpA PE=3 SV=1

31.   EZRA_BACLD    Mass: 65949    Score: 34    Expect: 2e+002  Matches: 4
      Septation ring formation regulator EzrA OS=Bacillus licheniformis (strain ATCC 14580 / DSM 13 / JCM 2505 / NBRC 12200 / NCIMB 9375 / NRRL NRS-1264 / Gibson 46) GN=ezrA PE=3 SV=1

32.   SSB_BPPH2    Mass: 13335    Score: 34    Expect: 2.1e+002  Matches: 6
      Single-strand-binding protein OS=Bacillus phage phi29 GN=5 PE=1 SV=1
```

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (820.9055,1+): <no title>
   Query2 (828.7350,1+): <no title>
   Query3 (828.8386,1+): <no title>
                       : seq(C-T)
   Query4 (832.8204,1+): <no title>
   Query5 (856.7492,1+): <no title>
   Query6 (859.9046,1+): <no title>
   Query7 (861.4703,1+): <no title>
   Query8 (861.6490,1+): <no title>
   Query9 (864.8382,1+): <no title>
  Query10 (872.6738,1+): <no title>
  Query11 (877.3892,1+): <no title>
  Query12 (877.4345,1+): <no title>
  Query13 (879.7843,1+): <no title>
  Query14 (907.8780,1+): <no title>
  Query15 (907.8818,1+): <no title>
                       : seq(C-K)
  Query16 (907.8818,1+): <no title>
                       : seq(C-Q)
```

CYC mass+c



Mascot Search Results

```
User            : Y Wang
Email           : kahn20003@gmail.com
Search title    :
Database        : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp       : 14 Apr 2016 at 07:29:21 GMT
Top Score       : 176 for CYC_HORSE, Cytochrome c OS=Equus caballus GN=CYCS PE=1 SV=2
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

```
1.   CYC_HORSE    Mass: 11941    Score: 176    Expect: 1.4e-012  Matches: 8
     Cytochrome c OS=Equus caballus GN=CYCS PE=1 SV=2
     CYC_EQUAS    Mass: 11927    Score: 118    Expect: 8.7e-007  Matches: 6
     Cytochrome c OS=Equus asinus GN=CYCS PE=1 SV=2
     CYC_EQUAS    Mass: 11927    Score: 118    Expect: 8.7e-007  Matches: 6
     Cytochrome c OS=Equus asinus GN=CYCS PE=1 SV=2
     CYC_EQUBU    Mass: 11927    Score: 118    Expect: 8.7e-007  Matches: 6
     Cytochrome c OS=Equus burchelli GN=CYCS PE=1 SV=2
     CYC_MACGI    Mass: 11743    Score: 81    Expect: 0.0048  Matches: 4
     Cytochrome c OS=Macropus giganteus GN=CYCS PE=1 SV=2
     CYC_HIPAM    Mass: 11770    Score: 53    Expect: 2.6  Matches: 3
     Cytochrome c OS=Hippopotamus amphibius GN=CYCS PE=1 SV=2
     CYC_MIRLE    Mass: 11726    Score: 53    Expect: 2.6  Matches: 3
     Cytochrome c OS=Mirounga leonina GN=CYCS PE=1 SV=2
     CYC_BOVIN    Mass: 11812    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Bos taurus GN=CYCS PE=1 SV=2
     CYC_CAMDR    Mass: 11756    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Camelus dromedarius GN=CYCS PE=1 SV=2
     CYC_CANLF    Mass: 11741    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Canis lupus familiaris GN=CYCS PE=1 SV=2
     CYC_ESCRO    Mass: 11756    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Eschrichtius robustus GN=CYCS PE=1 SV=2
     CYC_LAMGU    Mass: 11756    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Lama guanicoe GN=CYCS PE=1 SV=2
     CYC_MINSC    Mass: 11683    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Miniopterus schreibersii GN=CYCS PE=1 SV=2
     CYC_MOUSE    Mass: 11714    Score: 53    Expect: 3  Matches: 3
     Cytochrome c, somatic OS=Mus musculus GN=Cycs PE=1 SV=2
     CYC_PIG    Mass: 11812    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Sus scrofa GN=CYCS PE=1 SV=2
     CYC_RABIT    Mass: 11800    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Oryctolagus cuniculus GN=CYCS PE=1 SV=2
     CYC_RAT    Mass: 11714    Score: 53    Expect: 3  Matches: 3
     Cytochrome c, somatic OS=Rattus norvegicus GN=Cycs PE=1 SV=2
     CYC_SHEEP    Mass: 11812    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Ovis aries GN=CYCS PE=1 SV=2
     CYC_TARBA    Mass: 11728    Score: 53    Expect: 3  Matches: 3
     Cytochrome c OS=Tarsius bancanus GN=CYCS PE=3 SV=3

2.   CYC2_BOVIN    Mass: 11848    Score: 82    Expect: 0.0036  Matches: 4
     Cytochrome c 2 OS=Bos taurus GN=CYCT PE=3 SV=3

3.   CYC_ALLMI    Mass: 11733    Score: 81    Expect: 0.0043  Matches: 4
     Cytochrome c OS=Alligator mississippiensis PE=1 SV=2
```

Cytochrome c OS=Alligator mississippiensis PE=1 SV=2

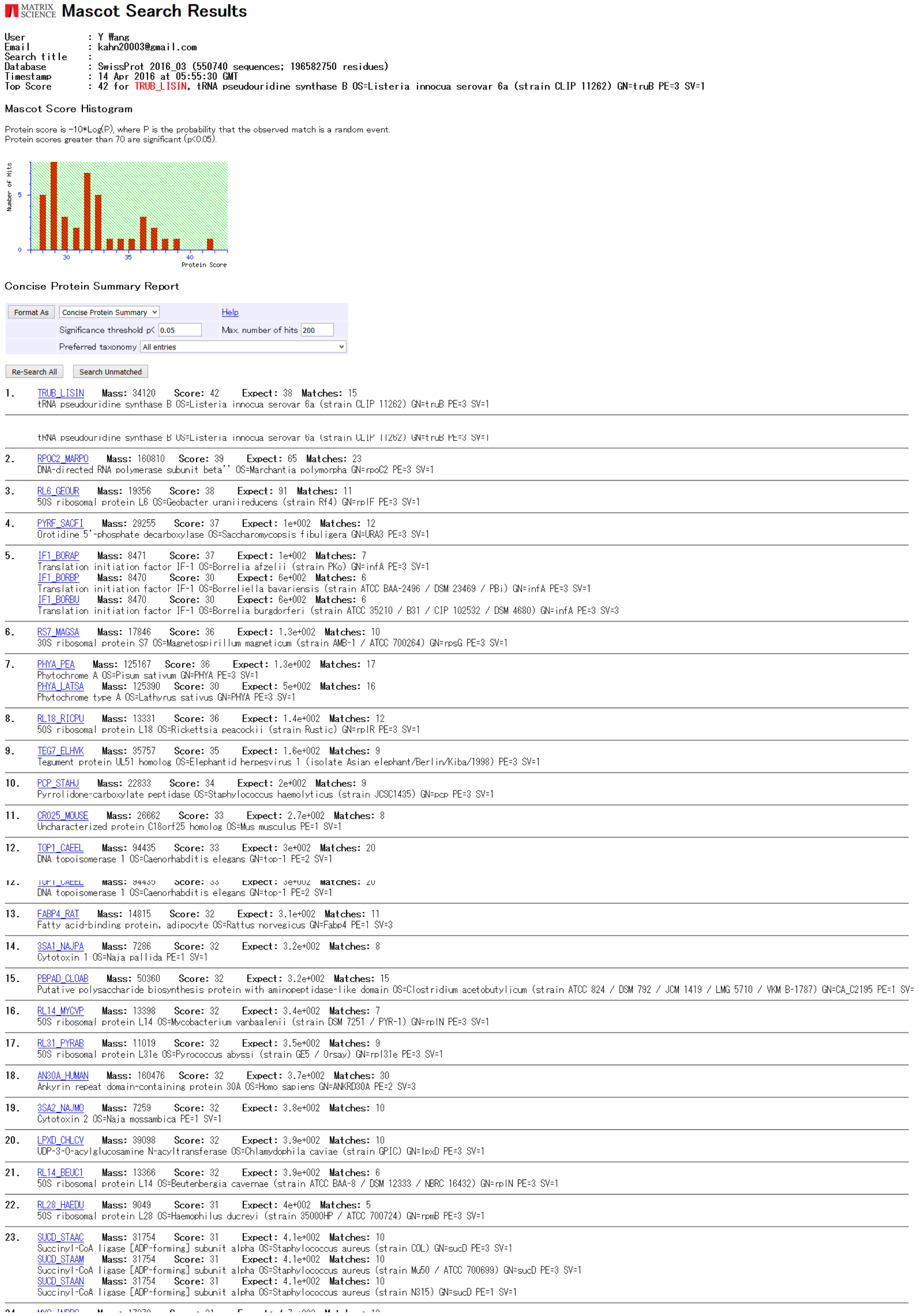
```
4.   BAF_BOVIN    Mass: 10284    Score: 60    Expect: 0.5  Matches: 3
     Barrier-to-autointegration factor OS=Bos taurus GN=BANF1 PE=3 SV=1
     BAF_HUMAN    Mass: 10284    Score: 60    Expect: 0.5  Matches: 3
     Barrier-to-autointegration factor OS=Homo sapiens GN=BANF1 PE=1 SV=1
     BAF_PONAB    Mass: 10284    Score: 60    Expect: 0.5  Matches: 3
     Barrier-to-autointegration factor OS=Pongo abelii GN=BANF1 PE=3 SV=1

5.   DC1L1_CHICK    Mass: 56206    Score: 44    Expect: 23  Matches: 3
     Cytoplasmic dynein 1 light intermediate chain 1 OS=Gallus gallus GN=DYNC1LI1 PE=1 SV=1

6.   RS19_NITHX    Mass: 10248    Score: 43    Expect: 30  Matches: 2
     30S ribosomal protein S19 OS=Nitrobacter hamburgensis (strain X14 / DSM 10229) GN=rpsS PE=3 SV=1
     RS19_NITWN    Mass: 10255    Score: 43    Expect: 30  Matches: 2
     30S ribosomal protein S19 OS=Nitrobacter winogradskyi (strain Nb-255 / ATCC 25391) GN=rpsS PE=3 SV=1
     RS19_OLICO    Mass: 10110    Score: 42    Expect: 37  Matches: 2
     30S ribosomal protein S19 OS=Oligotropha carboxidovorans (strain ATCC 49405 / DSM 1227 / OM5) GN=rpsS PE=3 SV=1
     RS19_BRADU    Mass: 10197    Score: 42    Expect: 37  Matches: 2
     30S ribosomal protein S19 OS=Bradyrhizobium diazoefficiens (strain JCM 10833 / IAM 13628 / NBRC 14792 / USDA 110) GN=rpsS PE=3 SV=1
     RS19_BRASB    Mass: 10211    Score: 42    Expect: 37  Matches: 2
     30S ribosomal protein S19 OS=Bradyrhizobium sp. (strain BTAi1 / ATCC BAA-1182) GN=rpsS PE=3 SV=1
     RS19_BRASO    Mass: 10211    Score: 42    Expect: 37  Matches: 2
     30S ribosomal protein S19 OS=Bradyrhizobium sp. (strain ORS278) GN=rpsS PE=3 SV=1
     RS19_RHOP2    Mass: 10213    Score: 42    Expect: 37  Matches: 2
     30S ribosomal protein S19 OS=Rhodopseudomonas palustris (strain HaA2) GN=rpsS PE=3 SV=1
     RS19_RHOP5    Mass: 10225    Score: 42    Expect: 37  Matches: 2
     30S ribosomal protein S19 OS=Rhodopseudomonas palustris (strain BisA53) GN=rpsS PE=3 SV=1
     RS19_RHOPA    Mass: 10213    Score: 42    Expect: 37  Matches: 2
     30S ribosomal protein S19 OS=Rhodopseudomonas palustris (strain ATCC BAA-98 / CGA009) GN=rpsS PE=1 SV=3
     RS19_RHOPT    Mass: 10213    Score: 42    Expect: 37  Matches: 2
     30S ribosomal protein S19 OS=Rhodopseudomonas palustris (strain TIE-1) GN=rpsS PE=3 SV=1

7.   MIC27_ZYGRC    Mass: 26638    Score: 40    Expect: 49  Matches: 2
     MICOS complex subunit MIC27 OS=Zygosaccharomyces rouxii (strain ATCC 2623 / CBS 732 / NBRC 1130 / NCYC 568 / NRRL Y-229) GN=MIC27 PE=3 SV=1

8.   SSL_DROME    Mass: 25580    Score: 40    Expect: 54  Matches: 2
     Suppressor-of-stellate-like protein OS=Drosophila melanogaster GN=Ssl PE=2 SV=1

9.   RL5_RHOFT    Mass: 20165    Score: 39    Expect: 65  Matches: 2
```

```
9.    RL5_RHOFT    Mass: 20165    Score: 39     Expect: 65  Matches: 2
      50S ribosomal protein L5 OS=Rhodoferax ferrireducens (strain ATCC BAA-621 / DSM 15236 / T118) GN=rplE PE=3 SV=1
      RL5_ACIET    Mass: 20038    Score: 38     Expect: 81  Matches: 2
      50S ribosomal protein L5 OS=Acidovorax ebreus (strain TPSY) GN=rplE PE=3 SV=1
      RL5_ACISJ    Mass: 20038    Score: 38     Expect: 81  Matches: 2
      50S ribosomal protein L5 OS=Acidovorax sp. (strain JS42) GN=rplE PE=3 SV=1
      RL5_POLNA    Mass: 20171    Score: 38     Expect: 81  Matches: 2
      50S ribosomal protein L5 OS=Polaromonas naphthalenivorans (strain CJ2) GN=rplE PE=3 SV=1
      RL5_POLSJ    Mass: 20219    Score: 38     Expect: 81  Matches: 2
      50S ribosomal protein L5 OS=Polaromonas sp. (strain JS666 / ATCC BAA-500) GN=rplE PE=3 SV=1
      RL5_BORA1    Mass: 20185    Score: 37     Expect: 1e+002  Matches: 2
      50S ribosomal protein L5 OS=Bordetella avium (strain 197N) GN=rplE PE=3 SV=1
      RL5_BORBR    Mass: 20261    Score: 37     Expect: 1e+002  Matches: 2
      50S ribosomal protein L5 OS=Bordetella bronchiseptica (strain ATCC BAA-588 / NCTC 13252 / RB50) GN=rplE PE=3 SV=1
      RL5_BORPA    Mass: 20261    Score: 37     Expect: 1e+002  Matches: 2
      50S ribosomal protein L5 OS=Bordetella parapertussis (strain 12822 / ATCC BAA-587 / NCTC 13253) GN=rplE PE=3 SV=1

10.   Y989_METJA    Mass: 12743    Score: 39     Expect: 66  Matches: 2
      Uncharacterized protein MJ0989 OS=Methanocaldococcus jannaschii (strain ATCC 43067 / DSM 2661 / JAL-1 / JCM 10045 / NBRC 100440) GN=MJ0989 PE=4 SV=1

11.   HIS5_LISW6    Mass: 23241    Score: 39     Expect: 74  Matches: 2
      Imidazole glycerol phosphate synthase subunit HisH OS=Listeria welshimeri serovar 6b (strain ATCC 35897 / DSM 20650 / SLCC5334) GN=hisH PE=3 SV=1

12.   RL14_ARTAT    Mass: 13280    Score: 38     Expect: 94  Matches: 2
      50S ribosomal protein L14 OS=Arthrobacter aurescens (strain TC1) GN=rplN PE=3 SV=1
      RL14_ARTCA    Mass: 13280    Score: 38     Expect: 94  Matches: 2
      50S ribosomal protein L14 OS=Arthrobacter chlorophenolicus (strain ATCC 700700 / DSM 12829 / JCM 12360 / NCIMB 13794 / A6) GN=rplN PE=3 SV=1
      RL14_ARTS2    Mass: 13280    Score: 38     Expect: 94  Matches: 2
      50S ribosomal protein L14 OS=Arthrobacter sp. (strain FB24) GN=rplN PE=3 SV=1
      RL14_RENSM    Mass: 13264    Score: 38     Expect: 94  Matches: 2
      50S ribosomal protein L14 OS=Renibacterium salmoninarum (strain ATCC 33209 / DSM 20767 / JCM 11484 / NBRC 15589 / NCIMB 2235) GN=rplN PE=3 SV=1
```

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da

Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (828.8386,1+): <no title>
                       : seq(C-T)
   Query2 (907.8818,1+): <no title>
                       : seq(C-K)
   Query3 (907.8818,1+): <no title>
                       : seq(C-Q)
   Query4 (992.8366,1+): <no title>
                       : seq(C-I)
   Query5 (992.8366,1+): <no title>
                       : seq(C-L)
  Query6 (1016.7344,1+): <no title>
                       : seq(C-F)
  Query7 (1038.6964,1+): <no title>
                       : seq(C-F)
  Query8 (1134.6925,1+): <no title>
                       : seq(C-E)
  Query9 (1274.6438,1+): <no title>
                       : seq(C-F)
 Query10 (1286.6981,1+): <no title>
                       : seq(C-F)
 Query11 (1296.8462,1+): <no title>
                       : seq(C-R)
 Query12 (1350.8030,1+): <no title>
                       : seq(C-I)
 Query13 (1350.8030,1+): <no title>
                       : seq(C-L)
 Query14 (1354.7767,1+): <no title>
                       : seq(C-N)
 Query15 (1408.7645,1+): <no title>
                       : seq(C-I)
 Query16 (1408.7645,1+): <no title>
                       : seq(C-L)
 Query17 (1424.8444,1+): <no title>
                       : seq(C-K)
 Query18 (1424.8444,1+): <no title>
                       : seq(C-Q)
 Query19 (1447.7640,1+): <no title>
                       : seq(C-D)
 Query20 (1466.7246,1+): <no title>
                       : seq(C-I)
```

MB mass



Mascot Search Results

```
User          : Y Wang
Email         : kahn20003@gmail.com
Search title  :
Database      : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp     : 14 Apr 2016 at 05:55:30 GMT
Top Score     : 42 for TRUB_LISIN, tRNA pseudouridine synthase B OS=Listeria innocua serovar 6a (strain CLIP 11262) GN=truB PE=3 SV=1
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

1. TRUB_LISIN **Mass:** 34120 **Score:** 42 **Expect:** 38 **Matches:** 15
tRNA pseudouridine synthase B OS=Listeria innocua serovar 6a (strain CLIP 11262) GN=truB PE=3 SV=1

tRNA pseudouridine synthase B OS=Listeria innocua serovar 6a (strain CLIP 11262) GN=truB PE=3 SV=1

2. RPOC2_MARPO **Mass:** 160810 **Score:** 39 **Expect:** 65 **Matches:** 23
DNA-directed RNA polymerase subunit beta'' OS=Marchantia polymorpha GN=rpoC2 PE=3 SV=1

3. RL6_GEOUR **Mass:** 19356 **Score:** 38 **Expect:** 91 **Matches:** 11
50S ribosomal protein L6 OS=Geobacter uraniireducens (strain Rf4) GN=rplF PE=3 SV=1

4. PYRF_SACFI **Mass:** 29255 **Score:** 37 **Expect:** 1e+002 **Matches:** 12
Orotidine 5'-phosphate decarboxylase OS=Saccharomycopsis fibuligera GN=URA3 PE=3 SV=1

5. IF1_BORAP **Mass:** 8471 **Score:** 37 **Expect:** 1e+002 **Matches:** 7
Translation initiation factor IF-1 OS=Borrelia afzelii (strain PKo) GN=infA PE=3 SV=1
IF1_BORBP **Mass:** 8470 **Score:** 30 **Expect:** 6e+002 **Matches:** 6
Translation initiation factor IF-1 OS=Borreliella bavariensis (strain ATCC BAA-2496 / DSM 23469 / PBi) GN=infA PE=3 SV=1
IF1_BORBU **Mass:** 8470 **Score:** 30 **Expect:** 6e+002 **Matches:** 6
Translation initiation factor IF-1 OS=Borrelia burgdorferi (strain ATCC 35210 / B31 / CIP 102532 / DSM 4680) GN=infA PE=3 SV=3

6. RS7_MAGSA **Mass:** 17846 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 10
30S ribosomal protein S7 OS=Magnetospirillum magneticum (strain AMB-1 / ATCC 700264) GN=rpsG PE=3 SV=1

7. PHYA_PEA **Mass:** 125167 **Score:** 36 **Expect:** 1.3e+002 **Matches:** 17
Phytochrome A OS=Pisum sativum GN=PHYA PE=3 SV=1
PHYA_LATSA **Mass:** 125390 **Score:** 30 **Expect:** 5e+002 **Matches:** 16
Phytochrome type A OS=Lathyrus sativus GN=PHYA PE=3 SV=1

8. RL18_RICPU **Mass:** 13331 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 12
50S ribosomal protein L18 OS=Rickettsia peacockii (strain Rustic) GN=rplR PE=3 SV=1

9. TEG7_ELHVK **Mass:** 35757 **Score:** 35 **Expect:** 1.6e+002 **Matches:** 9
Tegument protein UL51 homolog OS=Elephantid herpesvirus 1 (isolate Asian elephant/Berlin/Kiba/1998) PE=3 SV=1

10. PCP_STAHJ **Mass:** 22833 **Score:** 34 **Expect:** 2e+002 **Matches:** 9
Pyrrolidone-carboxylate peptidase OS=Staphylococcus haemolyticus (strain JCSC1435) GN=pcp PE=3 SV=1

11. CR025_MOUSE **Mass:** 26662 **Score:** 33 **Expect:** 2.7e+002 **Matches:** 8
Uncharacterized protein C18orf25 homolog OS=Mus musculus PE=1 SV=1

12. TOP1_CAEEL **Mass:** 94435 **Score:** 33 **Expect:** 3e+002 **Matches:** 20
DNA topoisomerase 1 OS=Caenorhabditis elegans GN=top-1 PE=2 SV=1

12. TOP1_CAEEL **Mass:** 94435 **Score:** 33 **Expect:** 3e+002 **Matches:** 20
DNA topoisomerase 1 OS=Caenorhabditis elegans GN=top-1 PE=2 SV=1

13. FABP4_RAT **Mass:** 14815 **Score:** 32 **Expect:** 3.1e+002 **Matches:** 11
Fatty acid-binding protein, adipocyte OS=Rattus norvegicus GN=Fabp4 PE=1 SV=3

14. 3SA1_NAJPA **Mass:** 7286 **Score:** 32 **Expect:** 3.2e+002 **Matches:** 8
Cytotoxin 1 OS=Naja pallida PE=1 SV=1

15. PBPAD_CLOAB **Mass:** 50360 **Score:** 32 **Expect:** 3.2e+002 **Matches:** 15
Putative polysaccharide biosynthesis protein with aminopeptidase-like domain OS=Clostridium acetobutylicum (strain ATCC 824 / DSM 792 / JCM 1419 / LMG 5710 / VKM B-1787) GN=CA_C2195 PE=1 SV=

16. RL14_MYCVP **Mass:** 13398 **Score:** 32 **Expect:** 3.4e+002 **Matches:** 7
50S ribosomal protein L14 OS=Mycobacterium vanbaalenii (strain DSM 7251 / PYR-1) GN=rplN PE=3 SV=1

17. RL31_PYRAB **Mass:** 11019 **Score:** 32 **Expect:** 3.5e+002 **Matches:** 9
50S ribosomal protein L31e OS=Pyrococcus abyssi (strain GE5 / Orsay) GN=rpl31e PE=3 SV=1

18. AN30A_HUMAN **Mass:** 160476 **Score:** 32 **Expect:** 3.7e+002 **Matches:** 30
Ankyrin repeat domain-containing protein 30A OS=Homo sapiens GN=ANKRD30A PE=2 SV=3

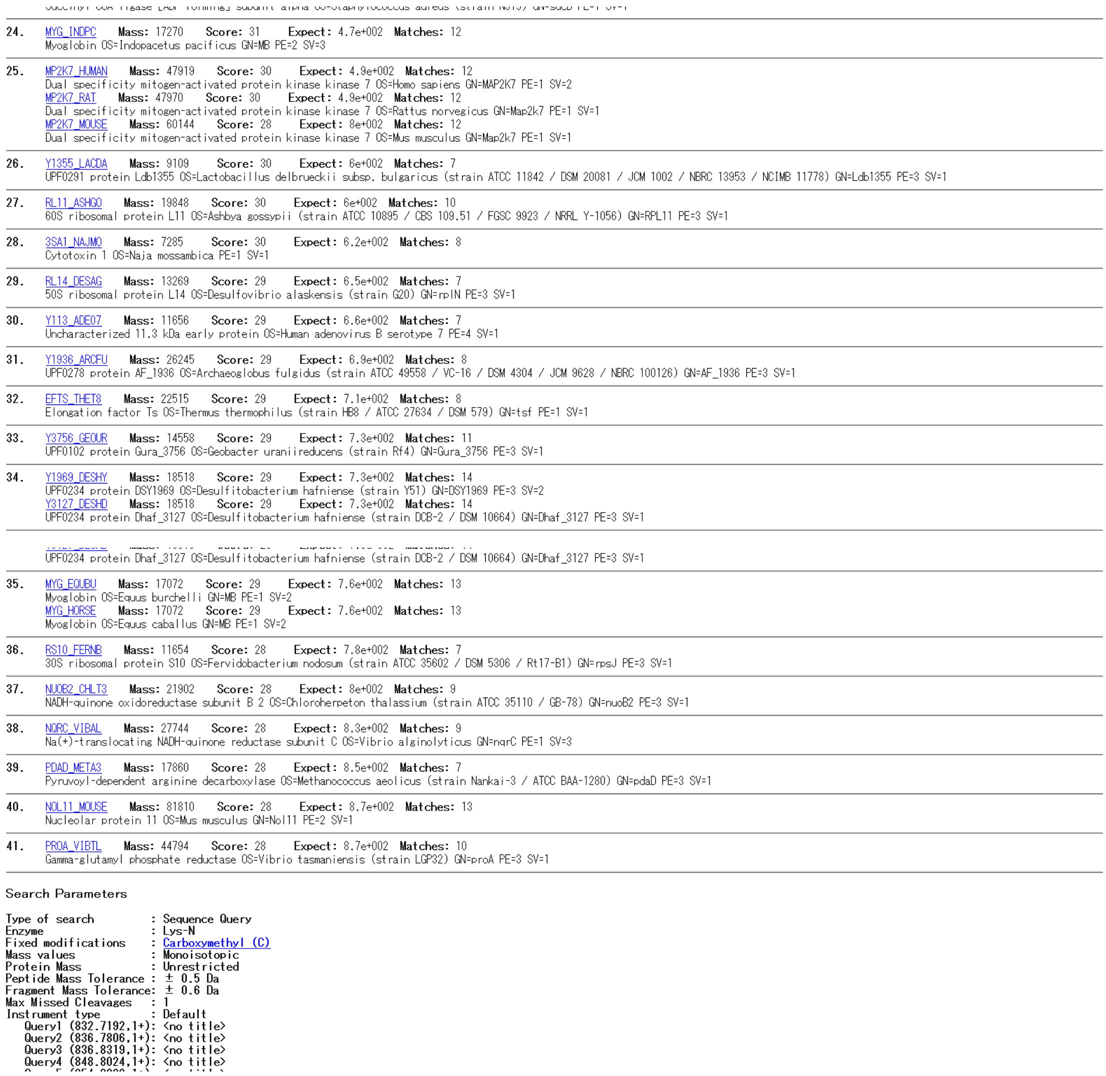
19. 3SA2_NAJMO **Mass:** 7259 **Score:** 32 **Expect:** 3.8e+002 **Matches:** 10
Cytotoxin 2 OS=Naja mossambica PE=1 SV=1

20. LPXD_CHLCV **Mass:** 39098 **Score:** 32 **Expect:** 3.9e+002 **Matches:** 10
UDP-3-O-acylglucosamine N-acyltransferase OS=Chlamydophila caviae (strain GPIC) GN=lpxD PE=3 SV=1

21. RL14_BEUC1 **Mass:** 13366 **Score:** 32 **Expect:** 3.9e+002 **Matches:** 6
50S ribosomal protein L14 OS=Beutenbergia cavernae (strain ATCC BAA-8 / DSM 12333 / NBRC 16432) GN=rplN PE=3 SV=1

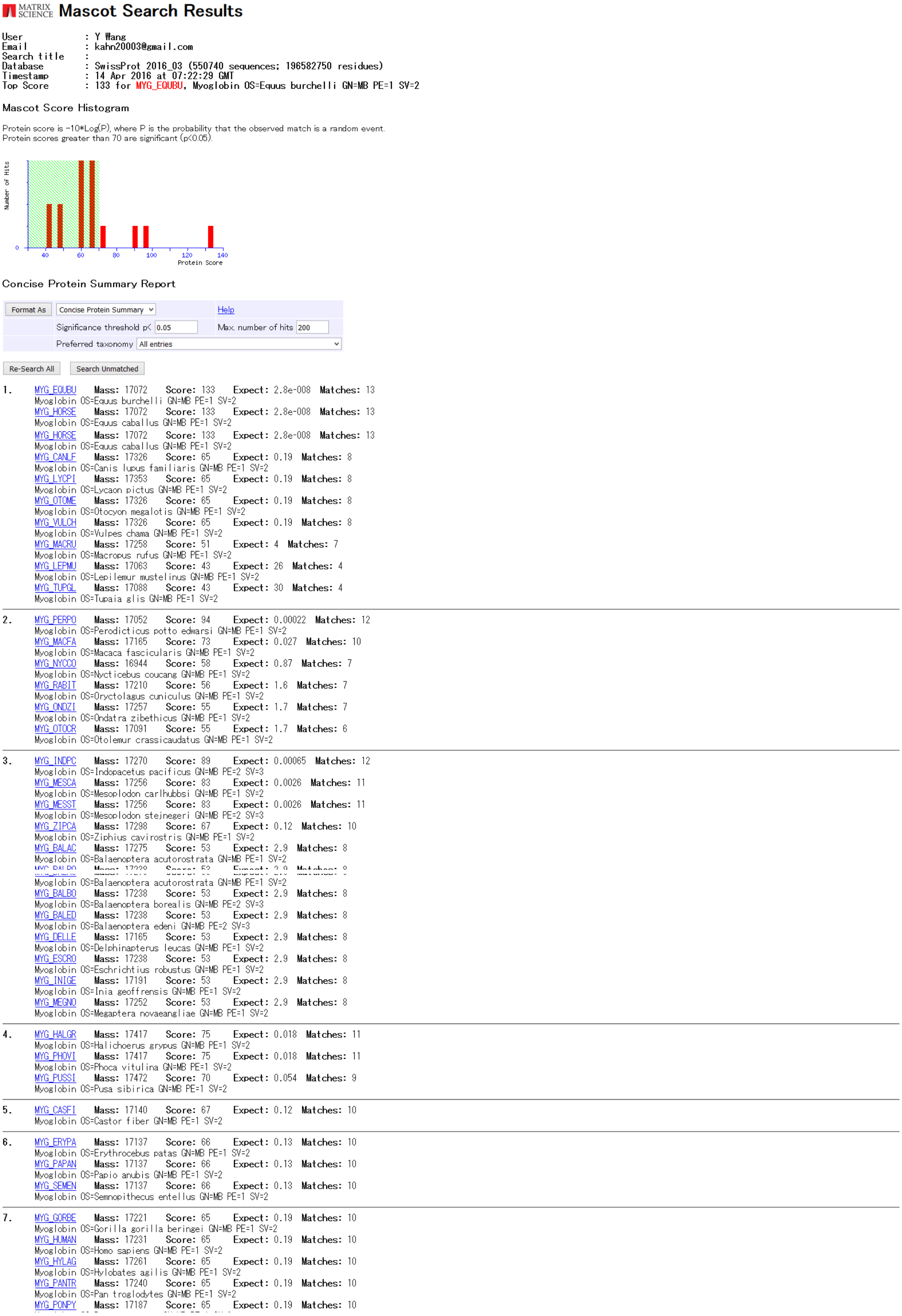
22. RL28_HAEDU **Mass:** 9049 **Score:** 31 **Expect:** 4e+002 **Matches:** 5
50S ribosomal protein L28 OS=Haemophilus ducreyi (strain 35000HP / ATCC 700724) GN=rpmB PE=3 SV=1

23. SUCD_STAAC **Mass:** 31754 **Score:** 31 **Expect:** 4.1e+002 **Matches:** 10
Succinyl-CoA ligase [ADP-forming] subunit alpha OS=Staphylococcus aureus (strain COL) GN=sucD PE=3 SV=1
SUCD_STAAM **Mass:** 31754 **Score:** 31 **Expect:** 4.1e+002 **Matches:** 10
Succinyl-CoA ligase [ADP-forming] subunit alpha OS=Staphylococcus aureus (strain Mu50 / ATCC 700699) GN=sucD PE=3 SV=1
SUCD_STAAN **Mass:** 31754 **Score:** 31 **Expect:** 4.1e+002 **Matches:** 10
Succinyl-CoA ligase [ADP-forming] subunit alpha OS=Staphylococcus aureus (strain N315) GN=sucD PE=1 SV=1



```
24.  MYG_INDPC    Mass: 17270    Score: 31     Expect: 4.7e+002  Matches: 12
     Myoglobin OS=Indopacetus pacificus GN=MB PE=2 SV=3

25.  MP2K7_HUMAN    Mass: 47919    Score: 30     Expect: 4.9e+002  Matches: 12
     Dual specificity mitogen-activated protein kinase kinase 7 OS=Homo sapiens GN=MAP2K7 PE=1 SV=2
     MP2K7_RAT    Mass: 47970    Score: 30     Expect: 4.9e+002  Matches: 12
     Dual specificity mitogen-activated protein kinase kinase 7 OS=Rattus norvegicus GN=Map2k7 PE=1 SV=1
     MP2K7_MOUSE    Mass: 60144    Score: 28     Expect: 8e+002  Matches: 12
     Dual specificity mitogen-activated protein kinase kinase 7 OS=Mus musculus GN=Map2k7 PE=1 SV=1

26.  Y1355_LACDA    Mass: 9109     Score: 30     Expect: 6e+002  Matches: 7
     UPF0291 protein Ldb1355 OS=Lactobacillus delbrueckii subsp. bulgaricus (strain ATCC 11842 / DSM 20081 / JCM 1002 / NBRC 13953 / NCIMB 11778) GN=Ldb1355 PE=3 SV=1

27.  RL11_ASHGO    Mass: 19848    Score: 30     Expect: 6e+002  Matches: 10
     60S ribosomal protein L11 OS=Ashbya gossypii (strain ATCC 10895 / CBS 109.51 / FGSC 9923 / NRRL Y-1056) GN=RPL11 PE=3 SV=1

28.  3SA1_NAJMO    Mass: 7285     Score: 30     Expect: 6.2e+002  Matches: 8
     Cytotoxin 1 OS=Naja mossambica PE=1 SV=1

29.  RL14_DESAG    Mass: 13269    Score: 29     Expect: 6.5e+002  Matches: 7
     50S ribosomal protein L14 OS=Desulfovibrio alaskensis (strain G20) GN=rplN PE=3 SV=1

30.  Y113_ADE07    Mass: 11656    Score: 29     Expect: 6.6e+002  Matches: 7
     Uncharacterized 11.3 kDa early protein OS=Human adenovirus B serotype 7 PE=4 SV=1

31.  Y1936_ARCFU    Mass: 26245    Score: 29     Expect: 6.9e+002  Matches: 8
     UPF0278 protein AF_1936 OS=Archaeoglobus fulgidus (strain ATCC 49558 / VC-16 / DSM 4304 / JCM 9628 / NBRC 100126) GN=AF_1936 PE=3 SV=1

32.  EFTS_THET8    Mass: 22515    Score: 29     Expect: 7.1e+002  Matches: 8
     Elongation factor Ts OS=Thermus thermophilus (strain HB8 / ATCC 27634 / DSM 579) GN=tsf PE=1 SV=1

33.  Y3756_GEOUR    Mass: 14558    Score: 29     Expect: 7.3e+002  Matches: 11
     UPF0102 protein Gura_3756 OS=Geobacter uraniireducens (strain Rf4) GN=Gura_3756 PE=3 SV=1

34.  Y1969_DESHY    Mass: 18518    Score: 29     Expect: 7.3e+002  Matches: 14
     UPF0234 protein DSY1969 OS=Desulfitobacterium hafniense (strain Y51) GN=DSY1969 PE=3 SV=2
     Y3127_DESHD    Mass: 18518    Score: 29     Expect: 7.3e+002  Matches: 14
     UPF0234 protein Dhaf_3127 OS=Desulfitobacterium hafniense (strain DCB-2 / DSM 10664) GN=Dhaf_3127 PE=3 SV=1

     UPF0234 protein Dhaf_3127 OS=Desulfitobacterium hafniense (strain DCB-2 / DSM 10664) GN=Dhaf_3127 PE=3 SV=1

35.  MYG_EQUBU    Mass: 17072    Score: 29     Expect: 7.6e+002  Matches: 13
     Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
     MYG_HORSE    Mass: 17072    Score: 29     Expect: 7.6e+002  Matches: 13
     Myoglobin OS=Equus caballus GN=MB PE=1 SV=2

36.  RS10_FERNB    Mass: 11654    Score: 28     Expect: 7.8e+002  Matches: 7
     30S ribosomal protein S10 OS=Fervidobacterium nodosum (strain ATCC 35602 / DSM 5306 / Rt17-B1) GN=rpsJ PE=3 SV=1

37.  NUOB2_CHLT3    Mass: 21902    Score: 28     Expect: 8e+002  Matches: 9
     NADH-quinone oxidoreductase subunit B 2 OS=Chloroherpeton thalassium (strain ATCC 35110 / GB-78) GN=nuoB2 PE=3 SV=1

38.  NQRC_VIBAL    Mass: 27744    Score: 28     Expect: 8.3e+002  Matches: 9
     Na(+)-translocating NADH-quinone reductase subunit C OS=Vibrio alginolyticus GN=nqrC PE=1 SV=3

39.  PDAD_META3    Mass: 17860    Score: 28     Expect: 8.5e+002  Matches: 7
     Pyruvoyl-dependent arginine decarboxylase OS=Methanococcus aeolicus (strain Nankai-3 / ATCC BAA-1280) GN=pdaD PE=3 SV=1

40.  NOL11_MOUSE    Mass: 81810    Score: 28     Expect: 8.7e+002  Matches: 13
     Nucleolar protein 11 OS=Mus musculus GN=Nol11 PE=2 SV=1

41.  PROA_VIBTL    Mass: 44794    Score: 28     Expect: 8.7e+002  Matches: 10
     Gamma-glutamyl phosphate reductase OS=Vibrio tasmaniensis (strain LGP32) GN=proA PE=3 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (832.7192,1+): <no title>
   Query2 (836.7806,1+): <no title>
   Query3 (836.8319,1+): <no title>
   Query4 (848.8024,1+): <no title>
```

MB mass/mass+c



Mascot Search Results

```
User           : Y Wang
Email          : kahn20003@gmail.com
Search title   :
Database       : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp      : 14 Apr 2016 at 07:22:29 GMT
Top Score      : 133 for MYG_EQUBU, Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

```
1.    MYG_EQUBU   Mass: 17072   Score: 133   Expect: 2.8e-008  Matches: 13
      Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
      MYG_HORSE   Mass: 17072   Score: 133   Expect: 2.8e-008  Matches: 13
      Myoglobin OS=Equus caballus GN=MB PE=1 SV=2
      MYG_HORSE   Mass: 17072   Score: 133   Expect: 2.8e-008  Matches: 13
      Myoglobin OS=Equus caballus GN=MB PE=1 SV=2
      MYG_CANLF   Mass: 17326   Score: 65    Expect: 0.19  Matches: 8
      Myoglobin OS=Canis lupus familiaris GN=MB PE=1 SV=2
      MYG_LYCPI   Mass: 17353   Score: 65    Expect: 0.19  Matches: 8
      Myoglobin OS=Lycaon pictus GN=MB PE=1 SV=2
      MYG_OTOME   Mass: 17326   Score: 65    Expect: 0.19  Matches: 8
      Myoglobin OS=Otocyon megalotis GN=MB PE=1 SV=2
      MYG_VULCH   Mass: 17326   Score: 65    Expect: 0.19  Matches: 8
      Myoglobin OS=Vulpes chama GN=MB PE=1 SV=2
      MYG_MACRU   Mass: 17258   Score: 51    Expect: 4  Matches: 7
      Myoglobin OS=Macropus rufus GN=MB PE=1 SV=2
      MYG_LEPMU   Mass: 17063   Score: 43    Expect: 26  Matches: 4
      Myoglobin OS=Lepilemur mustelinus GN=MB PE=1 SV=2
      MYG_TUPGL   Mass: 17088   Score: 43    Expect: 30  Matches: 4
      Myoglobin OS=Tupaia glis GN=MB PE=1 SV=2
```

```
2.    MYG_PERPO   Mass: 17052   Score: 94    Expect: 0.00022  Matches: 12
      Myoglobin OS=Perodicticus potto edwarsi GN=MB PE=1 SV=2
      MYG_MACFA   Mass: 17165   Score: 73    Expect: 0.027  Matches: 10
      Myoglobin OS=Macaca fascicularis GN=MB PE=1 SV=2
      MYG_NYCCO   Mass: 16944   Score: 58    Expect: 0.87  Matches: 7
      Myoglobin OS=Nycticebus coucang GN=MB PE=1 SV=2
      MYG_RABIT   Mass: 17210   Score: 56    Expect: 1.6  Matches: 7
      Myoglobin OS=Oryctolagus cuniculus GN=MB PE=1 SV=2
      MYG_ONDZI   Mass: 17257   Score: 55    Expect: 1.7  Matches: 7
      Myoglobin OS=Ondatra zibethicus GN=MB PE=1 SV=2
      MYG_OTOCR   Mass: 17091   Score: 55    Expect: 1.7  Matches: 6
      Myoglobin OS=Otolemur crassicaudatus GN=MB PE=1 SV=2
```

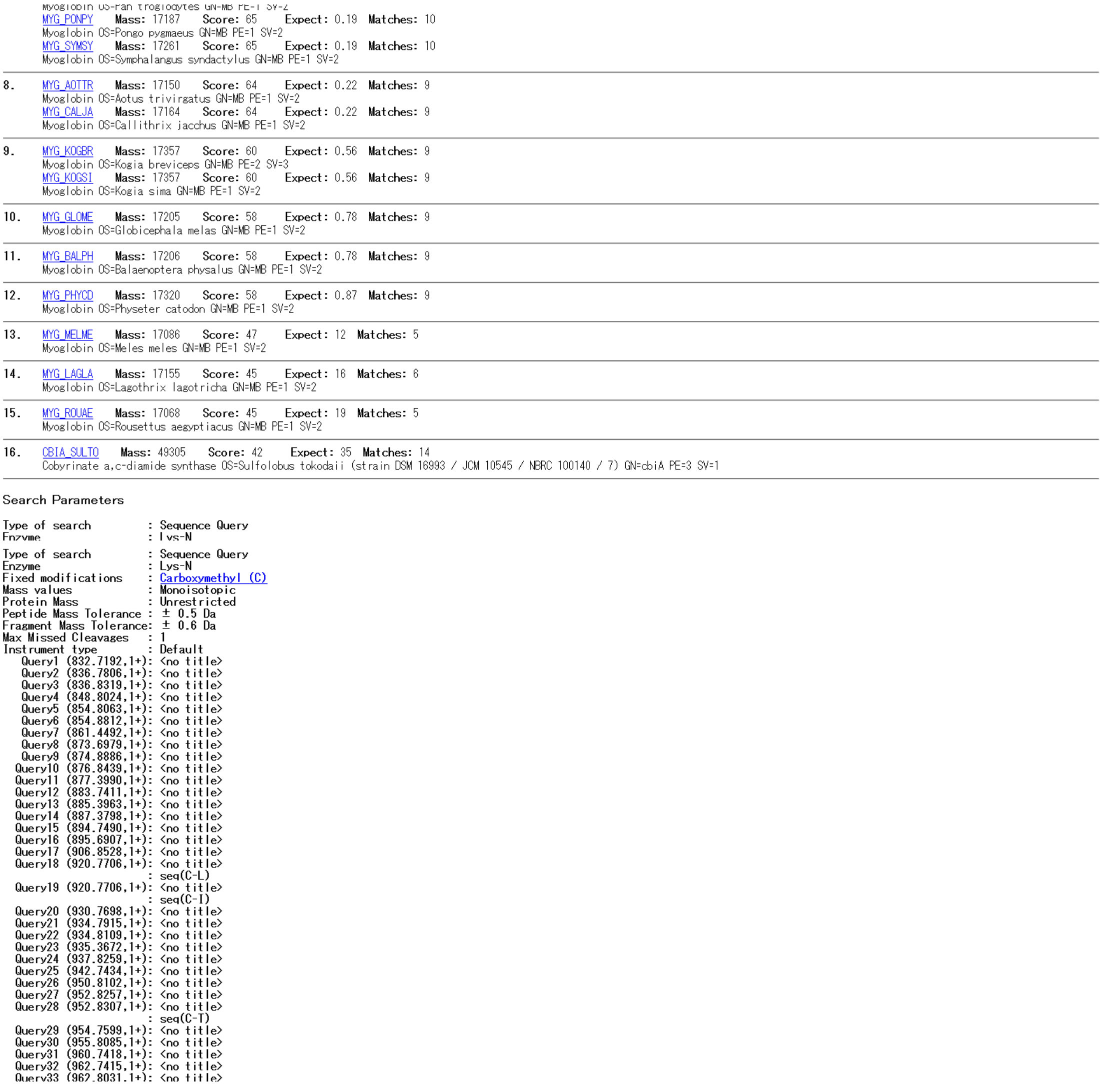
```
3.    MYG_INDPC   Mass: 17270   Score: 89    Expect: 0.00065  Matches: 12
      Myoglobin OS=Indopacetus pacificus GN=MB PE=2 SV=3
      MYG_MESCA   Mass: 17256   Score: 83    Expect: 0.0026  Matches: 11
      Myoglobin OS=Mesoplodon carlhubbsi GN=MB PE=1 SV=2
      MYG_MESST   Mass: 17256   Score: 83    Expect: 0.0026  Matches: 11
      Myoglobin OS=Mesoplodon stejnegeri GN=MB PE=2 SV=3
      MYG_ZIPCA   Mass: 17298   Score: 67    Expect: 0.12  Matches: 10
      Myoglobin OS=Ziphius cavirostris GN=MB PE=1 SV=2
      MYG_BALAC   Mass: 17275   Score: 53    Expect: 2.9  Matches: 8
      Myoglobin OS=Balaenoptera acutorostrata GN=MB PE=1 SV=2
      MYG_BALPO   Mass: 17238   Score: 53    Expect: 2.9  Matches: 8
      Myoglobin OS=Balaenoptera acutorostrata GN=MB PE=1 SV=2
      MYG_BALBO   Mass: 17238   Score: 53    Expect: 2.9  Matches: 8
      Myoglobin OS=Balaenoptera borealis GN=MB PE=2 SV=3
      MYG_BALED   Mass: 17238   Score: 53    Expect: 2.9  Matches: 8
      Myoglobin OS=Balaenoptera edeni GN=MB PE=2 SV=3
      MYG_DELLE   Mass: 17165   Score: 53    Expect: 2.9  Matches: 8
      Myoglobin OS=Delphinapterus leucas GN=MB PE=1 SV=2
      MYG_ESCRO   Mass: 17238   Score: 53    Expect: 2.9  Matches: 8
      Myoglobin OS=Eschrichtius robustus GN=MB PE=1 SV=2
      MYG_INIGE   Mass: 17191   Score: 53    Expect: 2.9  Matches: 8
      Myoglobin OS=Inia geoffrensis GN=MB PE=1 SV=2
      MYG_MEGNO   Mass: 17252   Score: 53    Expect: 2.9  Matches: 8
      Myoglobin OS=Megaptera novaeangliae GN=MB PE=1 SV=2
```

```
4.    MYG_HALGR   Mass: 17417   Score: 75    Expect: 0.018  Matches: 11
      Myoglobin OS=Halichoerus grypus GN=MB PE=1 SV=2
      MYG_PHOVI   Mass: 17417   Score: 75    Expect: 0.018  Matches: 11
      Myoglobin OS=Phoca vitulina GN=MB PE=1 SV=2
      MYG_PUSSI   Mass: 17472   Score: 70    Expect: 0.054  Matches: 9
      Myoglobin OS=Pusa sibirica GN=MB PE=1 SV=2
```

```
5.    MYG_CASFI   Mass: 17140   Score: 67    Expect: 0.12  Matches: 10
      Myoglobin OS=Castor fiber GN=MB PE=1 SV=2
```

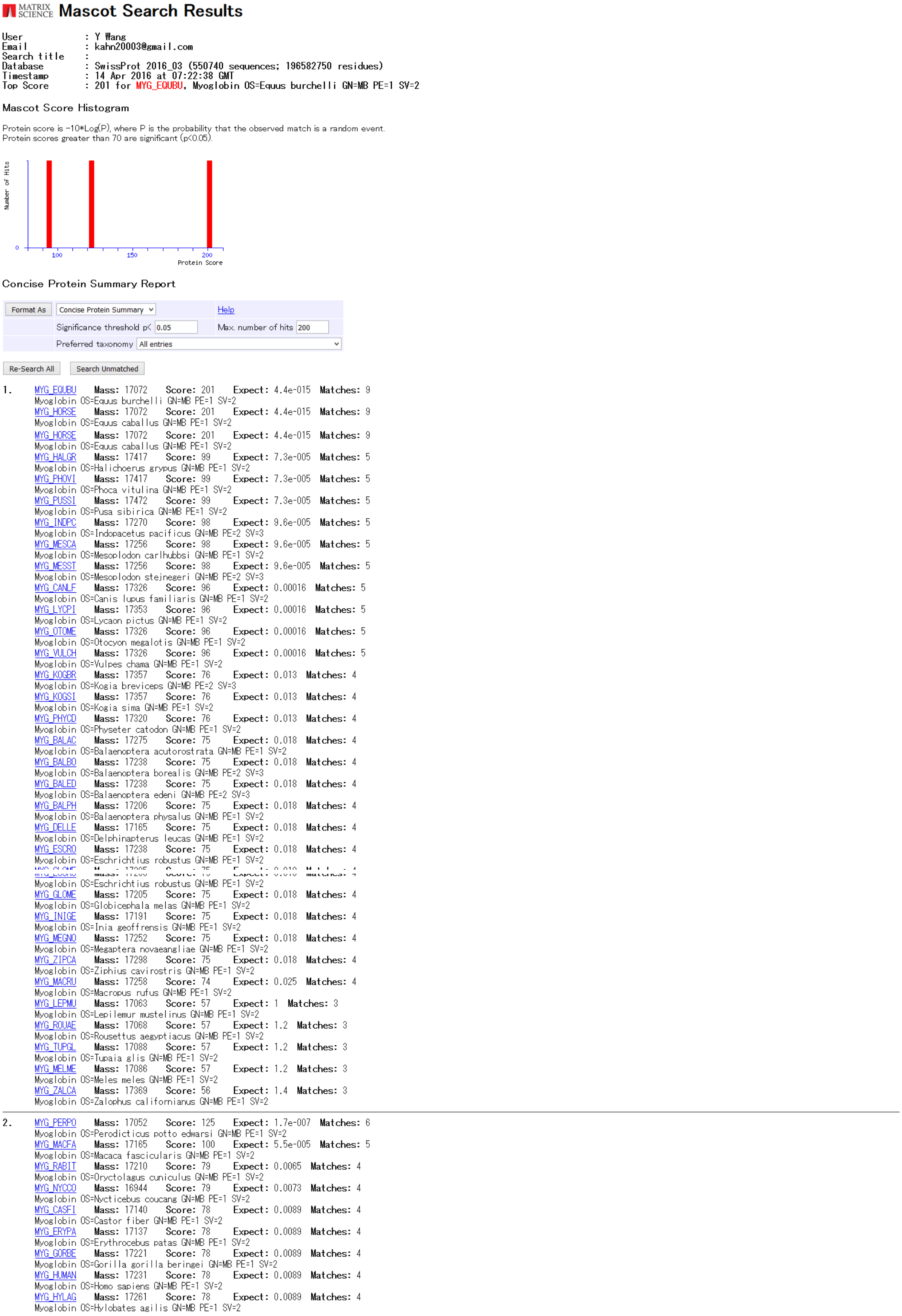
```
6.    MYG_ERYPA   Mass: 17137   Score: 66    Expect: 0.13  Matches: 10
      Myoglobin OS=Erythrocebus patas GN=MB PE=1 SV=2
      MYG_PAPAN   Mass: 17137   Score: 66    Expect: 0.13  Matches: 10
      Myoglobin OS=Papio anubis GN=MB PE=1 SV=2
      MYG_SEMEN   Mass: 17137   Score: 66    Expect: 0.13  Matches: 10
      Myoglobin OS=Semnopithecus entellus GN=MB PE=1 SV=2
```

```
7.    MYG_GORBE   Mass: 17221   Score: 65    Expect: 0.19  Matches: 10
      Myoglobin OS=Gorilla gorilla beringei GN=MB PE=1 SV=2
      MYG_HUMAN   Mass: 17231   Score: 65    Expect: 0.19  Matches: 10
      Myoglobin OS=Homo sapiens GN=MB PE=1 SV=2
      MYG_HYLAG   Mass: 17261   Score: 65    Expect: 0.19  Matches: 10
      Myoglobin OS=Hylobates agilis GN=MB PE=1 SV=2
      MYG_PANTR   Mass: 17240   Score: 65    Expect: 0.19  Matches: 10
      Myoglobin OS=Pan troglodytes GN=MB PE=1 SV=2
      MYG_PONPY   Mass: 17187   Score: 65    Expect: 0.19  Matches: 10
```



```
         Myoglobin OS=Pan troglodytes GN=MB PE=1 SV=2
         MYG_PONPY    Mass: 17187    Score: 65     Expect: 0.19  Matches: 10
         Myoglobin OS=Pongo pygmaeus GN=MB PE=1 SV=2
         MYG_SYMSY    Mass: 17261    Score: 65     Expect: 0.19  Matches: 10
         Myoglobin OS=Symphalangus syndactylus GN=MB PE=1 SV=2

8.       MYG_AOTTR    Mass: 17150    Score: 64     Expect: 0.22  Matches: 9
         Myoglobin OS=Aotus trivirgatus GN=MB PE=1 SV=2
         MYG_CALJA    Mass: 17164    Score: 64     Expect: 0.22  Matches: 9
         Myoglobin OS=Callithrix jacchus GN=MB PE=1 SV=2

9.       MYG_KOGBR    Mass: 17357    Score: 60     Expect: 0.56  Matches: 9
         Myoglobin OS=Kogia breviceps GN=MB PE=2 SV=3
         MYG_KOGSI    Mass: 17357    Score: 60     Expect: 0.56  Matches: 9
         Myoglobin OS=Kogia sima GN=MB PE=1 SV=2

10.      MYG_GLOME    Mass: 17205    Score: 58     Expect: 0.78  Matches: 9
         Myoglobin OS=Globicephala melas GN=MB PE=1 SV=2

11.      MYG_BALPH    Mass: 17206    Score: 58     Expect: 0.78  Matches: 9
         Myoglobin OS=Balaenoptera physalus GN=MB PE=1 SV=2

12.      MYG_PHYCD    Mass: 17320    Score: 58     Expect: 0.87  Matches: 9
         Myoglobin OS=Physeter catodon GN=MB PE=1 SV=2

13.      MYG_MELME    Mass: 17086    Score: 47     Expect: 12  Matches: 5
         Myoglobin OS=Meles meles GN=MB PE=1 SV=2

14.      MYG_LAGLA    Mass: 17155    Score: 45     Expect: 16  Matches: 6
         Myoglobin OS=Lagothrix lagotricha GN=MB PE=1 SV=2

15.      MYG_ROUAE    Mass: 17068    Score: 45     Expect: 19  Matches: 5
         Myoglobin OS=Rousettus aegyptiacus GN=MB PE=1 SV=2

16.      CBIA_SULTO    Mass: 49305    Score: 42     Expect: 35  Matches: 14
         Cobyrinate a,c-diamide synthase OS=Sulfolobus tokodaii (strain DSM 16993 / JCM 10545 / NBRC 100140 / 7) GN=cbiA PE=3 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (832.7192,1+): <no title>
   Query2 (836.7806,1+): <no title>
   Query3 (836.8319,1+): <no title>
   Query4 (848.8024,1+): <no title>
   Query5 (854.8063,1+): <no title>
   Query6 (854.8812,1+): <no title>
   Query7 (861.4492,1+): <no title>
   Query8 (873.6979,1+): <no title>
   Query9 (874.8886,1+): <no title>
  Query10 (876.8439,1+): <no title>
  Query11 (877.3990,1+): <no title>
  Query12 (883.7411,1+): <no title>
  Query13 (885.3963,1+): <no title>
  Query14 (887.3798,1+): <no title>
  Query15 (894.7490,1+): <no title>
  Query16 (895.6907,1+): <no title>
  Query17 (906.8528,1+): <no title>
  Query18 (920.7706,1+): <no title>
                       : seq(C-L)
  Query19 (920.7706,1+): <no title>
                       : seq(C-I)
  Query20 (930.7698,1+): <no title>
  Query21 (934.7915,1+): <no title>
  Query22 (934.8109,1+): <no title>
  Query23 (935.3672,1+): <no title>
  Query24 (937.8259,1+): <no title>
  Query25 (942.7434,1+): <no title>
  Query26 (950.8102,1+): <no title>
  Query27 (952.8257,1+): <no title>
  Query28 (952.8307,1+): <no title>
                       : seq(C-T)
  Query29 (954.7599,1+): <no title>
  Query30 (955.8085,1+): <no title>
  Query31 (960.7418,1+): <no title>
  Query32 (962.7415,1+): <no title>
  Query33 (962.8031,1+): <no title>
```

MB mass+c



Mascot Search Results

```
User         : Y Wang
Email        : kahn20003@gmail.com
Search title :
Database     : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp    : 14 Apr 2016 at 07:22:38 GMT
Top Score    : 201 for MYG_EQUBU, Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

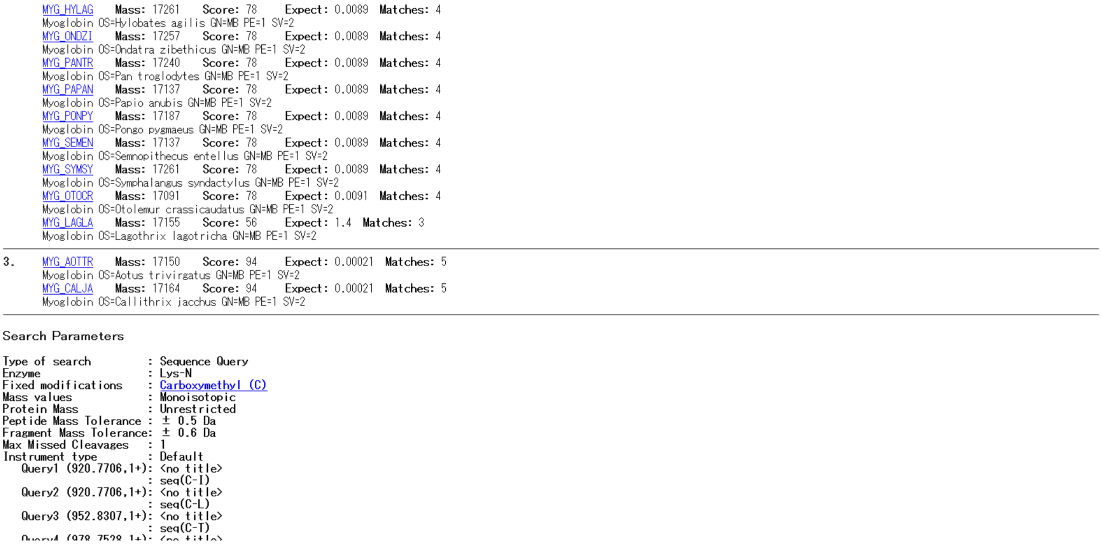
Concise Protein Summary Report

Significance threshold p< 0.05   Max. number of hits 200

Preferred taxonomy All entries

```
1.   MYG_EQUBU   Mass: 17072   Score: 201   Expect: 4.4e-015  Matches: 9
     Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
     MYG_HORSE   Mass: 17072   Score: 201   Expect: 4.4e-015  Matches: 9
     Myoglobin OS=Equus caballus GN=MB PE=1 SV=2
     MYG_HORSE   Mass: 17072   Score: 201   Expect: 4.4e-015  Matches: 9
     Myoglobin OS=Equus caballus GN=MB PE=1 SV=2
     MYG_HALGR   Mass: 17417   Score: 99    Expect: 7.3e-005  Matches: 5
     Myoglobin OS=Halichoerus grypus GN=MB PE=1 SV=2
     MYG_PHOVI   Mass: 17417   Score: 99    Expect: 7.3e-005  Matches: 5
     Myoglobin OS=Phoca vitulina GN=MB PE=1 SV=2
     MYG_PUSSI   Mass: 17472   Score: 99    Expect: 7.3e-005  Matches: 5
     Myoglobin OS=Pusa sibirica GN=MB PE=1 SV=2
     MYG_INDPC   Mass: 17270   Score: 98    Expect: 9.6e-005  Matches: 5
     Myoglobin OS=Indopacetus pacificus GN=MB PE=2 SV=3
     MYG_MESCA   Mass: 17256   Score: 98    Expect: 9.6e-005  Matches: 5
     Myoglobin OS=Mesoplodon carlhubbsi GN=MB PE=1 SV=2
     MYG_MESST   Mass: 17256   Score: 98    Expect: 9.6e-005  Matches: 5
     Myoglobin OS=Mesoplodon stejnegeri GN=MB PE=2 SV=3
     MYG_CANLF   Mass: 17326   Score: 96    Expect: 0.00016  Matches: 5
     Myoglobin OS=Canis lupus familiaris GN=MB PE=1 SV=2
     MYG_LYCPI   Mass: 17353   Score: 96    Expect: 0.00016  Matches: 5
     Myoglobin OS=Lycaon pictus GN=MB PE=1 SV=2
     MYG_OTOME   Mass: 17326   Score: 96    Expect: 0.00016  Matches: 5
     Myoglobin OS=Otocyon megalotis GN=MB PE=1 SV=2
     MYG_VULCH   Mass: 17326   Score: 96    Expect: 0.00016  Matches: 5
     Myoglobin OS=Vulpes chama GN=MB PE=1 SV=2
     MYG_KOGBR   Mass: 17357   Score: 76    Expect: 0.013  Matches: 4
     Myoglobin OS=Kogia breviceps GN=MB PE=2 SV=3
     MYG_KOGSI   Mass: 17357   Score: 76    Expect: 0.013  Matches: 4
     Myoglobin OS=Kogia sima GN=MB PE=1 SV=2
     MYG_PHYCD   Mass: 17320   Score: 76    Expect: 0.013  Matches: 4
     Myoglobin OS=Physeter catodon GN=MB PE=1 SV=2
     MYG_BALAC   Mass: 17275   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Balaenoptera acutorostrata GN=MB PE=1 SV=2
     MYG_BALBO   Mass: 17238   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Balaenoptera borealis GN=MB PE=2 SV=3
     MYG_BALED   Mass: 17238   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Balaenoptera edeni GN=MB PE=2 SV=3
     MYG_BALPH   Mass: 17206   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Balaenoptera physalus GN=MB PE=1 SV=2
     MYG_DELLE   Mass: 17165   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Delphinapterus leucas GN=MB PE=1 SV=2
     MYG_ESCRO   Mass: 17238   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Eschrichtius robustus GN=MB PE=1 SV=2
     Myoglobin OS=Eschrichtius robustus GN=MB PE=1 SV=2
     MYG_GLOME   Mass: 17205   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Globicephala melas GN=MB PE=1 SV=2
     MYG_INIGE   Mass: 17191   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Inia geoffrensis GN=MB PE=1 SV=2
     MYG_MEGNO   Mass: 17252   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Megaptera novaeangliae GN=MB PE=1 SV=2
     MYG_ZIPCA   Mass: 17298   Score: 75    Expect: 0.018  Matches: 4
     Myoglobin OS=Ziphius cavirostris GN=MB PE=1 SV=2
     MYG_MACRU   Mass: 17258   Score: 74    Expect: 0.025  Matches: 4
     Myoglobin OS=Macropus rufus GN=MB PE=1 SV=2
     MYG_LEPMU   Mass: 17063   Score: 57    Expect: 1  Matches: 3
     Myoglobin OS=Lepilemur mustelinus GN=MB PE=1 SV=2
     MYG_ROUAE   Mass: 17068   Score: 57    Expect: 1.2  Matches: 3
     Myoglobin OS=Rousettus aegyptiacus GN=MB PE=1 SV=2
     MYG_TUPGL   Mass: 17088   Score: 57    Expect: 1.2  Matches: 3
     Myoglobin OS=Tupaia glis GN=MB PE=1 SV=2
     MYG_MELME   Mass: 17086   Score: 57    Expect: 1.2  Matches: 3
     Myoglobin OS=Meles meles GN=MB PE=1 SV=2
     MYG_ZALCA   Mass: 17369   Score: 56    Expect: 1.4  Matches: 3
     Myoglobin OS=Zalophus californianus GN=MB PE=1 SV=2
```

```
2.   MYG_PERPO   Mass: 17052   Score: 125   Expect: 1.7e-007  Matches: 6
     Myoglobin OS=Perodicticus potto edwarsi GN=MB PE=1 SV=2
     MYG_MACFA   Mass: 17165   Score: 100   Expect: 5.5e-005  Matches: 5
     Myoglobin OS=Macaca fascicularis GN=MB PE=1 SV=2
     MYG_RABIT   Mass: 17210   Score: 79    Expect: 0.0065  Matches: 4
     Myoglobin OS=Oryctolagus cuniculus GN=MB PE=1 SV=2
     MYG_NYCCO   Mass: 16944   Score: 79    Expect: 0.0073  Matches: 4
     Myoglobin OS=Nycticebus coucang GN=MB PE=1 SV=2
     MYG_CASFI   Mass: 17140   Score: 78    Expect: 0.0089  Matches: 4
     Myoglobin OS=Castor fiber GN=MB PE=1 SV=2
     MYG_ERYPA   Mass: 17137   Score: 78    Expect: 0.0089  Matches: 4
     Myoglobin OS=Erythrocebus patas GN=MB PE=1 SV=2
     MYG_GORBE   Mass: 17221   Score: 78    Expect: 0.0089  Matches: 4
     Myoglobin OS=Gorilla gorilla beringei GN=MB PE=1 SV=2
     MYG_HUMAN   Mass: 17231   Score: 78    Expect: 0.0089  Matches: 4
     Myoglobin OS=Homo sapiens GN=MB PE=1 SV=2
     MYG_HYLAG   Mass: 17261   Score: 78    Expect: 0.0089  Matches: 4
     Myoglobin OS=Hylobates agilis GN=MB PE=1 SV=2
```



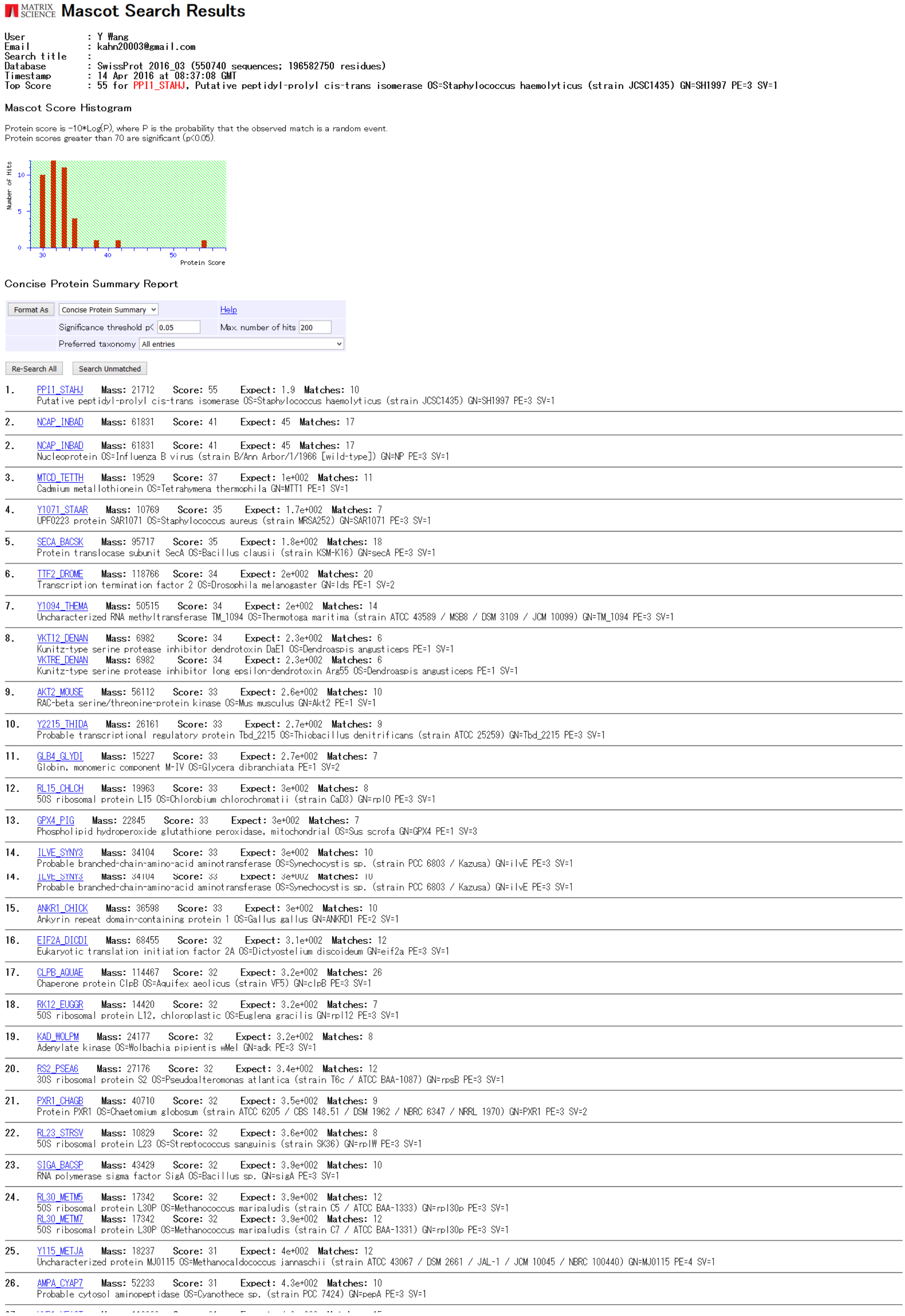
```
      MYG_HYLAG    Mass: 17261    Score: 78     Expect: 0.0089  Matches: 4
      Myoglobin OS=Hylobates agilis GN=MB PE=1 SV=2
      MYG_ONDZI    Mass: 17257    Score: 78     Expect: 0.0089  Matches: 4
      Myoglobin OS=Ondatra zibethicus GN=MB PE=1 SV=2
      MYG_PANTR    Mass: 17240    Score: 78     Expect: 0.0089  Matches: 4
      Myoglobin OS=Pan troglodytes GN=MB PE=1 SV=2
      MYG_PAPAN    Mass: 17137    Score: 78     Expect: 0.0089  Matches: 4
      Myoglobin OS=Papio anubis GN=MB PE=1 SV=2
      MYG_PONPY    Mass: 17187    Score: 78     Expect: 0.0089  Matches: 4
      Myoglobin OS=Pongo pygmaeus GN=MB PE=1 SV=2
      MYG_SEMEN    Mass: 17137    Score: 78     Expect: 0.0089  Matches: 4
      Myoglobin OS=Semnopithecus entellus GN=MB PE=1 SV=2
      MYG_SYMSY    Mass: 17261    Score: 78     Expect: 0.0089  Matches: 4
      Myoglobin OS=Symphalangus syndactylus GN=MB PE=1 SV=2
      MYG_OTOCR    Mass: 17091    Score: 78     Expect: 0.0091  Matches: 4
      Myoglobin OS=Otolemur crassicaudatus GN=MB PE=1 SV=2
      MYG_LAGLA    Mass: 17155    Score: 56     Expect: 1.4  Matches: 3
      Myoglobin OS=Lagothrix lagotricha GN=MB PE=1 SV=2
```

```
3.    MYG_AOTTR    Mass: 17150    Score: 94     Expect: 0.00021  Matches: 5
      Myoglobin OS=Aotus trivirgatus GN=MB PE=1 SV=2
      MYG_CALJA    Mass: 17164    Score: 94     Expect: 0.00021  Matches: 5
      Myoglobin OS=Callithrix jacchus GN=MB PE=1 SV=2
```

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (920.7706,1+): <no title>
                       : seq(C-I)
   Query2 (920.7706,1+): <no title>
                       : seq(C-L)
   Query3 (952.8307,1+): <no title>
                       : seq(C-T)
   Query4 (978.7528,1+): <no title>
```

MYH mass



Mascot Search Results

```
User            : Y Wang
Email           : kahn20003@gmail.com
Search title    :
Database        : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp       : 14 Apr 2016 at 08:37:08 GMT
Top Score       : 55 for PPI1_STAHJ, Putative peptidyl-prolyl cis-trans isomerase OS=Staphylococcus haemolyticus (strain JCSC1435) GN=SH1997 PE=3 SV=1
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

1. PPI1_STAHJ **Mass:** 21712 **Score:** 55 **Expect:** 1.9 **Matches:** 10
Putative peptidyl-prolyl cis-trans isomerase OS=Staphylococcus haemolyticus (strain JCSC1435) GN=SH1997 PE=3 SV=1

2. NCAP_INBAD **Mass:** 61831 **Score:** 41 **Expect:** 45 **Matches:** 17

2. NCAP_INBAD **Mass:** 61831 **Score:** 41 **Expect:** 45 **Matches:** 17
Nucleoprotein OS=Influenza B virus (strain B/Ann Arbor/1/1966 [wild-type]) GN=NP PE=3 SV=1

3. MTCD_TETTH **Mass:** 19529 **Score:** 37 **Expect:** 1e+002 **Matches:** 11
Cadmium metallothionein OS=Tetrahymena thermophila GN=MTT1 PE=1 SV=1

4. Y1071_STAAR **Mass:** 10769 **Score:** 35 **Expect:** 1.7e+002 **Matches:** 7
UPF0223 protein SAR1071 OS=Staphylococcus aureus (strain MRSA252) GN=SAR1071 PE=3 SV=1

5. SECA_BACSK **Mass:** 95717 **Score:** 35 **Expect:** 1.8e+002 **Matches:** 18
Protein translocase subunit SecA OS=Bacillus clausii (strain KSM-K16) GN=secA PE=3 SV=1

6. TTF2_DROME **Mass:** 118766 **Score:** 34 **Expect:** 2e+002 **Matches:** 20
Transcription termination factor 2 OS=Drosophila melanogaster GN=lds PE=1 SV=2

7. Y1094_THEMA **Mass:** 50515 **Score:** 34 **Expect:** 2e+002 **Matches:** 14
Uncharacterized RNA methyltransferase TM_1094 OS=Thermotoga maritima (strain ATCC 43589 / MSB8 / DSM 3109 / JCM 10099) GN=TM_1094 PE=3 SV=1

8. VKT12_DENAN **Mass:** 6982 **Score:** 34 **Expect:** 2.3e+002 **Matches:** 6
Kunitz-type serine protease inhibitor dendrotoxin DaE1 OS=Dendroaspis angusticeps PE=1 SV=1
VKTRE_DENAN **Mass:** 6982 **Score:** 34 **Expect:** 2.3e+002 **Matches:** 6
Kunitz-type serine protease inhibitor long epsilon-dendrotoxin Arg55 OS=Dendroaspis angusticeps PE=1 SV=1

9. AKT2_MOUSE **Mass:** 56112 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 10
RAC-beta serine/threonine-protein kinase OS=Mus musculus GN=Akt2 PE=1 SV=1

10. Y2215_THIDA **Mass:** 26161 **Score:** 33 **Expect:** 2.7e+002 **Matches:** 9
Probable transcriptional regulatory protein Tbd_2215 OS=Thiobacillus denitrificans (strain ATCC 25259) GN=Tbd_2215 PE=3 SV=1

11. GLB4_GLYDI **Mass:** 15227 **Score:** 33 **Expect:** 2.7e+002 **Matches:** 7
Globin, monomeric component M-IV OS=Glycera dibranchiata PE=1 SV=2

12. RL15_CHLCH **Mass:** 19963 **Score:** 33 **Expect:** 3e+002 **Matches:** 8
50S ribosomal protein L15 OS=Chlorobium chlorochromatii (strain CaD3) GN=rplO PE=3 SV=1

13. GPX4_PIG **Mass:** 22845 **Score:** 33 **Expect:** 3e+002 **Matches:** 7
Phospholipid hydroperoxide glutathione peroxidase, mitochondrial OS=Sus scrofa GN=GPX4 PE=1 SV=3

14. ILVE_SYNY3 **Mass:** 34104 **Score:** 33 **Expect:** 3e+002 **Matches:** 10
Probable branched-chain-amino-acid aminotransferase OS=Synechocystis sp. (strain PCC 6803 / Kazusa) GN=ilvE PE=3 SV=1

14. ILVE_SYNY3 **Mass:** 34104 **Score:** 33 **Expect:** 3e+002 **Matches:** 10
Probable branched-chain-amino-acid aminotransferase OS=Synechocystis sp. (strain PCC 6803 / Kazusa) GN=ilvE PE=3 SV=1

15. ANKR1_CHICK **Mass:** 36598 **Score:** 33 **Expect:** 3e+002 **Matches:** 10
Ankyrin repeat domain-containing protein 1 OS=Gallus gallus GN=ANKRD1 PE=2 SV=1

16. EIF2A_DICDI **Mass:** 68455 **Score:** 32 **Expect:** 3.1e+002 **Matches:** 12
Eukaryotic translation initiation factor 2A OS=Dictyostelium discoideum GN=eif2a PE=3 SV=1

17. CLPB_AQUAE **Mass:** 114467 **Score:** 32 **Expect:** 3.2e+002 **Matches:** 26
Chaperone protein ClpB OS=Aquifex aeolicus (strain VF5) GN=clpB PE=3 SV=1

18. RK12_EUGGR **Mass:** 14420 **Score:** 32 **Expect:** 3.2e+002 **Matches:** 7
50S ribosomal protein L12, chloroplastic OS=Euglena gracilis GN=rpl12 PE=3 SV=1

19. KAD_WOLPM **Mass:** 24177 **Score:** 32 **Expect:** 3.2e+002 **Matches:** 8
Adenylate kinase OS=Wolbachia pipientis wMel GN=adk PE=3 SV=1

20. RS2_PSEA6 **Mass:** 27176 **Score:** 32 **Expect:** 3.4e+002 **Matches:** 12
30S ribosomal protein S2 OS=Pseudoalteromonas atlantica (strain T6c / ATCC BAA-1087) GN=rpsB PE=3 SV=1

21. PXR1_CHAGB **Mass:** 40710 **Score:** 32 **Expect:** 3.5e+002 **Matches:** 9
Protein PXR1 OS=Chaetomium globosum (strain ATCC 6205 / CBS 148.51 / DSM 1962 / NBRC 6347 / NRRL 1970) GN=PXR1 PE=3 SV=2

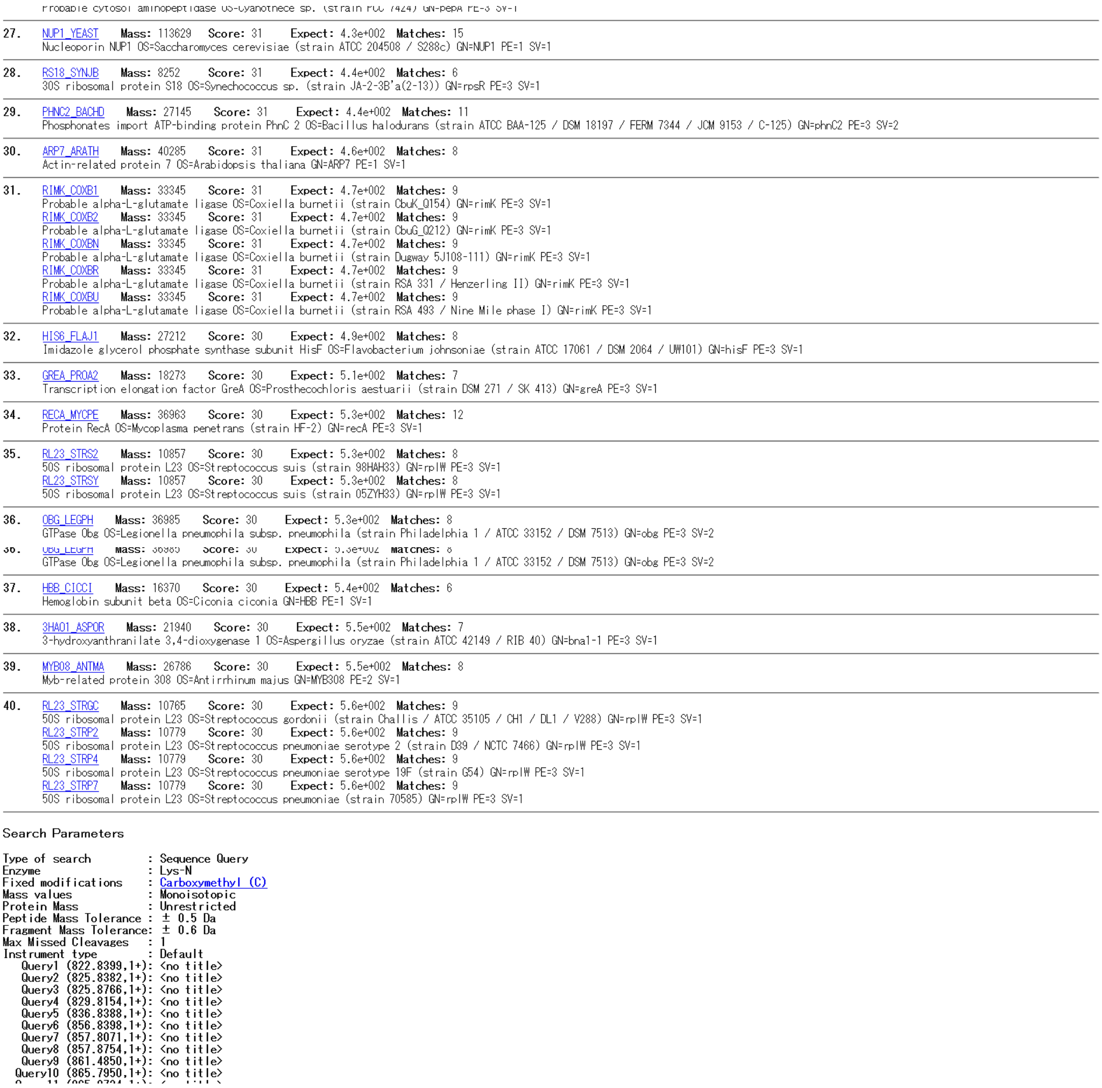
22. RL23_STRSV **Mass:** 10829 **Score:** 32 **Expect:** 3.6e+002 **Matches:** 8
50S ribosomal protein L23 OS=Streptococcus sanguinis (strain SK36) GN=rplW PE=3 SV=1

23. SIGA_BACSP **Mass:** 43429 **Score:** 32 **Expect:** 3.9e+002 **Matches:** 10
RNA polymerase sigma factor SigA OS=Bacillus sp. GN=sigA PE=3 SV=1

24. RL30_METM5 **Mass:** 17342 **Score:** 32 **Expect:** 3.9e+002 **Matches:** 12
50S ribosomal protein L30P OS=Methanococcus maripaludis (strain C5 / ATCC BAA-1333) GN=rpl30p PE=3 SV=1
RL30_METM7 **Mass:** 17342 **Score:** 32 **Expect:** 3.9e+002 **Matches:** 12
50S ribosomal protein L30P OS=Methanococcus maripaludis (strain C7 / ATCC BAA-1331) GN=rpl30p PE=3 SV=1

25. Y115_METJA **Mass:** 18237 **Score:** 31 **Expect:** 4e+002 **Matches:** 12
Uncharacterized protein MJ0115 OS=Methanocaldococcus jannaschii (strain ATCC 43067 / DSM 2661 / JAL-1 / JCM 10045 / NBRC 100440) GN=MJ0115 PE=4 SV=1

26. AMPA_CYAP7 **Mass:** 52233 **Score:** 31 **Expect:** 4.3e+002 **Matches:** 10
Probable cytosol aminopeptidase OS=Cyanothece sp. (strain PCC 7424) GN=pepA PE=3 SV=1



```
     Probable cytosol aminopeptidase OS=Cyanothece sp. (strain PCC 7424) GN=pepA PE=3 SV=1

27.  NUP1_YEAST    Mass: 113629   Score: 31     Expect: 4.3e+002  Matches: 15
     Nucleoporin NUP1 OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=NUP1 PE=1 SV=1

28.  RS18_SYNJB    Mass: 8252     Score: 31     Expect: 4.4e+002  Matches: 6
     30S ribosomal protein S18 OS=Synechococcus sp. (strain JA-2-3B'a(2-13)) GN=rpsR PE=3 SV=1

29.  PHNC2_BACHD    Mass: 27145    Score: 31     Expect: 4.4e+002  Matches: 11
     Phosphonates import ATP-binding protein PhnC 2 OS=Bacillus halodurans (strain ATCC BAA-125 / DSM 18197 / FERM 7344 / JCM 9153 / C-125) GN=phnC2 PE=3 SV=2

30.  ARP7_ARATH    Mass: 40285    Score: 31     Expect: 4.6e+002  Matches: 8
     Actin-related protein 7 OS=Arabidopsis thaliana GN=ARP7 PE=1 SV=1

31.  RIMK_COXB1    Mass: 33345    Score: 31     Expect: 4.7e+002  Matches: 9
     Probable alpha-L-glutamate ligase OS=Coxiella burnetii (strain CbuK_Q154) GN=rimK PE=3 SV=1
     RIMK_COXB2    Mass: 33345    Score: 31     Expect: 4.7e+002  Matches: 9
     Probable alpha-L-glutamate ligase OS=Coxiella burnetii (strain CbuG_Q212) GN=rimK PE=3 SV=1
     RIMK_COXBN    Mass: 33345    Score: 31     Expect: 4.7e+002  Matches: 9
     Probable alpha-L-glutamate ligase OS=Coxiella burnetii (strain Dugway 5J108-111) GN=rimK PE=3 SV=1
     RIMK_COXBR    Mass: 33345    Score: 31     Expect: 4.7e+002  Matches: 9
     Probable alpha-L-glutamate ligase OS=Coxiella burnetii (strain RSA 331 / Henzerling II) GN=rimK PE=3 SV=1
     RIMK_COXBU    Mass: 33345    Score: 31     Expect: 4.7e+002  Matches: 9
     Probable alpha-L-glutamate ligase OS=Coxiella burnetii (strain RSA 493 / Nine Mile phase I) GN=rimK PE=3 SV=1

32.  HIS6_FLAJ1    Mass: 27212    Score: 30     Expect: 4.9e+002  Matches: 8
     Imidazole glycerol phosphate synthase subunit HisF OS=Flavobacterium johnsoniae (strain ATCC 17061 / DSM 2064 / UW101) GN=hisF PE=3 SV=1

33.  GREA_PROA2    Mass: 18273    Score: 30     Expect: 5.1e+002  Matches: 7
     Transcription elongation factor GreA OS=Prosthecochloris aestuarii (strain DSM 271 / SK 413) GN=greA PE=3 SV=1

34.  RECA_MYCPE    Mass: 36963    Score: 30     Expect: 5.3e+002  Matches: 12
     Protein RecA OS=Mycoplasma penetrans (strain HF-2) GN=recA PE=3 SV=1

35.  RL23_STRS2    Mass: 10857    Score: 30     Expect: 5.3e+002  Matches: 8
     50S ribosomal protein L23 OS=Streptococcus suis (strain 98HAH33) GN=rplW PE=3 SV=1
     RL23_STRSY    Mass: 10857    Score: 30     Expect: 5.3e+002  Matches: 8
     50S ribosomal protein L23 OS=Streptococcus suis (strain 05ZYH33) GN=rplW PE=3 SV=1

36.  OBG_LEGPH    Mass: 36985    Score: 30     Expect: 5.3e+002  Matches: 8
     GTPase Obg OS=Legionella pneumophila subsp. pneumophila (strain Philadelphia 1 / ATCC 33152 / DSM 7513) GN=obg PE=3 SV=2
36.  OBG_LEGPH    Mass: 36985    Score: 30     Expect: 5.3e+002  Matches: 8
     GTPase Obg OS=Legionella pneumophila subsp. pneumophila (strain Philadelphia 1 / ATCC 33152 / DSM 7513) GN=obg PE=3 SV=2

37.  HBB_CICCI    Mass: 16370    Score: 30     Expect: 5.4e+002  Matches: 6
     Hemoglobin subunit beta OS=Ciconia ciconia GN=HBB PE=1 SV=1

38.  3HAO1_ASPOR    Mass: 21940    Score: 30     Expect: 5.5e+002  Matches: 7
     3-hydroxyanthranilate 3,4-dioxygenase 1 OS=Aspergillus oryzae (strain ATCC 42149 / RIB 40) GN=bna1-1 PE=3 SV=1

39.  MYB08_ANTMA    Mass: 26786    Score: 30     Expect: 5.5e+002  Matches: 8
     Myb-related protein 308 OS=Antirrhinum majus GN=MYB308 PE=2 SV=1

40.  RL23_STRGC    Mass: 10765    Score: 30     Expect: 5.6e+002  Matches: 9
     50S ribosomal protein L23 OS=Streptococcus gordonii (strain Challis / ATCC 35105 / CH1 / DL1 / V288) GN=rplW PE=3 SV=1
     RL23_STRP2    Mass: 10779    Score: 30     Expect: 5.6e+002  Matches: 9
     50S ribosomal protein L23 OS=Streptococcus pneumoniae serotype 2 (strain D39 / NCTC 7466) GN=rplW PE=3 SV=1
     RL23_STRP4    Mass: 10779    Score: 30     Expect: 5.6e+002  Matches: 9
     50S ribosomal protein L23 OS=Streptococcus pneumoniae serotype 19F (strain G54) GN=rplW PE=3 SV=1
     RL23_STRP7    Mass: 10779    Score: 30     Expect: 5.6e+002  Matches: 9
     50S ribosomal protein L23 OS=Streptococcus pneumoniae (strain 70585) GN=rplW PE=3 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (822.8399,1+): <no title>
   Query2 (825.8382,1+): <no title>
   Query3 (825.8766,1+): <no title>
   Query4 (829.8154,1+): <no title>
   Query5 (836.8388,1+): <no title>
   Query6 (856.8398,1+): <no title>
   Query7 (857.8071,1+): <no title>
   Query8 (857.8754,1+): <no title>
   Query9 (861.4850,1+): <no title>
  Query10 (865.7950,1+): <no title>
```

MYH mass/mass+c

Mascot Search Results

```
User            : Y Wang
Email           : kahn20003@gmail.com
Search title    :
Database        : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp       : 14 Apr 2016 at 08:36:17 GMT
Top Score       : 143 for MYH1_PIG, Myosin-1 OS=Sus scrofa GN=MYH1 PE=2 SV=1
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Format As: Concise Protein Summary

Significance threshold p< 0.05

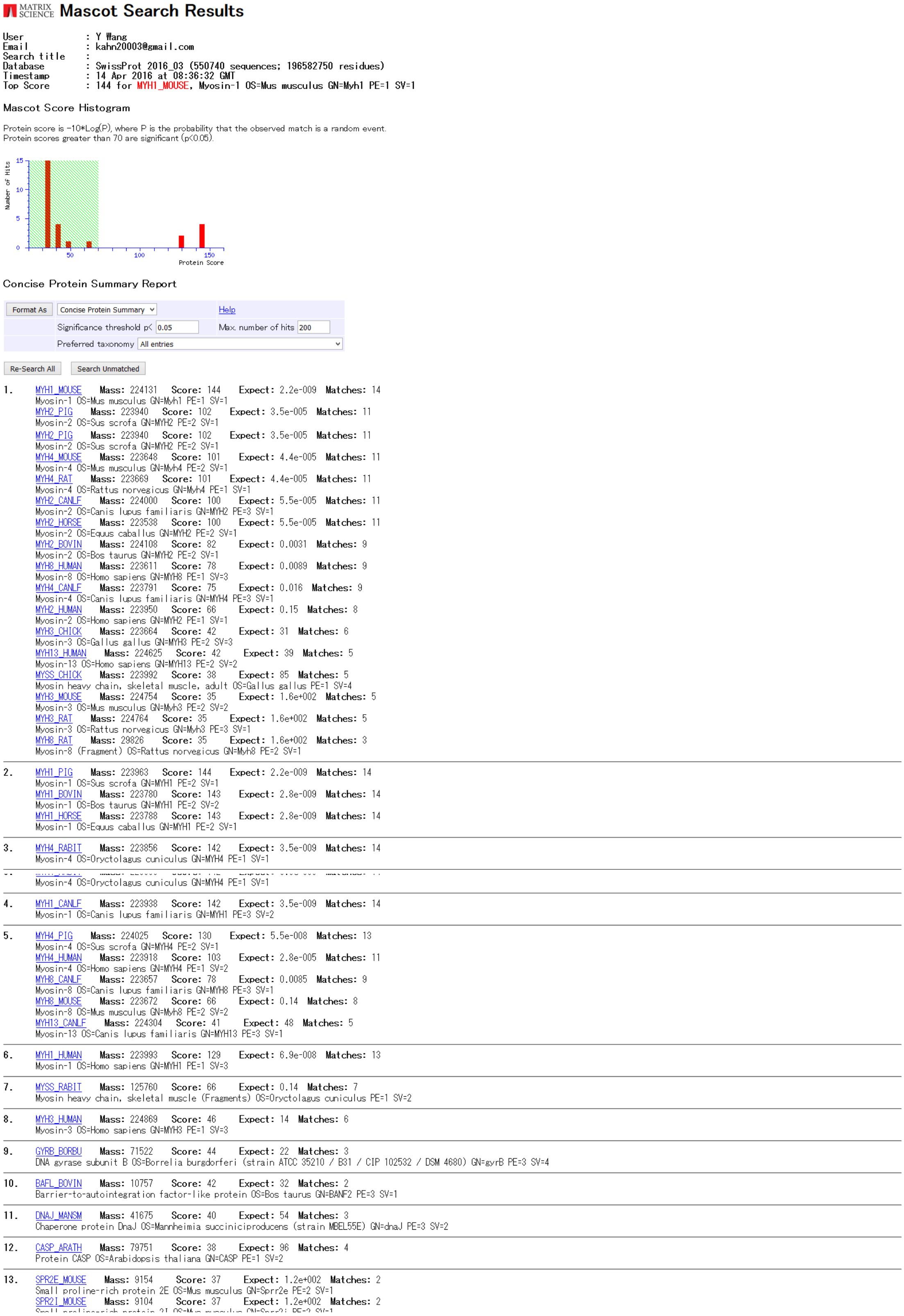
Max. number of hits 200

Preferred taxonomy All entries

```
1.    MYH1_PIG     Mass: 223963   Score: 143    Expect: 2.8e-009  Matches: 32
      Myosin-1 OS=Sus scrofa GN=MYH1 PE=2 SV=1

2.    MYH1_BOVIN     Mass: 223780   Score: 142    Expect: 3.5e-009  Matches: 31

2.    MYH1_BOVIN     Mass: 223780   Score: 142    Expect: 3.5e-009  Matches: 31
      Myosin-1 OS=Bos taurus GN=MYH1 PE=2 SV=2

3.    MYH1_CANLF     Mass: 223938   Score: 138    Expect: 8.7e-009  Matches: 32
      Myosin-1 OS=Canis lupus familiaris GN=MYH1 PE=3 SV=2

4.    MYH1_MOUSE     Mass: 224131   Score: 138    Expect: 8.7e-009  Matches: 33
      Myosin-1 OS=Mus musculus GN=Myh1 PE=1 SV=1

5.    MYH4_RABIT     Mass: 223856   Score: 134    Expect: 2.2e-008  Matches: 30
      Myosin-4 OS=Oryctolagus cuniculus GN=MYH4 PE=1 SV=1

6.    MYH1_HORSE     Mass: 223788   Score: 132    Expect: 3.5e-008  Matches: 30
      Myosin-1 OS=Equus caballus GN=MYH1 PE=2 SV=1

7.    MYH1_HUMAN     Mass: 223993   Score: 129    Expect: 6.9e-008  Matches: 35
      Myosin-1 OS=Homo sapiens GN=MYH1 PE=1 SV=3

8.    MYH4_PIG     Mass: 224025   Score: 127    Expect: 1.1e-007  Matches: 32
      Myosin-4 OS=Sus scrofa GN=MYH4 PE=2 SV=1

9.    MYH2_CANLF     Mass: 224000   Score: 99     Expect: 7.3e-005  Matches: 23
      Myosin-2 OS=Canis lupus familiaris GN=MYH2 PE=3 SV=1

10.   MYH2_PIG     Mass: 223940   Score: 98     Expect: 8.3e-005  Matches: 22
      Myosin-2 OS=Sus scrofa GN=MYH2 PE=2 SV=1

11.   MYH2_HORSE     Mass: 223538   Score: 96     Expect: 0.00014  Matches: 25
      Myosin-2 OS=Equus caballus GN=MYH2 PE=2 SV=1

12.   MYH4_MOUSE     Mass: 223648   Score: 94     Expect: 0.0002  Matches: 29
      Myosin-4 OS=Mus musculus GN=Myh4 PE=2 SV=1
      MYH4_RAT     Mass: 223669   Score: 94     Expect: 0.00022  Matches: 29
      Myosin-4 OS=Rattus norvegicus GN=Myh4 PE=1 SV=1

13.   MYH4_HUMAN     Mass: 223918   Score: 94     Expect: 0.00021  Matches: 30
      Myosin-4 OS=Homo sapiens GN=MYH4 PE=1 SV=2

14.   MYH8_HUMAN     Mass: 223611   Score: 90     Expect: 0.00058  Matches: 19
      Myosin-8 OS=Homo sapiens GN=MYH8 PE=1 SV=3
14.   MYH8_HUMAN     Mass: 223611   Score: 90     Expect: 0.00058  Matches: 19
      Myosin-8 OS=Homo sapiens GN=MYH8 PE=1 SV=3

15.   MYH2_BOVIN     Mass: 224108   Score: 89     Expect: 0.00065  Matches: 23
      Myosin-2 OS=Bos taurus GN=MYH2 PE=2 SV=1

16.   MYH8_CANLF     Mass: 223657   Score: 84     Expect: 0.0024  Matches: 22
      Myosin-8 OS=Canis lupus familiaris GN=MYH8 PE=3 SV=1

17.   MYH4_CANLF     Mass: 223791   Score: 70     Expect: 0.05  Matches: 26
      Myosin-4 OS=Canis lupus familiaris GN=MYH4 PE=3 SV=1

18.   MYH8_MOUSE     Mass: 223672   Score: 70     Expect: 0.056  Matches: 22
      Myosin-8 OS=Mus musculus GN=Myh8 PE=2 SV=2

19.   MYH2_HUMAN     Mass: 223950   Score: 70     Expect: 0.06  Matches: 24
      Myosin-2 OS=Homo sapiens GN=MYH2 PE=1 SV=1

20.   MYSS_RABIT     Mass: 125760   Score: 55     Expect: 1.7  Matches: 21
      Myosin heavy chain, skeletal muscle (Fragments) OS=Oryctolagus cuniculus PE=1 SV=2

21.   MYH13_HUMAN     Mass: 224625   Score: 45     Expect: 17  Matches: 20
      Myosin-13 OS=Homo sapiens GN=MYH13 PE=2 SV=2

22.   MYH13_CANLF     Mass: 224304   Score: 44     Expect: 22  Matches: 20
      Myosin-13 OS=Canis lupus familiaris GN=MYH13 PE=3 SV=1

23.   RS6E_METST     Mass: 14431    Score: 42     Expect: 32  Matches: 6
      30S ribosomal protein S6e OS=Methanosphaera stadtmanae (strain ATCC 43021 / DSM 3091 / JCM 11832 / MCB-3) GN=rps6e PE=3 SV=1

24.   PPI1_STAHJ     Mass: 21712    Score: 42     Expect: 33  Matches: 8
      Putative peptidyl-prolyl cis-trans isomerase OS=Staphylococcus haemolyticus (strain JCSC1435) GN=SH1997 PE=3 SV=1

25.   PTP14_STYPL     Mass: 13836    Score: 42     Expect: 38  Matches: 4
      Tyrosine-protein phosphatase 14 (Fragment) OS=Styela plicata GN=STY-14 PE=2 SV=1

26.   USE1_YEAST     Mass: 28089    Score: 41     Expect: 48  Matches: 5
      Protein transport protein USE1 OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=USE1 PE=1 SV=1

27.   GYRB_BORBU     Mass: 71522    Score: 40     Expect: 53  Matches: 7
      DNA gyrase subunit B OS=Borrelia burgdorferi (strain ATCC 35210 / B31 / CIP 102532 / DSM 4680) GN=gyrB PE=3 SV=4
```

```
     DNA gyrase subunit B OS=Borrelia burgdorferi (strain ATCC 35210 / B31 / CIP 102532 / DSM 4680) GN=gyrB PE=3 SV=4

28.  MTND_YEAST   Mass: 21098   Score: 40   Expect: 54  Matches: 4
     1,2-dihydroxy-3-keto-5-methylthiopentene dioxygenase OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=ADI1 PE=1 SV=1

29.  DNAJ_MANSM   Mass: 41675   Score: 39   Expect: 63  Matches: 7
     Chaperone protein DnaJ OS=Mannheimia succiniciproducens (strain MBEL55E) GN=dnaJ PE=3 SV=2

30.  EFP_FRASN   Mass: 20055   Score: 39   Expect: 68  Matches: 5
     Elongation factor P OS=Frankia sp. (strain EAN1pec) GN=efp PE=3 SV=1

31.  MYH3_MOUSE   Mass: 224754   Score: 39   Expect: 74  Matches: 19
     Myosin-3 OS=Mus musculus GN=Myh3 PE=2 SV=2

32.  SYY_STRUO   Mass: 47186   Score: 38   Expect: 89  Matches: 8
     Tyrosine--tRNA ligase OS=Streptococcus uberis (strain ATCC BAA-854 / 0140J) GN=tyrS PE=3 SV=1

33.  YCX8_CYAPA   Mass: 20097   Score: 37   Expect: 1.1e+002  Matches: 5
     Uncharacterized protein in petA-psaM intergenic region OS=Cyanophora paradoxa PE=1 SV=1

34.  TAL_DESHD   Mass: 23021   Score: 37   Expect: 1.1e+002  Matches: 4
     Probable transaldolase OS=Desulfitobacterium hafniense (strain DCB-2 / DSM 10664) GN=tal PE=3 SV=1
     TAL_DESHY   Mass: 23021   Score: 37   Expect: 1.1e+002  Matches: 4
     Probable transaldolase OS=Desulfitobacterium hafniense (strain Y51) GN=tal PE=3 SV=1

35.  YL287_YEAST   Mass: 41192   Score: 37   Expect: 1.2e+002  Matches: 10
     Uncharacterized protein YLR287C OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=YLR287C PE=1 SV=1

36.  CSE15_BACSU   Mass: 37704   Score: 36   Expect: 1.4e+002  Matches: 8
     Sporulation protein cse15 OS=Bacillus subtilis (strain 168) GN=cse15 PE=2 SV=1

37.  RAD50_SCHPO   Mass: 150212   Score: 36   Expect: 1.4e+002  Matches: 18
     DNA repair protein rad50 OS=Schizosaccharomyces pombe (strain 972 / ATCC 24843) GN=rad50 PE=1 SV=3

38.  BAFL_BOVIN   Mass: 10757   Score: 36   Expect: 1.4e+002  Matches: 2
     Barrier-to-autointegration factor-like protein OS=Bos taurus GN=BANF2 PE=3 SV=1

39.  Y1071_STAAR   Mass: 10769   Score: 36   Expect: 1.5e+002  Matches: 6
     UPF0223 protein SAR1071 OS=Staphylococcus aureus (strain MRSA252) GN=SAR1071 PE=3 SV=1

40.  MUTS_LEPBJ   Mass: 97582   Score: 36   Expect: 1.6e+002  Matches: 16
     DNA mismatch repair protein MutS OS=Leptospira borgpetersenii serovar Hardjo-bovis (strain JB197) GN=mutS PE=3 SV=1
     MUTS_LEPBL   Mass: 97582   Score: 36   Expect: 1.6e+002  Matches: 16
     DNA mismatch repair protein MutS OS=Leptospira borgpetersenii serovar Hardjo-bovis (strain L550) GN=mutS PE=3 SV=1

41.  MYH3_CHICK   Mass: 223664   Score: 35   Expect: 1.6e+002  Matches: 24
     Myosin-3 OS=Gallus gallus GN=MYH3 PE=2 SV=3

42.  RDR6_ARATH   Mass: 138234   Score: 35   Expect: 1.7e+002  Matches: 8
     RNA-dependent RNA polymerase 6 OS=Arabidopsis thaliana GN=RDR6 PE=1 SV=1

43.  SLA1_DEIAC   Mass: 17712   Score: 35   Expect: 1.8e+002  Matches: 4
     Snaclec agglucetin subunit alpha-1 OS=Deinagkistrodon acutus PE=1 SV=1

44.  AT5F1_HUMAN   Mass: 28948   Score: 35   Expect: 1.9e+002  Matches: 6
     ATP synthase F(0) complex subunit B1, mitochondrial OS=Homo sapiens GN=ATP5F1 PE=1 SV=2

45.  PINH_ECOLI   Mass: 9187   Score: 35   Expect: 1.9e+002  Matches: 3
     Putative DNA-invertase from prophage CP4-44 (Fragment) OS=Escherichia coli (strain K12) GN=pinH PE=5 SV=2

46.  RK23_PSEAK   Mass: 11021   Score: 34   Expect: 2e+002  Matches: 5
     50S ribosomal protein L23, chloroplastic OS=Pseudendoclonium akinetum GN=rpl23 PE=3 SV=1

47.  SPR2E_MOUSE   Mass: 9154   Score: 34   Expect: 2.1e+002  Matches: 3
     Small proline-rich protein 2E OS=Mus musculus GN=Sprr2e PE=2 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (822.8399,1+): <no title>
                       : seq(C-F)
   Query2 (825.8382,1+): <no title>
```

MYH mass+c



Mascot Search Results

```
User           : Y Wang
Email          : kahn20003@gmail.com
Search title   :
Database       : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp      : 14 Apr 2016 at 08:36:32 GMT
Top Score      : 144 for MYH1_MOUSE, Myosin-1 OS=Mus musculus GN=Myh1 PE=1 SV=1
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

```
1.    MYH1_MOUSE    Mass: 224131   Score: 144    Expect: 2.2e-009  Matches: 14
      Myosin-1 OS=Mus musculus GN=Myh1 PE=1 SV=1
      MYH2_PIG    Mass: 223940   Score: 102    Expect: 3.5e-005  Matches: 11
      Myosin-2 OS=Sus scrofa GN=MYH2 PE=2 SV=1
      MYH2_PIG    Mass: 223940   Score: 102    Expect: 3.5e-005  Matches: 11
      Myosin-2 OS=Sus scrofa GN=MYH2 PE=2 SV=1
      MYH4_MOUSE    Mass: 223648   Score: 101    Expect: 4.4e-005  Matches: 11
      Myosin-4 OS=Mus musculus GN=Myh4 PE=2 SV=1
      MYH4_RAT    Mass: 223669   Score: 101    Expect: 4.4e-005  Matches: 11
      Myosin-4 OS=Rattus norvegicus GN=Myh4 PE=1 SV=1
      MYH2_CANLF    Mass: 224000   Score: 100    Expect: 5.5e-005  Matches: 11
      Myosin-2 OS=Canis lupus familiaris GN=MYH2 PE=3 SV=1
      MYH2_HORSE    Mass: 223538   Score: 100    Expect: 5.5e-005  Matches: 11
      Myosin-2 OS=Equus caballus GN=MYH2 PE=2 SV=1
      MYH2_BOVIN    Mass: 224108   Score: 82     Expect: 0.0031  Matches: 9
      Myosin-2 OS=Bos taurus GN=MYH2 PE=2 SV=1
      MYH8_HUMAN    Mass: 223611   Score: 78     Expect: 0.0089  Matches: 9
      Myosin-8 OS=Homo sapiens GN=MYH8 PE=1 SV=3
      MYH4_CANLF    Mass: 223791   Score: 75     Expect: 0.016  Matches: 9
      Myosin-4 OS=Canis lupus familiaris GN=MYH4 PE=3 SV=1
      MYH2_HUMAN    Mass: 223950   Score: 66     Expect: 0.15  Matches: 8
      Myosin-2 OS=Homo sapiens GN=MYH2 PE=1 SV=1
      MYH3_CHICK    Mass: 223664   Score: 42     Expect: 31  Matches: 6
      Myosin-3 OS=Gallus gallus GN=MYH3 PE=2 SV=3
      MYH13_HUMAN    Mass: 224625   Score: 42     Expect: 39  Matches: 5
      Myosin-13 OS=Homo sapiens GN=MYH13 PE=2 SV=2
      MYSS_CHICK    Mass: 223992   Score: 38     Expect: 85  Matches: 5
      Myosin heavy chain, skeletal muscle, adult OS=Gallus gallus PE=1 SV=4
      MYH3_MOUSE    Mass: 224754   Score: 35     Expect: 1.6e+002  Matches: 5
      Myosin-3 OS=Mus musculus GN=Myh3 PE=2 SV=2
      MYH3_RAT    Mass: 224764   Score: 35     Expect: 1.6e+002  Matches: 5
      Myosin-3 OS=Rattus norvegicus GN=Myh3 PE=3 SV=1
      MYH8_RAT    Mass: 29826    Score: 35     Expect: 1.6e+002  Matches: 3
      Myosin-8 (Fragment) OS=Rattus norvegicus GN=Myh8 PE=2 SV=1

2.    MYH1_PIG    Mass: 223963   Score: 144    Expect: 2.2e-009  Matches: 14
      Myosin-1 OS=Sus scrofa GN=MYH1 PE=2 SV=1
      MYH1_BOVIN    Mass: 223780   Score: 143    Expect: 2.8e-009  Matches: 14
      Myosin-1 OS=Bos taurus GN=MYH1 PE=2 SV=2
      MYH1_HORSE    Mass: 223788   Score: 143    Expect: 2.8e-009  Matches: 14
      Myosin-1 OS=Equus caballus GN=MYH1 PE=2 SV=1

3.    MYH4_RABIT    Mass: 223856   Score: 142    Expect: 3.5e-009  Matches: 14
      Myosin-4 OS=Oryctolagus cuniculus GN=MYH4 PE=1 SV=1
```

Myosin-4 OS=Oryctolagus cuniculus GN=MYH4 PE=1 SV=1

```
4.    MYH1_CANLF    Mass: 223938   Score: 142    Expect: 3.5e-009  Matches: 14
      Myosin-1 OS=Canis lupus familiaris GN=MYH1 PE=3 SV=2

5.    MYH4_PIG    Mass: 224025   Score: 130    Expect: 5.5e-008  Matches: 13
      Myosin-4 OS=Sus scrofa GN=MYH4 PE=2 SV=1
      MYH4_HUMAN    Mass: 223918   Score: 103    Expect: 2.8e-005  Matches: 11
      Myosin-4 OS=Homo sapiens GN=MYH4 PE=1 SV=2
      MYH8_CANLF    Mass: 223657   Score: 78     Expect: 0.0085  Matches: 9
      Myosin-8 OS=Canis lupus familiaris GN=MYH8 PE=3 SV=1
      MYH8_MOUSE    Mass: 223672   Score: 66     Expect: 0.14  Matches: 8
      Myosin-8 OS=Mus musculus GN=Myh8 PE=2 SV=2
      MYH13_CANLF    Mass: 224304   Score: 41     Expect: 48  Matches: 5
      Myosin-13 OS=Canis lupus familiaris GN=MYH13 PE=3 SV=1

6.    MYH1_HUMAN    Mass: 223993   Score: 129    Expect: 6.9e-008  Matches: 13
      Myosin-1 OS=Homo sapiens GN=MYH1 PE=1 SV=3

7.    MYSS_RABIT    Mass: 125760   Score: 66     Expect: 0.14  Matches: 7
      Myosin heavy chain, skeletal muscle (Fragments) OS=Oryctolagus cuniculus PE=1 SV=2

8.    MYH3_HUMAN    Mass: 224869   Score: 46     Expect: 14  Matches: 6
      Myosin-3 OS=Homo sapiens GN=MYH3 PE=1 SV=3

9.    GYRB_BORBU    Mass: 71522    Score: 44     Expect: 22  Matches: 3
      DNA gyrase subunit B OS=Borrelia burgdorferi (strain ATCC 35210 / B31 / CIP 102532 / DSM 4680) GN=gyrB PE=3 SV=4

10.   BAFL_BOVIN    Mass: 10757    Score: 42     Expect: 32  Matches: 2
      Barrier-to-autointegration factor-like protein OS=Bos taurus GN=BANF2 PE=3 SV=1

11.   DNAJ_MANSM    Mass: 41675    Score: 40     Expect: 54  Matches: 3
      Chaperone protein DnaJ OS=Mannheimia succiniciproducens (strain MBEL55E) GN=dnaJ PE=3 SV=2

12.   CASP_ARATH    Mass: 79751    Score: 38     Expect: 96  Matches: 4
      Protein CASP OS=Arabidopsis thaliana GN=CASP PE=1 SV=2

13.   SPR2E_MOUSE    Mass: 9154     Score: 37     Expect: 1.2e+002  Matches: 2
      Small proline-rich protein 2E OS=Mus musculus GN=Sprr2e PE=2 SV=1
      SPR2I_MOUSE    Mass: 9104     Score: 37     Expect: 1.2e+002  Matches: 2
```

```
    SPR2I_MOUSE    Mass: 9104     Score: 37     Expect: 1.2e+002  Matches: 2
    Small proline-rich protein 2I OS=Mus musculus GN=Sprr2i PE=2 SV=1

14. PINH_ECOLI    Mass: 9187     Score: 36     Expect: 1.3e+002  Matches: 2
    Putative DNA-invertase from prophage CP4-44 (Fragment) OS=Escherichia coli (strain K12) GN=pinH PE=5 SV=2

15. PTP14_STYPL    Mass: 13836    Score: 36     Expect: 1.4e+002  Matches: 2
    Tyrosine-protein phosphatase 14 (Fragment) OS=Styela plicata GN=STY-14 PE=2 SV=1

16. HSLU_PSEMY    Mass: 49846    Score: 36     Expect: 1.4e+002  Matches: 3
    ATP-dependent protease ATPase subunit HslU OS=Pseudomonas mendocina (strain ymp) GN=hslU PE=3 SV=1

17. YDDA_BACSU    Mass: 12075    Score: 36     Expect: 1.6e+002  Matches: 2
    Uncharacterized protein YddA OS=Bacillus subtilis (strain 168) GN=yddA PE=4 SV=1

18. TCAL7_RAT    Mass: 12100    Score: 35     Expect: 1.7e+002  Matches: 2
    Transcription elongation factor A protein-like 7 OS=Rattus norvegicus GN=Tceal7 PE=3 SV=1

19. EFP_FRASN    Mass: 20055    Score: 35     Expect: 1.7e+002  Matches: 2
    Elongation factor P OS=Frankia sp. (strain EAN1pec) GN=efp PE=3 SV=1

20. MTND_YEAST    Mass: 21098    Score: 35     Expect: 1.8e+002  Matches: 2
    1,2-dihydroxy-3-keto-5-methylthiopentene dioxygenase OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=ADI1 PE=1 SV=1

21. ATG10_VANPO    Mass: 22918    Score: 35     Expect: 1.9e+002  Matches: 2
    Ubiquitin-like-conjugating enzyme ATG10 OS=Vanderwaltozyma polyspora (strain ATCC 22028 / DSM 70294) GN=ATG10 PE=3 SV=1

22. TRPA_CHLTR    Mass: 28270    Score: 35     Expect: 1.9e+002  Matches: 2
    Tryptophan synthase alpha chain OS=Chlamydia trachomatis (strain D/UW-3/Cx) GN=trpA PE=3 SV=1

23. PROF_HELAN    Mass: 14450    Score: 35     Expect: 1.9e+002  Matches: 2
    Profilin OS=Helianthus annuus PE=1 SV=1

24. TAL_DESHD    Mass: 23021    Score: 34     Expect: 2e+002  Matches: 2
    Probable transaldolase OS=Desulfitobacterium hafniense (strain DCB-2 / DSM 10664) GN=tal PE=3 SV=1
    TAL_DESHY    Mass: 23021    Score: 34     Expect: 2e+002  Matches: 2
    Probable transaldolase OS=Desulfitobacterium hafniense (strain Y51) GN=tal PE=3 SV=1

25. CH10_CALS4    Mass: 10270    Score: 34     Expect: 2.2e+002  Matches: 2
    10 kDa chaperonin OS=Caldanaerobacter subterraneus subsp. tengcongensis (strain DSM 15242 / JCM 11007 / NBRC 100824 / MB4) GN=groS PE=3 SV=1

    10 kDa chaperonin OS=Caldanaerobacter subterraneus subsp. tengcongensis (strain DSM 15242 / JCM 11007 / NBRC 100824 / MB4) GN=groS PE=3 SV=1

26. MPU1_MOUSE    Mass: 26770    Score: 34     Expect: 2.2e+002  Matches: 2
    Mannose-P-dolichol utilization defect 1 protein OS=Mus musculus GN=Mpdu1 PE=1 SV=1

27. ALB1_PSOTE    Mass: 19436    Score: 34     Expect: 2.3e+002  Matches: 2
    Albumin-1 OS=Psophocarpus tetragonolobus PE=1 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (822.8399,1+): <no title>
                       : seq(C-F)
   Query2 (825.8766,1+): <no title>
                       : seq(C-V)
   Query3 (836.8388,1+): <no title>
                       : seq(C-Y)
   Query4 (856.8398,1+): <no title>
                       : seq(C-T)
   Query5 (857.8071,1+): <no title>
                       : seq(C-Y)
   Query6 (857.8754,1+): <no title>
                       : seq(C-I)
   Query7 (857.8754,1+): <no title>
                       : seq(C-L)
   Query8 (865.8724,1+): <no title>
                       : seq(C-K)
   Query9 (865.8724,1+): <no title>
                       : seq(C-Q)
  Query10 (897.8425,1+): <no title>
                       : seq(C-F)
  Query11 (907.7966,1+): <no title>
                       : seq(C-K)
  Query12 (907.7966,1+): <no title>
                       : seq(C-Q)
```

PYGM mass

Mascot Search Results

```
User           : Y Wang
Email          : kahn20003@gmail.com
Search title   :
Database       : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp      : 14 Apr 2016 at 09:10:30 GMT
Top Score      : 39 for HIS2_METFK, Phosphoribosyl-ATP pyrophosphatase OS=Methylobacillus flagellatus (strain KT / ATCC 51484 / DSM 6875) GN=hisE PE=3 SV=1
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Format As: Concise Protein Summary
Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

```
1.    HIS2_METFK    Mass: 11579    Score: 39    Expect: 68  Matches: 7
      Phosphoribosyl-ATP pyrophosphatase OS=Methylobacillus flagellatus (strain KT / ATCC 51484 / DSM 6875) GN=hisE PE=3 SV=1

2.    PSTB_SACD2    Mass: 32647    Score: 38    Expect: 94  Matches: 9

2.    PSTB_SACD2    Mass: 32647    Score: 38    Expect: 94  Matches: 9
      Phosphate import ATP-binding protein PstB OS=Saccharophagus degradans (strain 2-40 / ATCC 43961 / DSM 17024) GN=pstB PE=3 SV=1

3.    RHOU_BOVIN    Mass: 28502    Score: 36    Expect: 1.3e+002  Matches: 10
      Rho-related GTP-binding protein RhoU OS=Bos taurus GN=RHOU PE=2 SV=1

4.    ERA_LACLA    Mass: 34773    Score: 35    Expect: 1.7e+002  Matches: 13
      GTPase Era OS=Lactococcus lactis subsp. lactis (strain IL1403) GN=era PE=3 SV=1
      ERA_LACLM    Mass: 34773    Score: 35    Expect: 1.7e+002  Matches: 13
      GTPase Era OS=Lactococcus lactis subsp. cremoris (strain MG1363) GN=era PE=3 SV=1
      ERA_LACLS    Mass: 34787    Score: 34    Expect: 2e+002  Matches: 13
      GTPase Era OS=Lactococcus lactis subsp. cremoris (strain SK11) GN=era PE=3 SV=1

5.    RAP1_PHYPO    Mass: 21313    Score: 35    Expect: 1.7e+002  Matches: 8
      Ras-related protein Rap-1 OS=Physarum polycephalum GN=RAP1 PE=2 SV=1

6.    RXT3_YEAST    Mass: 33907    Score: 34    Expect: 2e+002  Matches: 10
      Transcriptional regulatory protein RXT3 OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=RXT3 PE=1 SV=1

7.    RL1_CARHZ    Mass: 24778    Score: 34    Expect: 2e+002  Matches: 10
      50S ribosomal protein L1 OS=Carboxydothermus hydrogenoformans (strain ATCC BAA-161 / DSM 6008 / Z-2901) GN=rplA PE=3 SV=1

8.    NIR_EMENI    Mass: 124019   Score: 33    Expect: 2.7e+002  Matches: 23
      Nitrite reductase [NAD(P)H] OS=Emericella nidulans (strain FGSC A4 / ATCC 38163 / CBS 112.46 / NRRL 194 / M139) GN=niiA PE=3 SV=2

9.    PGK2_METFV    Mass: 35072    Score: 32    Expect: 3.2e+002  Matches: 9
      2-phosphoglycerate kinase OS=Methanothermus fervidus (strain ATCC 43054 / DSM 2088 / JCM 10308 / V24 S) GN=pgk2 PE=1 SV=1

10.   TRMD_FRATN    Mass: 28391    Score: 32    Expect: 3.3e+002  Matches: 12
      tRNA (guanine-N(1)-)-methyltransferase OS=Francisella tularensis subsp. novicida (strain U112) GN=trmD PE=3 SV=1

11.   PYRH_METS5    Mass: 24295    Score: 32    Expect: 3.4e+002  Matches: 7
      Uridylate kinase OS=Metallosphaera sedula (strain ATCC 51363 / DSM 5348) GN=pyrH PE=3 SV=1

12.   ARR8_ARATH    Mass: 25629    Score: 32    Expect: 3.7e+002  Matches: 9
      Two-component response regulator ARR8 OS=Arabidopsis thaliana GN=ARR8 PE=1 SV=1

13.   ARGB_BACAH    Mass: 27604    Score: 31    Expect: 4e+002  Matches: 12
      Acetylglutamate kinase OS=Bacillus thuringiensis (strain Al Hakam) GN=argB PE=3 SV=2

      Acetylglutamate kinase OS=Bacillus thuringiensis (strain Al Hakam) GN=argB PE=3 SV=2

14.   ILVC_HELPY    Mass: 36685    Score: 31    Expect: 4.3e+002  Matches: 13
      Ketol-acid reductoisomerase OS=Helicobacter pylori (strain ATCC 700392 / 26695) GN=ilvC PE=3 SV=1

15.   ALBA_METMJ    Mass: 9922     Score: 31    Expect: 4.4e+002  Matches: 8
      DNA/RNA-binding protein Alba OS=Methanoculleus marisnigri (strain ATCC 35101 / DSM 1498 / JR1) GN=albA PE=3 SV=1

16.   Y988_METJA    Mass: 37757    Score: 31    Expect: 4.8e+002  Matches: 10
      Uncharacterized protein MJ0988 OS=Methanocaldococcus jannaschii (strain ATCC 43067 / DSM 2661 / JAL-1 / JCM 10045 / NBRC 100440) GN=MJ0988 PE=4 SV=1

17.   RL28_LACC3    Mass: 6973     Score: 30    Expect: 5.1e+002  Matches: 7
      50S ribosomal protein L28 OS=Lactobacillus casei (strain ATCC 334) GN=rpmB PE=3 SV=1
      RL28_LACCB    Mass: 6973     Score: 30    Expect: 5.1e+002  Matches: 7
      50S ribosomal protein L28 OS=Lactobacillus casei (strain BL23) GN=rpmB PE=3 SV=1

18.   ALBU_PONAB    Mass: 71465    Score: 30    Expect: 5.3e+002  Matches: 18
      Serum albumin OS=Pongo abelii GN=ALB PE=2 SV=2

19.   APT_CHLPM    Mass: 19180    Score: 30    Expect: 5.6e+002  Matches: 6
      Adenine phosphoribosyltransferase OS=Chlorobium phaeovibrioides (strain DSM 265 / 1930) GN=apt PE=3 SV=1

20.   MURB_RICBR    Mass: 33322    Score: 30    Expect: 5.8e+002  Matches: 12
      UDP-N-acetylenolpyruvoylglucosamine reductase OS=Rickettsia bellii (strain RML369-C) GN=murB PE=3 SV=1

21.   CHLB_PROMS    Mass: 59218    Score: 30    Expect: 5.9e+002  Matches: 12
      Light-independent protochlorophyllide reductase subunit B OS=Prochlorococcus marinus (strain AS9601) GN=chlB PE=3 SV=1

22.   SURA_VIBPA    Mass: 47464    Score: 29    Expect: 6.3e+002  Matches: 12
      Chaperone SurA OS=Vibrio parahaemolyticus serotype O3:K6 (strain RIMD 2210633) GN=surA PE=3 SV=1

23.   FABA_VIBC3    Mass: 19146    Score: 29    Expect: 6.3e+002  Matches: 6
      3-hydroxydecanoyl-[acyl-carrier-protein] dehydratase OS=Vibrio cholerae serotype O1 (strain ATCC 39541 / Classical Ogawa 395 / O395) GN=fabA PE=3 SV=1
      FABA_VIBCH    Mass: 19146    Score: 29    Expect: 6.3e+002  Matches: 6
      3-hydroxydecanoyl-[acyl-carrier-protein] dehydratase OS=Vibrio cholerae serotype O1 (strain ATCC 39315 / El Tor Inaba N16961) GN=fabA PE=3 SV=1
      FABA_VIBCM    Mass: 19146    Score: 29    Expect: 6.3e+002  Matches: 6
      3-hydroxydecanoyl-[acyl-carrier-protein] dehydratase OS=Vibrio cholerae serotype O1 (strain M66-2) GN=fabA PE=3 SV=1

24.   MAM3_ARATH    Mass: 55832    Score: 29    Expect: 6.3e+002  Matches: 15
      Methylthioalkylmalate synthase 3, chloroplastic OS=Arabidopsis thaliana GN=MAM3 PE=1 SV=1
```

```
     Methylthioalkylmalate synthase 3, chloroplastic OS=Arabidopsis thaliana GN=MAM3 PE=1 SV=1
------------------------------------------------------------------------------------------------
25.  AEP2_YEAS1    Mass: 68173    Score: 29     Expect: 6.5e+002  Matches: 15
     ATPase expression protein 2, mitochondrial OS=Saccharomyces cerevisiae (strain RM11-1a) GN=AEP2 PE=3 SV=1
     AEP2_YEAS6    Mass: 68146    Score: 29     Expect: 6.5e+002  Matches: 15
     ATPase expression protein 2, mitochondrial OS=Saccharomyces cerevisiae (strain AWRI1631) GN=AEP2 PE=3 SV=1
     AEP2_YEAS8    Mass: 68173    Score: 29     Expect: 6.5e+002  Matches: 15
     ATPase expression protein 2, mitochondrial OS=Saccharomyces cerevisiae (strain Lalvin EC1118 / Prise de mousse) GN=AEP2 PE=3 SV=1
------------------------------------------------------------------------------------------------
26.  KAD_PSEFS    Mass: 23309    Score: 29     Expect: 6.8e+002  Matches: 6
     Adenylate kinase OS=Pseudomonas fluorescens (strain SBW25) GN=adk PE=3 SV=1
------------------------------------------------------------------------------------------------
27.  Y6387_DICDI    Mass: 4917     Score: 29     Expect: 7.1e+002  Matches: 7
     Putative uncharacterized protein DDB_G0285187 OS=Dictyostelium discoideum GN=DDB_G0285187 PE=4 SV=1
------------------------------------------------------------------------------------------------
28.  AROD_STAA1    Mass: 27063    Score: 29     Expect: 7.1e+002  Matches: 10
     3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain Mu3 / ATCC 700698) GN=aroD PE=3 SV=1
     AROD_STAA2    Mass: 27063    Score: 29     Expect: 7.1e+002  Matches: 10
     3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain JH1) GN=aroD PE=3 SV=1
     AROD_STAA9    Mass: 27063    Score: 29     Expect: 7.1e+002  Matches: 10
     3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain JH9) GN=aroD PE=3 SV=1
     AROD_STAAM    Mass: 27063    Score: 29     Expect: 7.1e+002  Matches: 10
     3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain Mu50 / ATCC 700699) GN=aroD PE=3 SV=1
     AROD_STAAN    Mass: 27063    Score: 29     Expect: 7.1e+002  Matches: 10
     3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain N315) GN=aroD PE=3 SV=1
------------------------------------------------------------------------------------------------
29.  MDH_ECO7I    Mass: 32491    Score: 29     Expect: 7.3e+002  Matches: 10
     Malate dehydrogenase OS=Escherichia coli O7:K1 (strain IAI39 / ExPEC) GN=mdh PE=3 SV=1
     MDH_ECOLU    Mass: 32490    Score: 29     Expect: 7.3e+002  Matches: 10
     Malate dehydrogenase OS=Escherichia coli O17:K52:H18 (strain UMN026 / ExPEC) GN=mdh PE=3 SV=1
     MDH_ECOSM    Mass: 32491    Score: 29     Expect: 7.3e+002  Matches: 10
     Malate dehydrogenase OS=Escherichia coli (strain SMS-3-5 / SECEC) GN=mdh PE=3 SV=1
------------------------------------------------------------------------------------------------
30.  NIA2_ARATH    Mass: 103706   Score: 29     Expect: 7.4e+002  Matches: 15
     Nitrate reductase [NADH] 2 OS=Arabidopsis thaliana GN=NIA2 PE=1 SV=1
------------------------------------------------------------------------------------------------
31.  SYC_LACAC    Mass: 55092    Score: 29     Expect: 7.4e+002  Matches: 11
     Cysteine--tRNA ligase OS=Lactobacillus acidophilus (strain ATCC 700396 / NCK56 / N2 / NCFM) GN=cysS PE=3 SV=1
------------------------------------------------------------------------------------------------
32.  YG3W_YEAST    Mass: 44504    Score: 29     Expect: 7.4e+002  Matches: 13
     Uncharacterized protein YGR168C OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=YGR168C PE=4 SV=1
32.  YG3W_YEAST    Mass: 44504    Score: 29     Expect: 7.4e+002  Matches: 13
     Uncharacterized protein YGR168C OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=YGR168C PE=4 SV=1
------------------------------------------------------------------------------------------------
33.  BUCC_APLCA    Mass: 53483    Score: 29     Expect: 7.6e+002  Matches: 9
     Buccalin OS=Aplysia californica PE=1 SV=2
------------------------------------------------------------------------------------------------
34.  AEP2_YEAS7    Mass: 68231    Score: 28     Expect: 7.8e+002  Matches: 17
     ATPase expression protein 2, mitochondrial OS=Saccharomyces cerevisiae (strain YJM789) GN=AEP2 PE=3 SV=1
------------------------------------------------------------------------------------------------
35.  RL17_MYCMO    Mass: 13613    Score: 28     Expect: 8.1e+002  Matches: 7
     50S ribosomal protein L17 OS=Mycoplasma mobile (strain ATCC 43663 / 163K / NCTC 11711) GN=rplQ PE=3 SV=1
------------------------------------------------------------------------------------------------
36.  ATPF_GUITH    Mass: 20228    Score: 28     Expect: 8.9e+002  Matches: 10
     ATP synthase subunit b, chloroplastic OS=Guillardia theta GN=atpF PE=3 SV=1
------------------------------------------------------------------------------------------------
37.  RL35A_MOUSE    Mass: 12662    Score: 28     Expect: 9.4e+002  Matches: 7
     60S ribosomal protein L35a OS=Mus musculus GN=Rpl35a PE=1 SV=2
------------------------------------------------------------------------------------------------

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (822.8594,1+): <no title>
   Query2 (825.8621,1+): <no title>
   Query3 (829.8763,1+): <no title>
   Query4 (831.8586,1+): <no title>
   Query5 (831.8587,1+): <no title>
   Query6 (832.8786,1+): <no title>
   Query7 (837.8337,1+): <no title>
   Query8 (845.8452,1+): <no title>
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  Query10 (854.3198,1+): <no title>
  Query11 (855.9137,1+): <no title>
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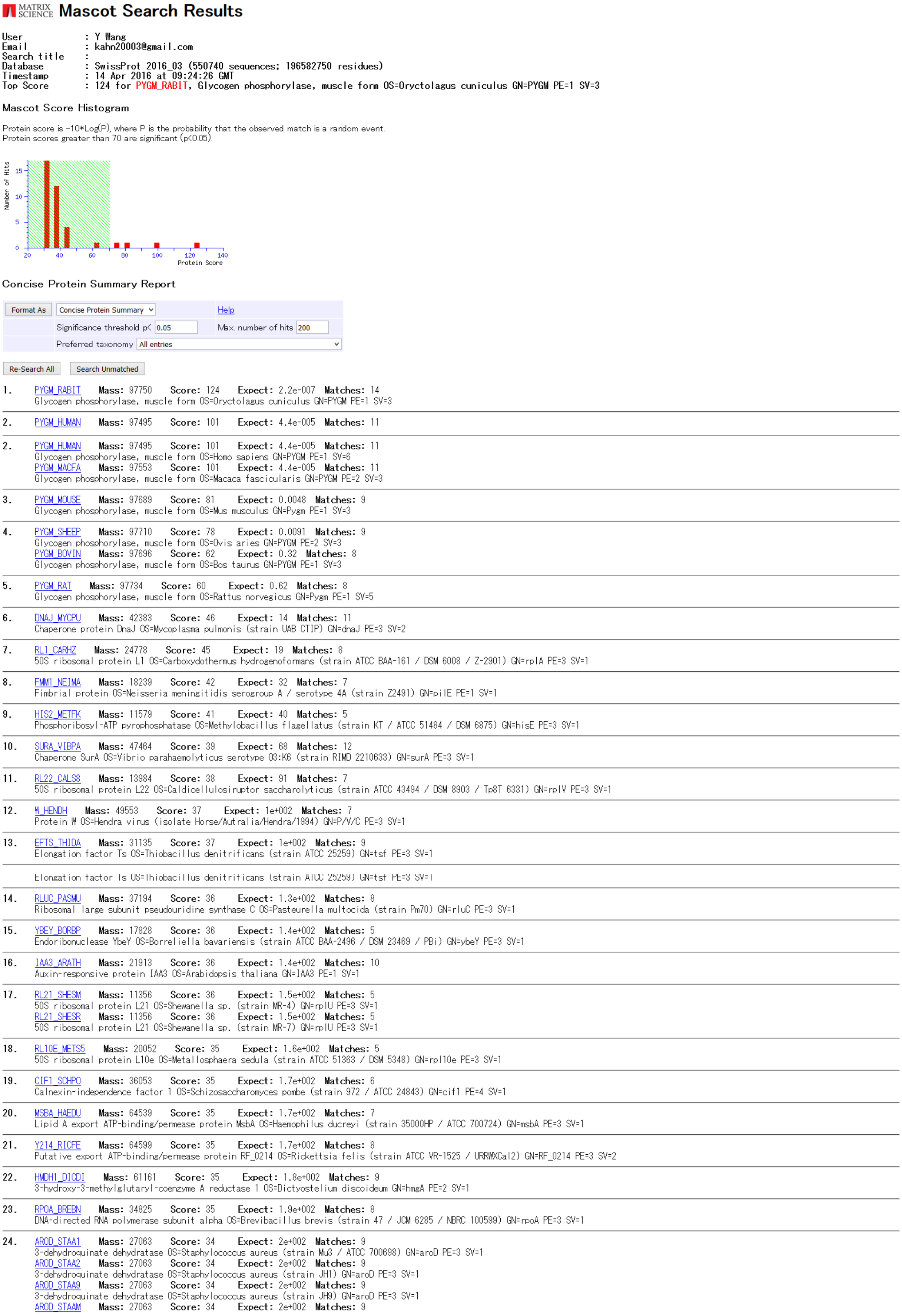
PYGM mass/mass+c



Mascot Search Results

```
User           : Y Wang
Email          : kahn20003@gmail.com
Search title   :
Database       : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp      : 14 Apr 2016 at 09:24:26 GMT
Top Score      : 124 for PYGM_RABIT, Glycogen phosphorylase, muscle form OS=Oryctolagus cuniculus GN=PYGM PE=1 SV=3
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Format As: Concise Protein Summary
Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy: All entries

```
1.    PYGM_RABIT    Mass: 97750    Score: 124    Expect: 2.2e-007  Matches: 14
      Glycogen phosphorylase, muscle form OS=Oryctolagus cuniculus GN=PYGM PE=1 SV=3

2.    PYGM_HUMAN    Mass: 97495    Score: 101    Expect: 4.4e-005  Matches: 11

2.    PYGM_HUMAN    Mass: 97495    Score: 101    Expect: 4.4e-005  Matches: 11
      Glycogen phosphorylase, muscle form OS=Homo sapiens GN=PYGM PE=1 SV=6
      PYGM_MACFA    Mass: 97553    Score: 101    Expect: 4.4e-005  Matches: 11
      Glycogen phosphorylase, muscle form OS=Macaca fascicularis GN=PYGM PE=2 SV=3

3.    PYGM_MOUSE    Mass: 97689    Score: 81     Expect: 0.0048  Matches: 9
      Glycogen phosphorylase, muscle form OS=Mus musculus GN=Pygm PE=1 SV=3

4.    PYGM_SHEEP    Mass: 97710    Score: 78     Expect: 0.0091  Matches: 9
      Glycogen phosphorylase, muscle form OS=Ovis aries GN=PYGM PE=2 SV=3
      PYGM_BOVIN    Mass: 97696    Score: 62     Expect: 0.32  Matches: 8
      Glycogen phosphorylase, muscle form OS=Bos taurus GN=PYGM PE=1 SV=3

5.    PYGM_RAT    Mass: 97734    Score: 60     Expect: 0.62  Matches: 8
      Glycogen phosphorylase, muscle form OS=Rattus norvegicus GN=Pygm PE=1 SV=5

6.    DNAJ_MYCPU    Mass: 42383    Score: 46     Expect: 14  Matches: 11
      Chaperone protein DnaJ OS=Mycoplasma pulmonis (strain UAB CTIP) GN=dnaJ PE=3 SV=2

7.    RL1_CARHZ    Mass: 24778    Score: 45     Expect: 19  Matches: 8
      50S ribosomal protein L1 OS=Carboxydothermus hydrogenoformans (strain ATCC BAA-161 / DSM 6008 / Z-2901) GN=rplA PE=3 SV=1

8.    FMM1_NEIMA    Mass: 18239    Score: 42     Expect: 32  Matches: 7
      Fimbrial protein OS=Neisseria meningitidis serogroup A / serotype 4A (strain Z2491) GN=pilE PE=1 SV=1

9.    HIS2_METFK    Mass: 11579    Score: 41     Expect: 40  Matches: 5
      Phosphoribosyl-ATP pyrophosphatase OS=Methylobacillus flagellatus (strain KT / ATCC 51484 / DSM 6875) GN=hisE PE=3 SV=1

10.   SURA_VIBPA    Mass: 47464    Score: 39     Expect: 68  Matches: 12
      Chaperone SurA OS=Vibrio parahaemolyticus serotype O3:K6 (strain RIMD 2210633) GN=surA PE=3 SV=1

11.   RL22_CALS8    Mass: 13984    Score: 38     Expect: 91  Matches: 7
      50S ribosomal protein L22 OS=Caldicellulosiruptor saccharolyticus (strain ATCC 43494 / DSM 8903 / Tp8T 6331) GN=rplV PE=3 SV=1

12.   W_HENDH    Mass: 49553    Score: 37     Expect: 1e+002  Matches: 7
      Protein W OS=Hendra virus (isolate Horse/Autralia/Hendra/1994) GN=P/V/C PE=3 SV=1

13.   EFTS_THIDA    Mass: 31135    Score: 37     Expect: 1e+002  Matches: 9
      Elongation factor Ts OS=Thiobacillus denitrificans (strain ATCC 25259) GN=tsf PE=3 SV=1
```

Elongation factor Ts OS=Thiobacillus denitrificans (strain ATCC 25259) GN=tsf PE=3 SV=1

```
14.   RLUC_PASMU    Mass: 37194    Score: 36     Expect: 1.3e+002  Matches: 8
      Ribosomal large subunit pseudouridine synthase C OS=Pasteurella multocida (strain Pm70) GN=rluC PE=3 SV=1

15.   YBEY_BORBP    Mass: 17828    Score: 36     Expect: 1.4e+002  Matches: 5
      Endoribonuclease YbeY OS=Borreliella bavariensis (strain ATCC BAA-2496 / DSM 23469 / PBi) GN=ybeY PE=3 SV=1

16.   IAA3_ARATH    Mass: 21913    Score: 36     Expect: 1.4e+002  Matches: 10
      Auxin-responsive protein IAA3 OS=Arabidopsis thaliana GN=IAA3 PE=1 SV=1

17.   RL21_SHESM    Mass: 11356    Score: 36     Expect: 1.5e+002  Matches: 5
      50S ribosomal protein L21 OS=Shewanella sp. (strain MR-4) GN=rplU PE=3 SV=1
      RL21_SHESR    Mass: 11356    Score: 36     Expect: 1.5e+002  Matches: 5
      50S ribosomal protein L21 OS=Shewanella sp. (strain MR-7) GN=rplU PE=3 SV=1

18.   RL10E_METS5    Mass: 20052    Score: 35     Expect: 1.6e+002  Matches: 5
      50S ribosomal protein L10e OS=Metallosphaera sedula (strain ATCC 51363 / DSM 5348) GN=rpl10e PE=3 SV=1

19.   CIF1_SCHPO    Mass: 36053    Score: 35     Expect: 1.7e+002  Matches: 6
      Calnexin-independence factor 1 OS=Schizosaccharomyces pombe (strain 972 / ATCC 24843) GN=cif1 PE=4 SV=1

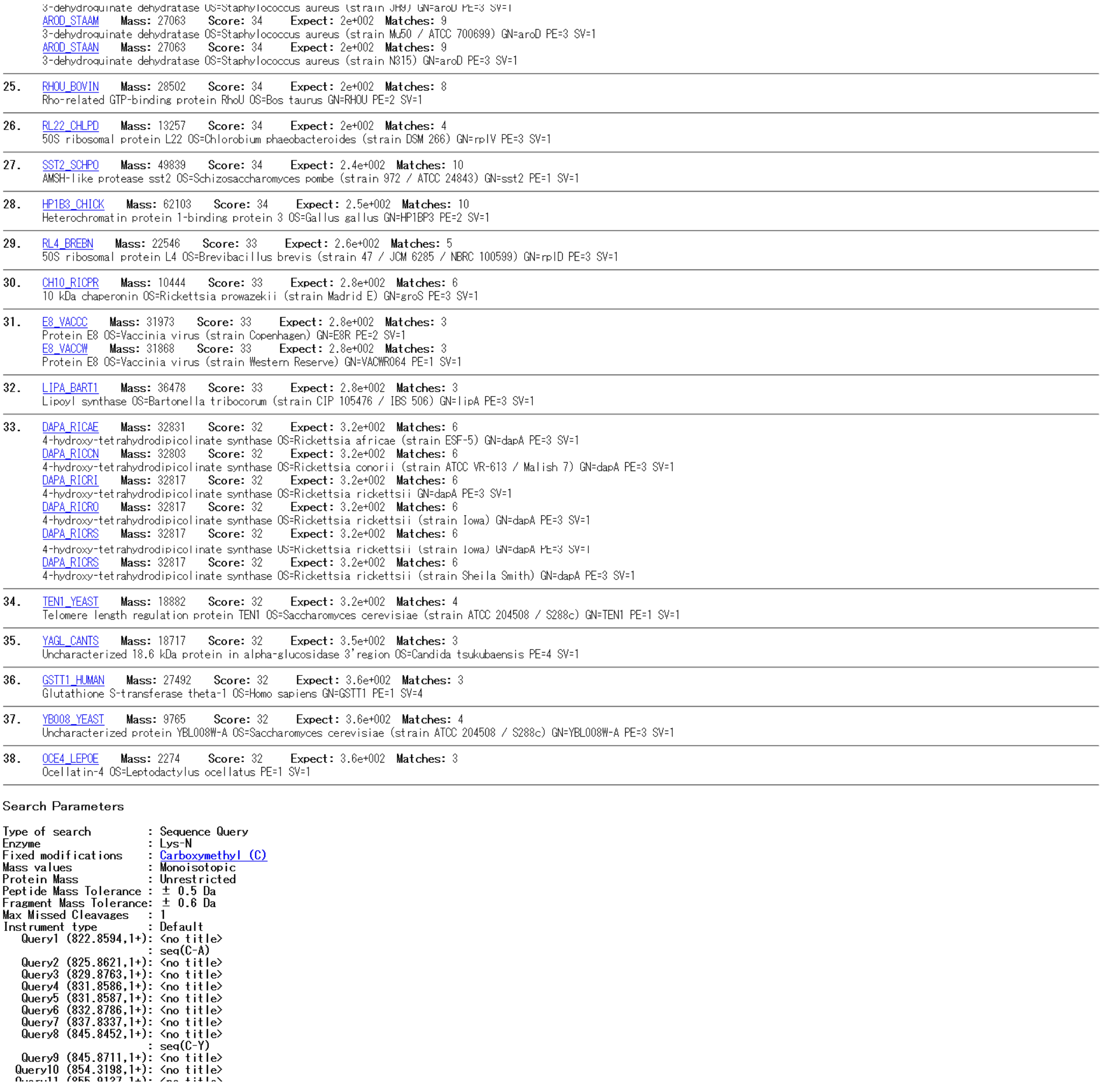
20.   MSBA_HAEDU    Mass: 64539    Score: 35     Expect: 1.7e+002  Matches: 7
      Lipid A export ATP-binding/permease protein MsbA OS=Haemophilus ducreyi (strain 35000HP / ATCC 700724) GN=msbA PE=3 SV=1

21.   Y214_RICFE    Mass: 64599    Score: 35     Expect: 1.7e+002  Matches: 8
      Putative export ATP-binding/permease protein RF_0214 OS=Rickettsia felis (strain ATCC VR-1525 / URRWXCal2) GN=RF_0214 PE=3 SV=2

22.   HMDH1_DICDI    Mass: 61161    Score: 35     Expect: 1.8e+002  Matches: 9
      3-hydroxy-3-methylglutaryl-coenzyme A reductase 1 OS=Dictyostelium discoideum GN=hmgA PE=2 SV=1

23.   RPOA_BREBN    Mass: 34825    Score: 35     Expect: 1.9e+002  Matches: 8
      DNA-directed RNA polymerase subunit alpha OS=Brevibacillus brevis (strain 47 / JCM 6285 / NBRC 100599) GN=rpoA PE=3 SV=1

24.   AROD_STAA1    Mass: 27063    Score: 34     Expect: 2e+002  Matches: 9
      3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain Mu3 / ATCC 700698) GN=aroD PE=3 SV=1
      AROD_STAA2    Mass: 27063    Score: 34     Expect: 2e+002  Matches: 9
      3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain JH1) GN=aroD PE=3 SV=1
      AROD_STAA9    Mass: 27063    Score: 34     Expect: 2e+002  Matches: 9
      3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain JH9) GN=aroD PE=3 SV=1
      AROD_STAAM    Mass: 27063    Score: 34     Expect: 2e+002  Matches: 9
```



```
     3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain JH9) GN=aroD PE=3 SV=1
     AROD_STAAM    Mass: 27063    Score: 34     Expect: 2e+002  Matches: 9
     3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain Mu50 / ATCC 700699) GN=aroD PE=3 SV=1
     AROD_STAAN    Mass: 27063    Score: 34     Expect: 2e+002  Matches: 9
     3-dehydroquinate dehydratase OS=Staphylococcus aureus (strain N315) GN=aroD PE=3 SV=1
----------------------------------------------------------------------------------------------------------
25.  RHOU_BOVIN    Mass: 28502    Score: 34     Expect: 2e+002  Matches: 8
     Rho-related GTP-binding protein RhoU OS=Bos taurus GN=RHOU PE=2 SV=1
----------------------------------------------------------------------------------------------------------
26.  RL22_CHLPD    Mass: 13257    Score: 34     Expect: 2e+002  Matches: 4
     50S ribosomal protein L22 OS=Chlorobium phaeobacteroides (strain DSM 266) GN=rplV PE=3 SV=1
----------------------------------------------------------------------------------------------------------
27.  SST2_SCHPO    Mass: 49839    Score: 34     Expect: 2.4e+002  Matches: 10
     AMSH-like protease sst2 OS=Schizosaccharomyces pombe (strain 972 / ATCC 24843) GN=sst2 PE=1 SV=1
----------------------------------------------------------------------------------------------------------
28.  HP1B3_CHICK    Mass: 62103    Score: 34     Expect: 2.5e+002  Matches: 10
     Heterochromatin protein 1-binding protein 3 OS=Gallus gallus GN=HP1BP3 PE=2 SV=1
----------------------------------------------------------------------------------------------------------
29.  RL4_BREBN    Mass: 22546    Score: 33     Expect: 2.6e+002  Matches: 5
     50S ribosomal protein L4 OS=Brevibacillus brevis (strain 47 / JCM 6285 / NBRC 100599) GN=rplD PE=3 SV=1
----------------------------------------------------------------------------------------------------------
30.  CH10_RICPR    Mass: 10444    Score: 33     Expect: 2.8e+002  Matches: 6
     10 kDa chaperonin OS=Rickettsia prowazekii (strain Madrid E) GN=groS PE=3 SV=1
----------------------------------------------------------------------------------------------------------
31.  E8_VACCC    Mass: 31973    Score: 33     Expect: 2.8e+002  Matches: 3
     Protein E8 OS=Vaccinia virus (strain Copenhagen) GN=E8R PE=2 SV=1
     E8_VACCW    Mass: 31868    Score: 33     Expect: 2.8e+002  Matches: 3
     Protein E8 OS=Vaccinia virus (strain Western Reserve) GN=VACWR064 PE=1 SV=1
----------------------------------------------------------------------------------------------------------
32.  LIPA_BART1    Mass: 36478    Score: 33     Expect: 2.8e+002  Matches: 3
     Lipoyl synthase OS=Bartonella tribocorum (strain CIP 105476 / IBS 506) GN=lipA PE=3 SV=1
----------------------------------------------------------------------------------------------------------
33.  DAPA_RICAE    Mass: 32831    Score: 32     Expect: 3.2e+002  Matches: 6
     4-hydroxy-tetrahydrodipicolinate synthase OS=Rickettsia africae (strain ESF-5) GN=dapA PE=3 SV=1
     DAPA_RICCN    Mass: 32803    Score: 32     Expect: 3.2e+002  Matches: 6
     4-hydroxy-tetrahydrodipicolinate synthase OS=Rickettsia conorii (strain ATCC VR-613 / Malish 7) GN=dapA PE=3 SV=1
     DAPA_RICRI    Mass: 32817    Score: 32     Expect: 3.2e+002  Matches: 6
     4-hydroxy-tetrahydrodipicolinate synthase OS=Rickettsia rickettsii GN=dapA PE=3 SV=1
     DAPA_RICRO    Mass: 32817    Score: 32     Expect: 3.2e+002  Matches: 6
     4-hydroxy-tetrahydrodipicolinate synthase OS=Rickettsia rickettsii (strain Iowa) GN=dapA PE=3 SV=1
     DAPA_RICRS    Mass: 32817    Score: 32     Expect: 3.2e+002  Matches: 6
     4-hydroxy-tetrahydrodipicolinate synthase OS=Rickettsia rickettsii (strain Iowa) GN=dapA PE=3 SV=1
     DAPA_RICRS    Mass: 32817    Score: 32     Expect: 3.2e+002  Matches: 6
     4-hydroxy-tetrahydrodipicolinate synthase OS=Rickettsia rickettsii (strain Sheila Smith) GN=dapA PE=3 SV=1
----------------------------------------------------------------------------------------------------------
34.  TEN1_YEAST    Mass: 18882    Score: 32     Expect: 3.2e+002  Matches: 4
     Telomere length regulation protein TEN1 OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=TEN1 PE=1 SV=1
----------------------------------------------------------------------------------------------------------
35.  YAGL_CANTS    Mass: 18717    Score: 32     Expect: 3.5e+002  Matches: 3
     Uncharacterized 18.6 kDa protein in alpha-glucosidase 3'region OS=Candida tsukubaensis PE=4 SV=1
----------------------------------------------------------------------------------------------------------
36.  GSTT1_HUMAN    Mass: 27492    Score: 32     Expect: 3.6e+002  Matches: 3
     Glutathione S-transferase theta-1 OS=Homo sapiens GN=GSTT1 PE=1 SV=4
----------------------------------------------------------------------------------------------------------
37.  YB008_YEAST    Mass: 9765    Score: 32     Expect: 3.6e+002  Matches: 4
     Uncharacterized protein YBL008W-A OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=YBL008W-A PE=3 SV=1
----------------------------------------------------------------------------------------------------------
38.  OCE4_LEPOE    Mass: 2274    Score: 32     Expect: 3.6e+002  Matches: 3
     Ocellatin-4 OS=Leptodactylus ocellatus PE=1 SV=1
----------------------------------------------------------------------------------------------------------

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (822.8594,1+): <no title>
                       : seq(C-A)
   Query2 (825.8621,1+): <no title>
   Query3 (829.8763,1+): <no title>
   Query4 (831.8586,1+): <no title>
   Query5 (831.8587,1+): <no title>
   Query6 (832.8786,1+): <no title>
   Query7 (837.8337,1+): <no title>
   Query8 (845.8452,1+): <no title>
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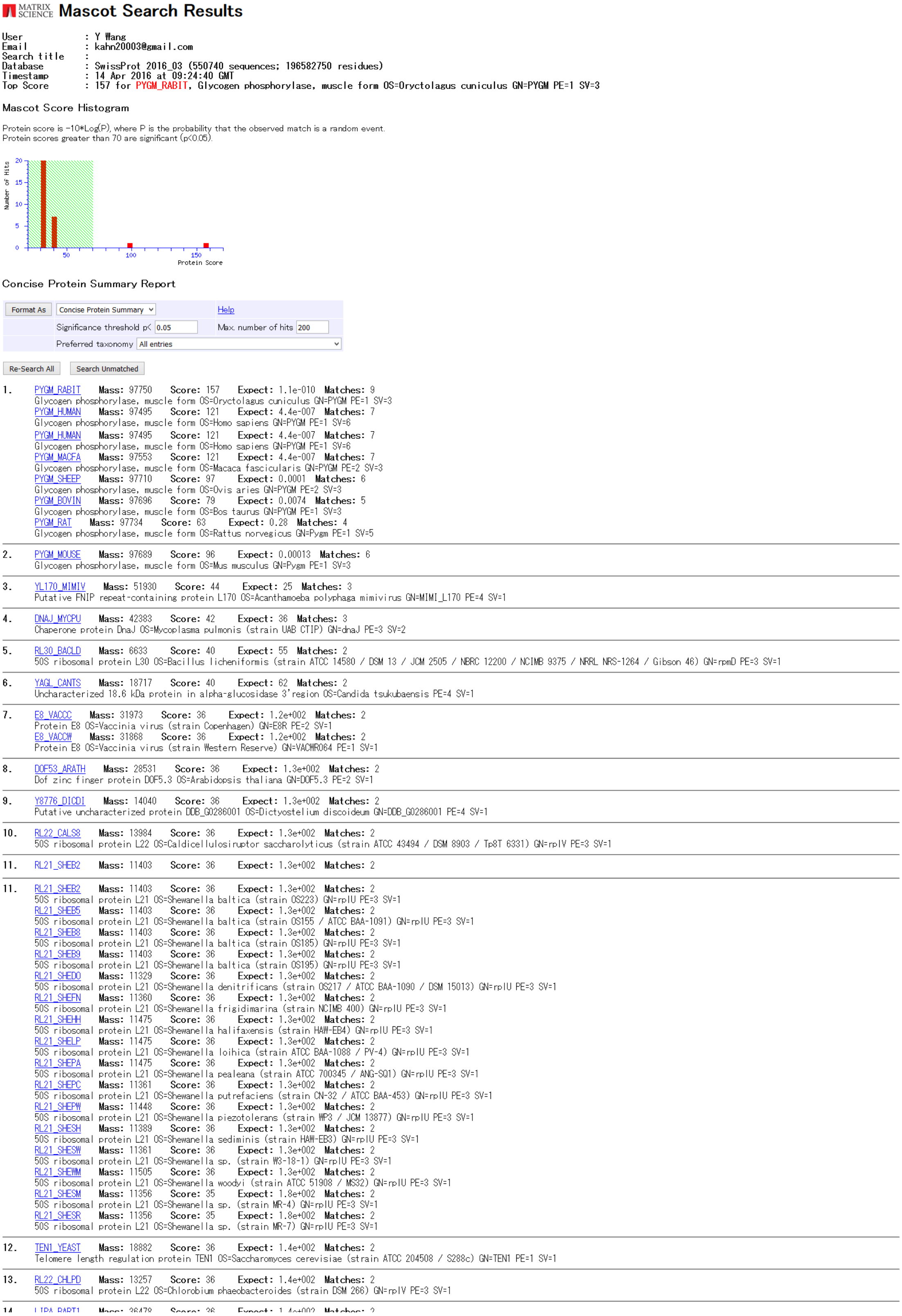
PYGM mass+c



Mascot Search Results

```
User            : Y Wang
Email           : kahn20003@gmail.com
Search title    :
Database        : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp       : 14 Apr 2016 at 09:24:40 GMT
Top Score       : 157 for PYGM_RABIT, Glycogen phosphorylase, muscle form OS=Oryctolagus cuniculus GN=PYGM PE=1 SV=3
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

```
1.    PYGM_RABIT    Mass: 97750    Score: 157    Expect: 1.1e-010  Matches: 9
      Glycogen phosphorylase, muscle form OS=Oryctolagus cuniculus GN=PYGM PE=1 SV=3
      PYGM_HUMAN    Mass: 97495    Score: 121    Expect: 4.4e-007  Matches: 7
      Glycogen phosphorylase, muscle form OS=Homo sapiens GN=PYGM PE=1 SV=6
      PYGM_HUMAN    Mass: 97495    Score: 121    Expect: 4.4e-007  Matches: 7
      Glycogen phosphorylase, muscle form OS=Homo sapiens GN=PYGM PE=1 SV=6
      PYGM_MACFA    Mass: 97553    Score: 121    Expect: 4.4e-007  Matches: 7
      Glycogen phosphorylase, muscle form OS=Macaca fascicularis GN=PYGM PE=2 SV=3
      PYGM_SHEEP    Mass: 97710    Score: 97     Expect: 0.0001  Matches: 6
      Glycogen phosphorylase, muscle form OS=Ovis aries GN=PYGM PE=2 SV=3
      PYGM_BOVIN    Mass: 97696    Score: 79     Expect: 0.0074  Matches: 5
      Glycogen phosphorylase, muscle form OS=Bos taurus GN=PYGM PE=1 SV=3
      PYGM_RAT    Mass: 97734    Score: 63    Expect: 0.28  Matches: 4
      Glycogen phosphorylase, muscle form OS=Rattus norvegicus GN=Pygm PE=1 SV=5

2.    PYGM_MOUSE    Mass: 97689    Score: 96     Expect: 0.00013  Matches: 6
      Glycogen phosphorylase, muscle form OS=Mus musculus GN=Pygm PE=1 SV=3

3.    YL170_MIMIV    Mass: 51930    Score: 44     Expect: 25  Matches: 3
      Putative FNIP repeat-containing protein L170 OS=Acanthamoeba polyphaga mimivirus GN=MIMI_L170 PE=4 SV=1

4.    DNAJ_MYCPU    Mass: 42383    Score: 42     Expect: 36  Matches: 3
      Chaperone protein DnaJ OS=Mycoplasma pulmonis (strain UAB CTIP) GN=dnaJ PE=3 SV=2

5.    RL30_BACLD    Mass: 6633     Score: 40     Expect: 55  Matches: 2
      50S ribosomal protein L30 OS=Bacillus licheniformis (strain ATCC 14580 / DSM 13 / JCM 2505 / NBRC 12200 / NCIMB 9375 / NRRL NRS-1264 / Gibson 46) GN=rpmD PE=3 SV=1

6.    YAGL_CANTS    Mass: 18717    Score: 40     Expect: 62  Matches: 2
      Uncharacterized 18.6 kDa protein in alpha-glucosidase 3'region OS=Candida tsukubaensis PE=4 SV=1

7.    E8_VACCC    Mass: 31973    Score: 36     Expect: 1.2e+002  Matches: 2
      Protein E8 OS=Vaccinia virus (strain Copenhagen) GN=E8R PE=2 SV=1
      E8_VACCW    Mass: 31868    Score: 36     Expect: 1.2e+002  Matches: 2
      Protein E8 OS=Vaccinia virus (strain Western Reserve) GN=VACWR064 PE=1 SV=1

8.    DOF53_ARATH    Mass: 28531    Score: 36     Expect: 1.3e+002  Matches: 2
      Dof zinc finger protein DOF5.3 OS=Arabidopsis thaliana GN=DOF5.3 PE=2 SV=1

9.    Y8776_DICDI    Mass: 14040    Score: 36     Expect: 1.3e+002  Matches: 2
      Putative uncharacterized protein DDB_G0286001 OS=Dictyostelium discoideum GN=DDB_G0286001 PE=4 SV=1

10.   RL22_CALS8    Mass: 13984    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L22 OS=Caldicellulosiruptor saccharolyticus (strain ATCC 43494 / DSM 8903 / Tp8T 6331) GN=rplV PE=3 SV=1

11.   RL21_SHEB2    Mass: 11403    Score: 36     Expect: 1.3e+002  Matches: 2

11.   RL21_SHEB2    Mass: 11403    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella baltica (strain OS223) GN=rplU PE=3 SV=1
      RL21_SHEB5    Mass: 11403    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella baltica (strain OS155 / ATCC BAA-1091) GN=rplU PE=3 SV=1
      RL21_SHEB8    Mass: 11403    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella baltica (strain OS185) GN=rplU PE=3 SV=1
      RL21_SHEB9    Mass: 11403    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella baltica (strain OS195) GN=rplU PE=3 SV=1
      RL21_SHEDO    Mass: 11329    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella denitrificans (strain OS217 / ATCC BAA-1090 / DSM 15013) GN=rplU PE=3 SV=1
      RL21_SHEFN    Mass: 11360    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella frigidimarina (strain NCIMB 400) GN=rplU PE=3 SV=1
      RL21_SHEHH    Mass: 11475    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella halifaxensis (strain HAW-EB4) GN=rplU PE=3 SV=1
      RL21_SHELP    Mass: 11475    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella loihica (strain ATCC BAA-1088 / PV-4) GN=rplU PE=3 SV=1
      RL21_SHEPA    Mass: 11475    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella pealeana (strain ATCC 700345 / ANG-SQ1) GN=rplU PE=3 SV=1
      RL21_SHEPC    Mass: 11361    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella putrefaciens (strain CN-32 / ATCC BAA-453) GN=rplU PE=3 SV=1
      RL21_SHEPW    Mass: 11448    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella piezotolerans (strain WP3 / JCM 13877) GN=rplU PE=3 SV=1
      RL21_SHESH    Mass: 11389    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella sediminis (strain HAW-EB3) GN=rplU PE=3 SV=1
      RL21_SHESW    Mass: 11361    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella sp. (strain W3-18-1) GN=rplU PE=3 SV=1
      RL21_SHEWM    Mass: 11505    Score: 36     Expect: 1.3e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella woodyi (strain ATCC 51908 / MS32) GN=rplU PE=3 SV=1
      RL21_SHESM    Mass: 11356    Score: 35     Expect: 1.8e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella sp. (strain MR-4) GN=rplU PE=3 SV=1
      RL21_SHESR    Mass: 11356    Score: 35     Expect: 1.8e+002  Matches: 2
      50S ribosomal protein L21 OS=Shewanella sp. (strain MR-7) GN=rplU PE=3 SV=1

12.   TEN1_YEAST    Mass: 18882    Score: 36     Expect: 1.4e+002  Matches: 2
      Telomere length regulation protein TEN1 OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=TEN1 PE=1 SV=1

13.   RL22_CHLPD    Mass: 13257    Score: 36     Expect: 1.4e+002  Matches: 2
      50S ribosomal protein L22 OS=Chlorobium phaeobacteroides (strain DSM 266) GN=rplV PE=3 SV=1
```

```
14.  LIPA_BART1    Mass: 36478    Score: 36     Expect: 1.4e+002  Matches: 2
     Lipoyl synthase OS=Bartonella tribocorum (strain CIP 105476 / IBS 506) GN=lipA PE=3 SV=1

15.  GSTT1_HUMAN    Mass: 27492    Score: 36     Expect: 1.5e+002  Matches: 2
     Glutathione S-transferase theta-1 OS=Homo sapiens GN=GSTT1 PE=1 SV=4

16.  RL10E_METS5    Mass: 20052    Score: 36     Expect: 1.6e+002  Matches: 2
     50S ribosomal protein L10e OS=Metallosphaera sedula (strain ATCC 51363 / DSM 5348) GN=rpl10e PE=3 SV=1

17.  UPPP2_PSEHT    Mass: 28077    Score: 35     Expect: 1.7e+002  Matches: 2
     Undecaprenyl-diphosphatase 2 OS=Pseudoalteromonas haloplanktis (strain TAC 125) GN=uppP2 PE=3 SV=1

18.  NIP7_DICDI    Mass: 20975    Score: 35     Expect: 1.9e+002  Matches: 2
     60S ribosome subunit biogenesis protein NIP7 homolog OS=Dictyostelium discoideum GN=nip7 PE=3 SV=1

19.  SYS_CHLFF    Mass: 48647    Score: 34     Expect: 2.2e+002  Matches: 2
     Serine--tRNA ligase OS=Chlamydophila felis (strain Fe/C-56) GN=serS PE=3 SV=1

20.  FOLD1_SYNWW    Mass: 29876    Score: 34     Expect: 2.3e+002  Matches: 2
     Bifunctional protein FolD 1 OS=Syntrophomonas wolfei subsp. wolfei (strain DSM 2245B / Goettingen) GN=folD1 PE=3 SV=1

21.  FAT3_HUMAN    Mass: 508747   Score: 33     Expect: 2.6e+002  Matches: 4
     Protocadherin Fat 3 OS=Homo sapiens GN=FAT3 PE=2 SV=2

22.  SUCC_ACICJ    Mass: 42214    Score: 33     Expect: 2.6e+002  Matches: 2
     Succinyl-CoA ligase [ADP-forming] subunit beta OS=Acidiphilium cryptum (strain JF-5) GN=sucC PE=3 SV=1

23.  L_NIPAV    Mass: 259389   Score: 33     Expect: 2.8e+002  Matches: 3
     RNA-directed RNA polymerase L OS=Nipah virus GN=L PE=3 SV=1

24.  HIS4_METPP    Mass: 26322    Score: 33     Expect: 2.9e+002  Matches: 2
     1-(5-phosphoribosyl)-5-[(5-phosphoribosylamino)methylideneamino] imidazole-4-carboxamide isomerase OS=Methylibium petroleiphilum (strain PM1) GN=hisA PE=3 SV=1

25.  YB008_YEAST    Mass: 9765     Score: 33     Expect: 3e+002  Matches: 2
     Uncharacterized protein YBL008W-A OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=YBL008W-A PE=3 SV=1

26.  LPXK_PSEA6    Mass: 34416    Score: 32     Expect: 3.2e+002  Matches: 2
     Tetraacyldisaccharide 4'-kinase OS=Pseudoalteromonas atlantica (strain T6c / ATCC BAA-1087) GN=lpxK PE=3 SV=1
```

27   TMX2A_DANRE   Mass: 35671   Score: 32   Expect: 3.3e+002 Matches: 2

```
27.  TMX2A_DANRE    Mass: 35671    Score: 32     Expect: 3.3e+002  Matches: 2
     Thioredoxin-related transmembrane protein 2-A OS=Danio rerio GN=tmx2a PE=2 SV=1

28.  W_HENDH    Mass: 49553    Score: 32     Expect: 3.3e+002  Matches: 2
     Protein W OS=Hendra virus (isolate Horse/Autralia/Hendra/1994) GN=P/V/C PE=3 SV=1

29.  POF7_SCHPO    Mass: 42428    Score: 32     Expect: 3.4e+002  Matches: 2
     F-box protein pof7 OS=Schizosaccharomyces pombe (strain 972 / ATCC 24843) GN=pof7 PE=1 SV=2
```

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (822.8594,1+): <no title>
                       : seq(C-A)
   Query2 (845.8452,1+): <no title>
                       : seq(C-Y)
   Query3 (855.9137,1+): <no title>
                       : seq(C-I)
   Query4 (855.9137,1+): <no title>
                       : seq(C-L)
   Query5 (867.8736,1+): <no title>
                       : seq(C-I)
   Query6 (867.8736,1+): <no title>
                       : seq(C-L)
   Query7 (872.8096,1+): <no title>
                       : seq(C-Y)
   Query8 (900.8648,1+): <no title>
                       : seq(C-P)
   Query9 (918.8184,1+): <no title>
                       : seq(C-I)
  Query10 (918.8184,1+): <no title>
                       : seq(C-L)
  Query11 (923.8497,1+): <no title>
                       : seq(C-I)
```

CA mass

Mascot Search Results

User : Y Wang
Email : kahn20003@gmail.com
Search title :
Database : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp : 14 Apr 2016 at 09:50:30 GMT
Top Score : 48 for CAH2_BOVIN, Carbonic anhydrase 2 OS=Bos taurus GN=CA2 PE=1 SV=3

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

1. CAH2_BOVIN **Mass:** 29096 **Score:** 48 **Expect:** 8.5 **Matches:** 11
Carbonic anhydrase 2 OS=Bos taurus GN=CA2 PE=1 SV=3

2. RS16_SHEFN **Mass:** 9318 **Score:** 38 **Expect:** 78 **Matches:** 5

2. RS16_SHEFN **Mass:** 9318 **Score:** 38 **Expect:** 78 **Matches:** 5
30S ribosomal protein S16 OS=Shewanella frigidimarina (strain NCIMB 400) GN=rpsP PE=3 SV=1

3. METN2_STAAB **Mass:** 38372 **Score:** 38 **Expect:** 98 **Matches:** 9
Methionine import ATP-binding protein MetN 2 OS=Staphylococcus aureus (strain bovine RF122 / ET3-1) GN=metN2 PE=3 SV=1
METN2_STAAM **Mass:** 38351 **Score:** 38 **Expect:** 98 **Matches:** 9
Methionine import ATP-binding protein MetN 2 OS=Staphylococcus aureus (strain Mu50 / ATCC 700699) GN=metN2 PE=1 SV=1
METN2_STAAN **Mass:** 38351 **Score:** 38 **Expect:** 98 **Matches:** 9
Methionine import ATP-binding protein MetN 2 OS=Staphylococcus aureus (strain N315) GN=metN2 PE=1 SV=1

4. AP4S1_HUMAN **Mass:** 17168 **Score:** 37 **Expect:** 1.1e+002 **Matches:** 8
AP-4 complex subunit sigma-1 OS=Homo sapiens GN=AP4S1 PE=2 SV=1

5. AP4S1_MOUSE **Mass:** 16981 **Score:** 37 **Expect:** 1.1e+002 **Matches:** 8
AP-4 complex subunit sigma-1 OS=Mus musculus GN=Ap4s1 PE=2 SV=1

6. COBD_PYRFU **Mass:** 32432 **Score:** 37 **Expect:** 1.2e+002 **Matches:** 7
Probable cobalamin biosynthesis protein CobD OS=Pyrococcus furiosus (strain ATCC 43587 / DSM 3638 / JCM 8422 / Vc1) GN=cobD PE=3 SV=1

7. BIOD_CLOBB **Mass:** 25775 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 8
ATP-dependent dethiobiotin synthetase BioD OS=Clostridium botulinum (strain Eklund 17B / Type B) GN=bioD PE=3 SV=1

8. LEUD_CYTH3 **Mass:** 22298 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 8
3-isopropylmalate dehydratase small subunit OS=Cytophaga hutchinsonii (strain ATCC 33406 / NCIMB 9469) GN=leuD PE=3 SV=1

9. RAN_SALSA **Mass:** 24532 **Score:** 36 **Expect:** 1.6e+002 **Matches:** 9
GTP-binding nuclear protein Ran OS=Salmo salar GN=ran PE=2 SV=1

10. SLAP1_BACAN **Mass:** 86568 **Score:** 35 **Expect:** 1.7e+002 **Matches:** 20
S-layer protein sap OS=Bacillus anthracis GN=sap PE=1 SV=1

11. RS17_STRCO **Mass:** 10714 **Score:** 35 **Expect:** 1.7e+002 **Matches:** 9
30S ribosomal protein S17 OS=Streptomyces coelicolor (strain ATCC BAA-471 / A3(2) / M145) GN=rpsQ PE=3 SV=1

12. DST2_DICDI **Mass:** 131280 **Score:** 35 **Expect:** 1.8e+002 **Matches:** 19
Serine/threonine-protein kinase dst2 OS=Dictyostelium discoideum GN=dst2 PE=3 SV=1

13. Y723_THEEB **Mass:** 12245 **Score:** 34 **Expect:** 2e+002 **Matches:** 7
Nucleoid-associated protein tlr0723 OS=Thermosynechococcus elongatus (strain BP-1) GN=tlr0723 PE=3 SV=1

Nucleoid-associated protein tlr0723 OS=Thermosynechococcus elongatus (strain BP-1) GN=tlr0723 PE=3 SV=1

14. PLSX_PEPD6 **Mass:** 37029 **Score:** 34 **Expect:** 2.3e+002 **Matches:** 12
Phosphate acyltransferase OS=Peptoclostridium difficile (strain 630) GN=plsX PE=3 SV=1

15. ATPF_VIBC3 **Mass:** 17707 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 9
ATP synthase subunit b OS=Vibrio cholerae serotype O1 (strain ATCC 39541 / Classical Ogawa 395 / O395) GN=atpF PE=3 SV=1
ATPF_VIBCH **Mass:** 17707 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 9
ATP synthase subunit b OS=Vibrio cholerae serotype O1 (strain ATCC 39315 / El Tor Inaba N16961) GN=atpF PE=3 SV=1

16. RL18_PYRIL **Mass:** 23217 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 10
50S ribosomal protein L18 OS=Pyrobaculum islandicum (strain DSM 4184 / JCM 9189 / GEO3) GN=rpl18 PE=3 SV=1

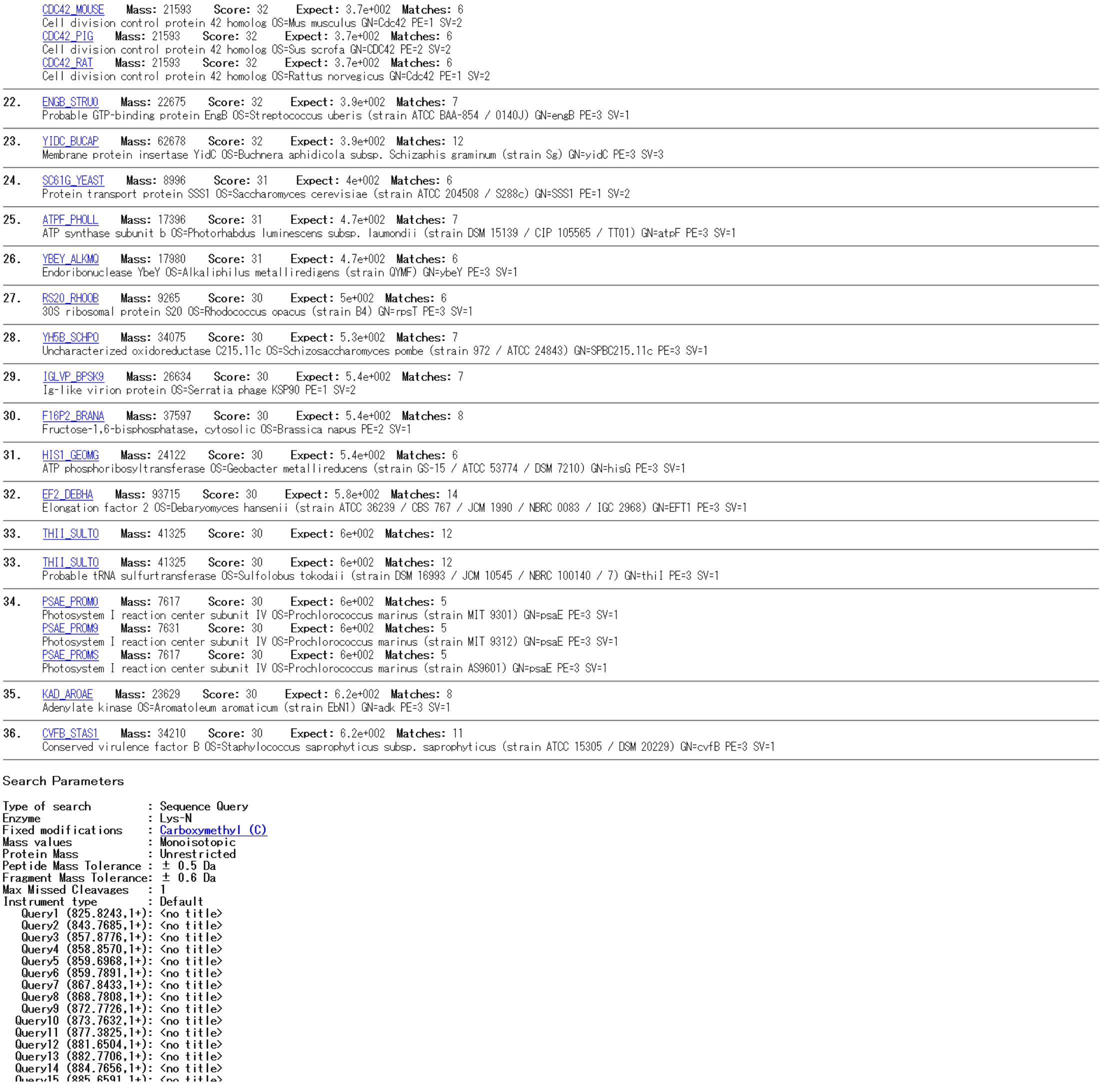
17. CAH2_SHEEP **Mass:** 29193 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 7
Carbonic anhydrase 2 OS=Ovis aries GN=CA2 PE=1 SV=2

18. OCAD2_HUMAN **Mass:** 17291 **Score:** 33 **Expect:** 2.9e+002 **Matches:** 7
OCIA domain-containing protein 2 OS=Homo sapiens GN=OCIAD2 PE=1 SV=1

19. RL15_RICAE **Mass:** 16738 **Score:** 32 **Expect:** 3.5e+002 **Matches:** 10
50S ribosomal protein L15 OS=Rickettsia africae (strain ESF-5) GN=rplO PE=3 SV=1
RL15_RICCN **Mass:** 16738 **Score:** 32 **Expect:** 3.5e+002 **Matches:** 10
50S ribosomal protein L15 OS=Rickettsia conorii (strain ATCC VR-613 / Malish 7) GN=rplO PE=3 SV=1
RL15_RICPU **Mass:** 16742 **Score:** 32 **Expect:** 3.5e+002 **Matches:** 10
50S ribosomal protein L15 OS=Rickettsia peacockii (strain Rustic) GN=rplO PE=3 SV=1

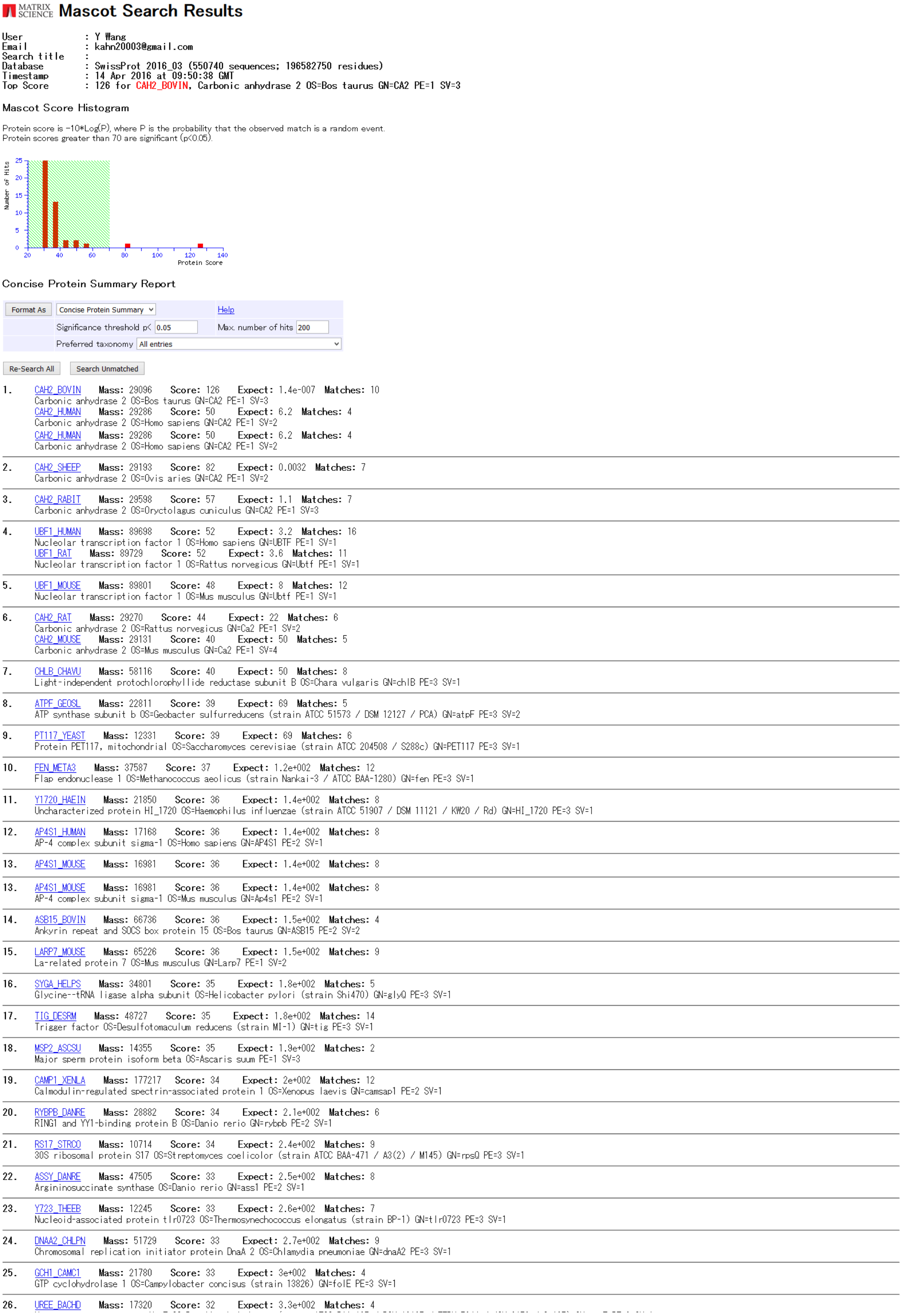
20. RNA14_CANAL **Mass:** 91733 **Score:** 32 **Expect:** 3.6e+002 **Matches:** 20
mRNA 3'-end-processing protein RNA14 OS=Candida albicans (strain SC5314 / ATCC MYA-2876) GN=RNA14 PE=3 SV=1

21. CDC42_BOVIN **Mass:** 21593 **Score:** 32 **Expect:** 3.7e+002 **Matches:** 6
Cell division control protein 42 homolog OS=Bos taurus GN=CDC42 PE=1 SV=1
CDC42_CANLF **Mass:** 21593 **Score:** 32 **Expect:** 3.7e+002 **Matches:** 6
Cell division control protein 42 homolog OS=Canis lupus familiaris GN=CDC42 PE=2 SV=2
CDC42_CHICK **Mass:** 21607 **Score:** 32 **Expect:** 3.7e+002 **Matches:** 6
Cell division control protein 42 homolog OS=Gallus gallus GN=CDC42 PE=2 SV=1
CDC42_HUMAN **Mass:** 21593 **Score:** 32 **Expect:** 3.7e+002 **Matches:** 6
Cell division control protein 42 homolog OS=Homo sapiens GN=CDC42 PE=1 SV=2
CDC42_MACFA **Mass:** 21592 **Score:** 32 **Expect:** 3.7e+002 **Matches:** 6
Cell division control protein 42 homolog OS=Macaca fascicularis GN=CDC42 PE=2 SV=1
CDC42_MOUSE **Mass:** 21593 **Score:** 32 **Expect:** 3.7e+002 **Matches:** 6
Cell division control protein 42 homolog OS=Mus musculus GN=Cdc42 PE=1 SV=2



```
         CDC42_MOUSE    Mass: 21593    Score: 32     Expect: 3.7e+002  Matches: 6
         Cell division control protein 42 homolog OS=Mus musculus GN=Cdc42 PE=1 SV=2
         CDC42_PIG    Mass: 21593    Score: 32     Expect: 3.7e+002  Matches: 6
         Cell division control protein 42 homolog OS=Sus scrofa GN=CDC42 PE=2 SV=2
         CDC42_RAT    Mass: 21593    Score: 32     Expect: 3.7e+002  Matches: 6
         Cell division control protein 42 homolog OS=Rattus norvegicus GN=Cdc42 PE=1 SV=2

22.      ENGB_STRU0    Mass: 22675    Score: 32     Expect: 3.9e+002  Matches: 7
         Probable GTP-binding protein EngB OS=Streptococcus uberis (strain ATCC BAA-854 / 0140J) GN=engB PE=3 SV=1

23.      YIDC_BUCAP    Mass: 62678    Score: 32     Expect: 3.9e+002  Matches: 12
         Membrane protein insertase YidC OS=Buchnera aphidicola subsp. Schizaphis graminum (strain Sg) GN=yidC PE=3 SV=3

24.      SC61G_YEAST    Mass: 8996    Score: 31     Expect: 4e+002  Matches: 6
         Protein transport protein SSS1 OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=SSS1 PE=1 SV=2

25.      ATPF_PHOLL    Mass: 17396    Score: 31     Expect: 4.7e+002  Matches: 7
         ATP synthase subunit b OS=Photorhabdus luminescens subsp. laumondii (strain DSM 15139 / CIP 105565 / TT01) GN=atpF PE=3 SV=1

26.      YBEY_ALKMQ    Mass: 17980    Score: 31     Expect: 4.7e+002  Matches: 6
         Endoribonuclease YbeY OS=Alkaliphilus metalliredigens (strain QYMF) GN=ybeY PE=3 SV=1

27.      RS20_RHOOB    Mass: 9265    Score: 30     Expect: 5e+002  Matches: 6
         30S ribosomal protein S20 OS=Rhodococcus opacus (strain B4) GN=rpsT PE=3 SV=1

28.      YH5B_SCHPO    Mass: 34075    Score: 30     Expect: 5.3e+002  Matches: 7
         Uncharacterized oxidoreductase C215.11c OS=Schizosaccharomyces pombe (strain 972 / ATCC 24843) GN=SPBC215.11c PE=3 SV=1

29.      IGLVP_BPSK9    Mass: 26634    Score: 30     Expect: 5.4e+002  Matches: 7
         Ig-like virion protein OS=Serratia phage KSP90 PE=1 SV=2

30.      F16P2_BRANA    Mass: 37597    Score: 30     Expect: 5.4e+002  Matches: 8
         Fructose-1,6-bisphosphatase, cytosolic OS=Brassica napus PE=2 SV=1

31.      HIS1_GEOMG    Mass: 24122    Score: 30     Expect: 5.4e+002  Matches: 6
         ATP phosphoribosyltransferase OS=Geobacter metallireducens (strain GS-15 / ATCC 53774 / DSM 7210) GN=hisG PE=3 SV=1

32.      EF2_DEBHA    Mass: 93715    Score: 30     Expect: 5.8e+002  Matches: 14
         Elongation factor 2 OS=Debaryomyces hansenii (strain ATCC 36239 / CBS 767 / JCM 1990 / NBRC 0083 / IGC 2968) GN=EFT1 PE=3 SV=1

33.      THII_SULTO    Mass: 41325    Score: 30     Expect: 6e+002  Matches: 12

33.      THII_SULTO    Mass: 41325    Score: 30     Expect: 6e+002  Matches: 12
         Probable tRNA sulfurtransferase OS=Sulfolobus tokodaii (strain DSM 16993 / JCM 10545 / NBRC 100140 / 7) GN=thiI PE=3 SV=1

34.      PSAE_PROM0    Mass: 7617    Score: 30     Expect: 6e+002  Matches: 5
         Photosystem I reaction center subunit IV OS=Prochlorococcus marinus (strain MIT 9301) GN=psaE PE=3 SV=1
         PSAE_PROM9    Mass: 7631    Score: 30     Expect: 6e+002  Matches: 5
         Photosystem I reaction center subunit IV OS=Prochlorococcus marinus (strain MIT 9312) GN=psaE PE=3 SV=1
         PSAE_PROMS    Mass: 7617    Score: 30     Expect: 6e+002  Matches: 5
         Photosystem I reaction center subunit IV OS=Prochlorococcus marinus (strain AS9601) GN=psaE PE=3 SV=1

35.      KAD_AROAE    Mass: 23629    Score: 30     Expect: 6.2e+002  Matches: 8
         Adenylate kinase OS=Aromatoleum aromaticum (strain EbN1) GN=adk PE=3 SV=1

36.      CVFB_STAS1    Mass: 34210    Score: 30     Expect: 6.2e+002  Matches: 11
         Conserved virulence factor B OS=Staphylococcus saprophyticus subsp. saprophyticus (strain ATCC 15305 / DSM 20229) GN=cvfB PE=3 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (825.8243,1+): <no title>
   Query2 (843.7685,1+): <no title>
   Query3 (857.8776,1+): <no title>
   Query4 (858.8570,1+): <no title>
   Query5 (859.6968,1+): <no title>
   Query6 (859.7891,1+): <no title>
   Query7 (867.8433,1+): <no title>
   Query8 (868.7808,1+): <no title>
   Query9 (872.7726,1+): <no title>
  Query10 (873.7632,1+): <no title>
  Query11 (877.3825,1+): <no title>
  Query12 (881.6504,1+): <no title>
  Query13 (882.7706,1+): <no title>
  Query14 (884.7656,1+): <no title>
  Query15 (885.6591,1+): <no title>
```

CA mass/mass+c



Mascot Search Results

User : Y Wang
Email : kahn20003@gmail.com
Search title :
Database : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp : 14 Apr 2016 at 09:50:38 GMT
Top Score : 126 for CAH2_BOVIN, Carbonic anhydrase 2 OS=Bos taurus GN=CA2 PE=1 SV=3

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

1\. CAH2_BOVIN **Mass:** 29096 **Score:** 126 **Expect:** 1.4e-007 **Matches:** 10
Carbonic anhydrase 2 OS=Bos taurus GN=CA2 PE=1 SV=3
CAH2_HUMAN **Mass:** 29286 **Score:** 50 **Expect:** 6.2 **Matches:** 4
Carbonic anhydrase 2 OS=Homo sapiens GN=CA2 PE=1 SV=2
CAH2_HUMAN **Mass:** 29286 **Score:** 50 **Expect:** 6.2 **Matches:** 4
Carbonic anhydrase 2 OS=Homo sapiens GN=CA2 PE=1 SV=2

2\. CAH2_SHEEP **Mass:** 29193 **Score:** 82 **Expect:** 0.0032 **Matches:** 7
Carbonic anhydrase 2 OS=Ovis aries GN=CA2 PE=1 SV=2

3\. CAH2_RABIT **Mass:** 29598 **Score:** 57 **Expect:** 1.1 **Matches:** 7
Carbonic anhydrase 2 OS=Oryctolagus cuniculus GN=CA2 PE=1 SV=3

4\. UBF1_HUMAN **Mass:** 89698 **Score:** 52 **Expect:** 3.2 **Matches:** 16
Nucleolar transcription factor 1 OS=Homo sapiens GN=UBTF PE=1 SV=1
UBF1_RAT **Mass:** 89729 **Score:** 52 **Expect:** 3.6 **Matches:** 11
Nucleolar transcription factor 1 OS=Rattus norvegicus GN=Ubtf PE=1 SV=1

5\. UBF1_MOUSE **Mass:** 89801 **Score:** 48 **Expect:** 8 **Matches:** 12
Nucleolar transcription factor 1 OS=Mus musculus GN=Ubtf PE=1 SV=1

6\. CAH2_RAT **Mass:** 29270 **Score:** 44 **Expect:** 22 **Matches:** 6
Carbonic anhydrase 2 OS=Rattus norvegicus GN=Ca2 PE=1 SV=2
CAH2_MOUSE **Mass:** 29131 **Score:** 40 **Expect:** 50 **Matches:** 5
Carbonic anhydrase 2 OS=Mus musculus GN=Ca2 PE=1 SV=4

7\. CHLB_CHAVU **Mass:** 58116 **Score:** 40 **Expect:** 50 **Matches:** 8
Light-independent protochlorophyllide reductase subunit B OS=Chara vulgaris GN=chlB PE=3 SV=1

8\. ATPF_GEOSL **Mass:** 22811 **Score:** 39 **Expect:** 69 **Matches:** 5
ATP synthase subunit b OS=Geobacter sulfurreducens (strain ATCC 51573 / DSM 12127 / PCA) GN=atpF PE=3 SV=2

9\. PT117_YEAST **Mass:** 12331 **Score:** 39 **Expect:** 69 **Matches:** 6
Protein PET117, mitochondrial OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=PET117 PE=3 SV=1

10\. FEN_META3 **Mass:** 37587 **Score:** 37 **Expect:** 1.2e+002 **Matches:** 12
Flap endonuclease 1 OS=Methanococcus aeolicus (strain Nankai-3 / ATCC BAA-1280) GN=fen PE=3 SV=1

11\. Y1720_HAEIN **Mass:** 21850 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 8
Uncharacterized protein HI_1720 OS=Haemophilus influenzae (strain ATCC 51907 / DSM 11121 / KW20 / Rd) GN=HI_1720 PE=3 SV=1

12\. AP4S1_HUMAN **Mass:** 17168 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 8
AP-4 complex subunit sigma-1 OS=Homo sapiens GN=AP4S1 PE=2 SV=1

13\. AP4S1_MOUSE **Mass:** 16981 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 8

13\. AP4S1_MOUSE **Mass:** 16981 **Score:** 36 **Expect:** 1.4e+002 **Matches:** 8
AP-4 complex subunit sigma-1 OS=Mus musculus GN=Ap4s1 PE=2 SV=1

14\. ASB15_BOVIN **Mass:** 66736 **Score:** 36 **Expect:** 1.5e+002 **Matches:** 4
Ankyrin repeat and SOCS box protein 15 OS=Bos taurus GN=ASB15 PE=2 SV=2

15\. LARP7_MOUSE **Mass:** 65226 **Score:** 36 **Expect:** 1.5e+002 **Matches:** 9
La-related protein 7 OS=Mus musculus GN=Larp7 PE=1 SV=2

16\. SYGA_HELPS **Mass:** 34801 **Score:** 35 **Expect:** 1.8e+002 **Matches:** 5
Glycine--tRNA ligase alpha subunit OS=Helicobacter pylori (strain Shi470) GN=glyQ PE=3 SV=1

17\. TIG_DESRM **Mass:** 48727 **Score:** 35 **Expect:** 1.8e+002 **Matches:** 14
Trigger factor OS=Desulfotomaculum reducens (strain MI-1) GN=tig PE=3 SV=1

18\. MSP2_ASCSU **Mass:** 14355 **Score:** 35 **Expect:** 1.9e+002 **Matches:** 2
Major sperm protein isoform beta OS=Ascaris suum PE=1 SV=3

19\. CAMP1_XENLA **Mass:** 177217 **Score:** 34 **Expect:** 2e+002 **Matches:** 12
Calmodulin-regulated spectrin-associated protein 1 OS=Xenopus laevis GN=camsap1 PE=2 SV=1

20\. RYBPB_DANRE **Mass:** 28882 **Score:** 34 **Expect:** 2.1e+002 **Matches:** 6
RING1 and YY1-binding protein B OS=Danio rerio GN=rybpb PE=2 SV=1

21\. RS17_STRCO **Mass:** 10714 **Score:** 34 **Expect:** 2.4e+002 **Matches:** 9
30S ribosomal protein S17 OS=Streptomyces coelicolor (strain ATCC BAA-471 / A3(2) / M145) GN=rpsQ PE=3 SV=1

22\. ASSY_DANRE **Mass:** 47505 **Score:** 33 **Expect:** 2.5e+002 **Matches:** 8
Argininosuccinate synthase OS=Danio rerio GN=ass1 PE=2 SV=1

23\. Y723_THEEB **Mass:** 12245 **Score:** 33 **Expect:** 2.6e+002 **Matches:** 7
Nucleoid-associated protein tlr0723 OS=Thermosynechococcus elongatus (strain BP-1) GN=tlr0723 PE=3 SV=1

24\. DNAA2_CHLPN **Mass:** 51729 **Score:** 33 **Expect:** 2.7e+002 **Matches:** 9
Chromosomal replication initiator protein DnaA 2 OS=Chlamydia pneumoniae GN=dnaA2 PE=3 SV=1

25\. GCH1_CAMC1 **Mass:** 21780 **Score:** 33 **Expect:** 3e+002 **Matches:** 4
GTP cyclohydrolase 1 OS=Campylobacter concisus (strain 13826) GN=folE PE=3 SV=1

26\. UREE_BACHD **Mass:** 17320 **Score:** 32 **Expect:** 3.3e+002 **Matches:** 4

```
26.  UREE_BACHD    Mass: 17320    Score: 32     Expect: 3.3e+002  Matches: 4
     Urease accessory protein UreE OS=Bacillus halodurans (strain ATCC BAA-125 / DSM 18197 / FERM 7344 / JCM 9153 / C-125) GN=ureE PE=3 SV=1

27.  EXO1_BPT4    Mass: 39315    Score: 32     Expect: 3.3e+002  Matches: 5
     Exonuclease subunit 1 OS=Enterobacteria phage T4 GN=47 PE=1 SV=1

28.  CSN5_DEBHA    Mass: 53267    Score: 32     Expect: 3.5e+002  Matches: 5
     COP9 signalosome complex subunit 5 OS=Debaryomyces hansenii (strain ATCC 36239 / CBS 767 / JCM 1990 / NBRC 0083 / IGC 2968) GN=RRI1 PE=3 SV=2

29.  MURA_BUCBP    Mass: 45326    Score: 32     Expect: 3.6e+002  Matches: 9
     UDP-N-acetylglucosamine 1-carboxyvinyltransferase OS=Buchnera aphidicola subsp. Baizongia pistaciae (strain Bp) GN=murA PE=3 SV=1

30.  PURA_SYNFM    Mass: 47713    Score: 32     Expect: 3.6e+002  Matches: 3
     Adenylosuccinate synthetase OS=Syntrophobacter fumaroxidans (strain DSM 10017 / MPOB) GN=purA PE=3 SV=1

31.  MINE_ALCBS    Mass: 9450     Score: 32     Expect: 3.6e+002  Matches: 3
     Cell division topological specificity factor OS=Alcanivorax borkumensis (strain ATCC 700651 / DSM 11573 / NCIMB 13689 / SK2) GN=minE PE=3 SV=1

32.  SYGA_HELPG    Mass: 34711    Score: 32     Expect: 3.6e+002  Matches: 3
     Glycine--tRNA ligase alpha subunit OS=Helicobacter pylori (strain G27) GN=glyQ PE=3 SV=1

33.  RPC1_DEBHA    Mass: 163618   Score: 32     Expect: 3.8e+002  Matches: 12
     DNA-directed RNA polymerase III subunit RPC1 OS=Debaryomyces hansenii (strain ATCC 36239 / CBS 767 / JCM 1990 / NBRC 0083 / IGC 2968) GN=RPC1 PE=3 SV=2

34.  RS15_THEAC    Mass: 16504    Score: 32     Expect: 3.9e+002  Matches: 6
     30S ribosomal protein S15 OS=Thermoplasma acidophilum (strain ATCC 25905 / DSM 1728 / JCM 9062 / NBRC 15155 / AMRC-C165) GN=rps15 PE=3 SV=1

35.  RL21_BAUCH    Mass: 12172    Score: 31     Expect: 4e+002  Matches: 5
     50S ribosomal protein L21 OS=Baumannia cicadellinicola subsp. Homalodisca coagulata GN=rplU PE=3 SV=1

36.  UL24_ELHVK    Mass: 26729    Score: 31     Expect: 4.1e+002  Matches: 6
     Protein UL24 homolog OS=Elephantid herpesvirus 1 (isolate Asian elephant/Berlin/Kiba/1998) PE=2 SV=1

37.  P2C76_ARATH    Mass: 46106    Score: 31     Expect: 4.2e+002  Matches: 7
     Probable protein phosphatase 2C 76 OS=Arabidopsis thaliana GN=At5g53140 PE=2 SV=1

38.  LIPA_RHOPS    Mass: 35521    Score: 31     Expect: 4.2e+002  Matches: 3
     Lipoyl synthase OS=Rhodopseudomonas palustris (strain BisB5) GN=lipA PE=3 SV=1

39.  PDAD_META3    Mass: 17860    Score: 31     Expect: 4.4e+002  Matches: 4

39.  PDAD_META3    Mass: 17860    Score: 31     Expect: 4.4e+002  Matches: 4
     Pyruvoyl-dependent arginine decarboxylase OS=Methanococcus aeolicus (strain Nankai-3 / ATCC BAA-1280) GN=pdaD PE=3 SV=1

40.  ATPF_VIBC3    Mass: 17707    Score: 31     Expect: 4.4e+002  Matches: 7
     ATP synthase subunit b OS=Vibrio cholerae serotype O1 (strain ATCC 39541 / Classical Ogawa 395 / O395) GN=atpF PE=3 SV=1
     ATPF_VIBCH    Mass: 17707    Score: 31     Expect: 4.4e+002  Matches: 7
     ATP synthase subunit b OS=Vibrio cholerae serotype O1 (strain ATCC 39315 / El Tor Inaba N16961) GN=atpF PE=3 SV=1

41.  RS7_EXIS2    Mass: 17976    Score: 31     Expect: 4.4e+002  Matches: 4
     30S ribosomal protein S7 OS=Exiguobacterium sibiricum (strain DSM 17290 / JCM 13490 / 255-15) GN=rpsG PE=3 SV=1

42.  NPH2_VACCC    Mass: 77899    Score: 31     Expect: 4.5e+002  Matches: 6
     RNA helicase NPH-II OS=Vaccinia virus (strain Copenhagen) GN=NPH2 PE=2 SV=1

43.  ILV5_ORYSJ    Mass: 62686    Score: 31     Expect: 4.6e+002  Matches: 5
     Ketol-acid reductoisomerase, chloroplastic OS=Oryza sativa subsp. japonica GN=Os05g0573700 PE=1 SV=1

44.  KAD_PELPB    Mass: 24348    Score: 31     Expect: 4.7e+002  Matches: 4
     Adenylate kinase OS=Pelodictyon phaeoclathratiforme (strain DSM 5477 / BU-1) GN=adk PE=3 SV=1

45.  MCE_ASFB7    Mass: 100427   Score: 31     Expect: 4.7e+002  Matches: 10
     Probable mRNA-capping enzyme OS=African swine fever virus (strain Badajoz 1971 Vero-adapted) GN=Ba71V-101 PE=3 SV=1
     MCE_ASFP4    Mass: 100439   Score: 31     Expect: 4.7e+002  Matches: 10
     Probable mRNA-capping enzyme OS=African swine fever virus (isolate Tick/South Africa/Pretoriuskop Pr4/1996) GN=Pret-113 PE=3 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (825.8243,1+): <no title>
   Query2 (843.7685,1+): <no title>
   Query3 (857.8776,1+): <no title>
   Query4 (858.8570,1+): <no title>
   Query5 (859.6968,1+): <no title>
```

CA mass+c

Mascot Search Results

```
User           : Y Wang
Email          : kahn20003@gmail.com
Search title   :
Database       : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp      : 14 Apr 2016 at 09:50:46 GMT
Top Score      : 142 for CAH2_BOVIN, Carbonic anhydrase 2 OS=Bos taurus GN=CA2 PE=1 SV=3
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Concise Protein Summary

Significance threshold p< 0.05

Max. number of hits 200

Preferred taxonomy All entries

```
1.    CAH2_BOVIN    Mass: 29096    Score: 142    Expect: 3.5e-009  Matches: 6
      Carbonic anhydrase 2 OS=Bos taurus GN=CA2 PE=1 SV=3
      CAH2_RABIT    Mass: 29598    Score: 62     Expect: 0.35  Matches: 3
      Carbonic anhydrase 2 OS=Oryctolagus cuniculus GN=CA2 PE=1 SV=3
      CAH2_RABIT    Mass: 29598    Score: 62     Expect: 0.35  Matches: 3
      Carbonic anhydrase 2 OS=Oryctolagus cuniculus GN=CA2 PE=1 SV=3
      CAH2_HUMAN    Mass: 29286    Score: 60     Expect: 0.53  Matches: 3
      Carbonic anhydrase 2 OS=Homo sapiens GN=CA2 PE=1 SV=2
      CAH2_RAT    Mass: 29270    Score: 38     Expect: 85  Matches: 2
      Carbonic anhydrase 2 OS=Rattus norvegicus GN=Ca2 PE=1 SV=2
      CAH2_MOUSE    Mass: 29131    Score: 38     Expect: 85  Matches: 2
      Carbonic anhydrase 2 OS=Mus musculus GN=Ca2 PE=1 SV=4

2.    CAH2_SHEEP    Mass: 29193    Score: 89     Expect: 0.00076  Matches: 4
      Carbonic anhydrase 2 OS=Ovis aries GN=CA2 PE=1 SV=2

3.    UBF1_HUMAN    Mass: 89698    Score: 53     Expect: 3  Matches: 4
      Nucleolar transcription factor 1 OS=Homo sapiens GN=UBTF PE=1 SV=1
      UBF1_RAT    Mass: 89729    Score: 52     Expect: 3.2  Matches: 4
      Nucleolar transcription factor 1 OS=Rattus norvegicus GN=Ubtf PE=1 SV=1
      UBF1_MOUSE    Mass: 89801    Score: 52     Expect: 3.2  Matches: 4
      Nucleolar transcription factor 1 OS=Mus musculus GN=Ubtf PE=1 SV=1

4.    MSP2_ASCSU    Mass: 14355    Score: 46     Expect: 13  Matches: 2
      Major sperm protein isoform beta OS=Ascaris suum PE=1 SV=3

5.    PT117_YEAST    Mass: 12331    Score: 40     Expect: 53  Matches: 2
      Protein PET117, mitochondrial OS=Saccharomyces cerevisiae (strain ATCC 204508 / S288c) GN=PET117 PE=3 SV=1

6.    ARGB_LACLA    Mass: 30827    Score: 40     Expect: 54  Matches: 2
      Acetylglutamate kinase OS=Lactococcus lactis subsp. lactis (strain IL1403) GN=argB PE=3 SV=1

7.    LIPA_PARL1    Mass: 36066    Score: 39     Expect: 69  Matches: 2
      Lipoyl synthase OS=Parvibaculum lavamentivorans (strain DS-1 / DSM 13023 / NCIMB 13966) GN=lipA PE=3 SV=1

8.    PURA_SYNFM    Mass: 47713    Score: 39     Expect: 74  Matches: 2
      Adenylosuccinate synthetase OS=Syntrophobacter fumaroxidans (strain DSM 10017 / MPOB) GN=purA PE=3 SV=1

9.    SPRR1_MACMU    Mass: 10309    Score: 38     Expect: 78  Matches: 2
      Cornifin OS=Macaca mulatta GN=SPRR1 PE=2 SV=1

10.   SYGA_HELP2    Mass: 34768    Score: 37     Expect: 1e+002  Matches: 2
      Glycine--tRNA ligase alpha subunit OS=Helicobacter pylori (strain P12) GN=glyQ PE=3 SV=1
      SYGA_HELPG    Mass: 34711    Score: 37     Expect: 1e+002  Matches: 2
      Glycine--tRNA ligase alpha subunit OS=Helicobacter pylori (strain G27) GN=glyQ PE=3 SV=1
      SYGA_HELPG    Mass: 34711    Score: 37     Expect: 1e+002  Matches: 2
      Glycine--tRNA ligase alpha subunit OS=Helicobacter pylori (strain G27) GN=glyQ PE=3 SV=1
      SYGA_HELPJ    Mass: 34760    Score: 37     Expect: 1e+002  Matches: 2
      Glycine--tRNA ligase alpha subunit OS=Helicobacter pylori (strain J99 / ATCC 700824) GN=glyQ PE=3 SV=1
      SYGA_HELPY    Mass: 35204    Score: 37     Expect: 1e+002  Matches: 2
      Glycine--tRNA ligase alpha subunit OS=Helicobacter pylori (strain ATCC 700392 / 26695) GN=glyQ PE=3 SV=1
      SYGA_HELPS    Mass: 34801    Score: 37     Expect: 1.2e+002  Matches: 2
      Glycine--tRNA ligase alpha subunit OS=Helicobacter pylori (strain Shi470) GN=glyQ PE=3 SV=1

11.   ASB15_BOVIN    Mass: 66736    Score: 37     Expect: 1e+002  Matches: 2
      Ankyrin repeat and SOCS box protein 15 OS=Bos taurus GN=ASB15 PE=2 SV=2

12.   LIPA_RHOPS    Mass: 35521    Score: 37     Expect: 1.1e+002  Matches: 2
      Lipoyl synthase OS=Rhodopseudomonas palustris (strain BisB5) GN=lipA PE=3 SV=1

13.   DSE3_CANGA    Mass: 49252    Score: 37     Expect: 1.2e+002  Matches: 2
      Protein DSE3 OS=Candida glabrata (strain ATCC 2001 / CBS 138 / JCM 3761 / NBRC 0622 / NRRL Y-65) GN=DSE3 PE=3 SV=1

14.   ATPF_GEOSL    Mass: 22811    Score: 36     Expect: 1.3e+002  Matches: 2
      ATP synthase subunit b OS=Geobacter sulfurreducens (strain ATCC 51573 / DSM 12127 / PCA) GN=atpF PE=3 SV=2

15.   HSLU_CAUCN    Mass: 46672    Score: 36     Expect: 1.4e+002  Matches: 2
      ATP-dependent protease ATPase subunit HslU OS=Caulobacter crescentus (strain NA1000 / CB15N) GN=hslU PE=3 SV=1
      HSLU_CAUCR    Mass: 46672    Score: 36     Expect: 1.4e+002  Matches: 2
      ATP-dependent protease ATPase subunit HslU OS=Caulobacter crescentus (strain ATCC 19089 / CB15) GN=hslU PE=3 SV=1

16.   YR773_MIMIV    Mass: 57161    Score: 35     Expect: 1.7e+002  Matches: 2
      Putative BTB/POZ domain-containing protein R773 OS=Acanthamoeba polyphaga mimivirus GN=MIMI_R773 PE=3 SV=1

17.   BIOW_SULSY    Mass: 28542    Score: 35     Expect: 1.8e+002  Matches: 2
      6-carboxyhexanoate--CoA ligase OS=Sulfurihydrogenibium sp. (strain YO3AOP1) GN=bioW PE=3 SV=1

18.   CHLB_CHAVU    Mass: 58116    Score: 35     Expect: 1.9e+002  Matches: 2
      Light-independent protochlorophyllide reductase subunit B OS=Chara vulgaris GN=chlB PE=3 SV=1

19.   ILV5_ORYSJ    Mass: 62686    Score: 35     Expect: 1.9e+002  Matches: 2
      Ketol-acid reductoisomerase, chloroplastic OS=Oryza sativa subsp. japonica GN=Os05g0573700 PE=1 SV=1

20.   RS15_THEAC    Mass: 16504    Score: 34     Expect: 2.2e+002  Matches: 2
      30S ribosomal protein S15 OS=Thermoplasma acidophilum (strain ATCC 25905 / DSM 1728 / JCM 9062 / NBRC 15155 / AMRC-C165) GN=rps15 PE=3 SV=1
```

30S ribosomal protein S15 OS=Thermoplasma acidophilum (strain ATCC 25905 / DSM 1728 / JCM 9062 / NBRC 15155 / AMRC-C165) GN=rps15 PE=3 SV=1

21. Y1720_HAEIN **Mass:** 21850 **Score:** 34 **Expect:** 2.3e+002 **Matches:** 2
Uncharacterized protein HI_1720 OS=Haemophilus influenzae (strain ATCC 51907 / DSM 11121 / KW20 / Rd) GN=HI_1720 PE=3 SV=1

22. CAMP1_XENLA **Mass:** 177217 **Score:** 32 **Expect:** 3.6e+002 **Matches:** 3
Calmodulin-regulated spectrin-associated protein 1 OS=Xenopus laevis GN=camsap1 PE=2 SV=1

23. URGCP_MOUSE **Mass:** 105313 **Score:** 32 **Expect:** 3.8e+002 **Matches:** 2
Up-regulator of cell proliferation OS=Mus musculus GN=Urgcp PE=2 SV=1

24. RR7_CHAVU **Mass:** 17886 **Score:** 31 **Expect:** 4.2e+002 **Matches:** 2
30S ribosomal protein S7, chloroplastic OS=Chara vulgaris GN=rps7 PE=3 SV=1

25. LIPA_LEPIC **Mass:** 34167 **Score:** 31 **Expect:** 4.5e+002 **Matches:** 2
Lipoyl synthase OS=Leptospira interrogans serogroup Icterohaemorrhagiae serovar copenhageni (strain Fiocruz L1-130) GN=lipA PE=3 SV=1
LIPA_LEPIN **Mass:** 34225 **Score:** 31 **Expect:** 4.5e+002 **Matches:** 2
Lipoyl synthase OS=Leptospira interrogans serogroup Icterohaemorrhagiae serovar Lai (strain 56601) GN=lipA PE=3 SV=1

26. PXR1_CANAL **Mass:** 31373 **Score:** 30 **Expect:** 5e+002 **Matches:** 2
Protein PXR1 OS=Candida albicans (strain SC5314 / ATCC MYA-2876) GN=PXR1 PE=3 SV=1

27. CBIJ_METMI **Mass:** 28039 **Score:** 30 **Expect:** 5e+002 **Matches:** 2
Cobalt-precorrin-6A reductase OS=Methanococcus maripaludis GN=cbiJ PE=3 SV=2

28. EAF6_YARLI **Mass:** 15451 **Score:** 30 **Expect:** 5.4e+002 **Matches:** 2
Chromatin modification-related protein EAF6 OS=Yarrowia lipolytica (strain CLIB 122 / E 150) GN=EAF6 PE=3 SV=1

29. RYBPB_DANRE **Mass:** 28882 **Score:** 30 **Expect:** 5.5e+002 **Matches:** 2
RING1 and YY1-binding protein B OS=Danio rerio GN=rybpb PE=2 SV=1

30. DDRGK_DROME **Mass:** 34311 **Score:** 30 **Expect:** 5.8e+002 **Matches:** 2
DDRGK domain-containing protein 1 OS=Drosophila melanogaster GN=CG5862 PE=1 SV=1

31. HDRA_METMP **Mass:** 73033 **Score:** 30 **Expect:** 6e+002 **Matches:** 2
CoB--CoM heterodisulfide reductase iron-sulfur subunit A OS=Methanococcus maripaludis (strain S2 / LL) GN=hdrA PE=3 SV=2

32. PNP_CORGB **Mass:** 81413 **Score:** 30 **Expect:** 6.2e+002 **Matches:** 2
Polyribonucleotide nucleotidyltransferase OS=Corynebacterium glutamicum (strain R) GN=pnp PE=3 SV=1
PNP_CORGL **Mass:** 81395 **Score:** 30 **Expect:** 6.2e+002 **Matches:** 2
Polyribonucleotide nucleotidyltransferase OS=Corynebacterium glutamicum (strain ATCC 13032 / DSM 20300 / JCM 1318 / LMG 3730 / NCIMB 10025) GN=pnp PE=3 SV=1
PNP_CORGL **Mass:** 81395 **Score:** 30 **Expect:** 6.2e+002 **Matches:** 2
Polyribonucleotide nucleotidyltransferase OS=Corynebacterium glutamicum (strain ATCC 13032 / DSM 20300 / JCM 1318 / LMG 3730 / NCIMB 10025) GN=pnp PE=3 SV=1

33. SYY_EHRCJ **Mass:** 48023 **Score:** 29 **Expect:** 6.5e+002 **Matches:** 2
Tyrosine--tRNA ligase OS=Ehrlichia canis (strain Jake) GN=tyrS PE=3 SV=1

34. NPH2_VACCC **Mass:** 77899 **Score:** 29 **Expect:** 6.5e+002 **Matches:** 2
RNA helicase NPH-II OS=Vaccinia virus (strain Copenhagen) GN=NPH2 PE=2 SV=1

35. YQ72_SCHPO **Mass:** 64889 **Score:** 29 **Expect:** 6.8e+002 **Matches:** 2
UPF0649 protein C1442.02 OS=Schizosaccharomyces pombe (strain 972 / ATCC 24843) GN=SPCC1442.02 PE=1 SV=1

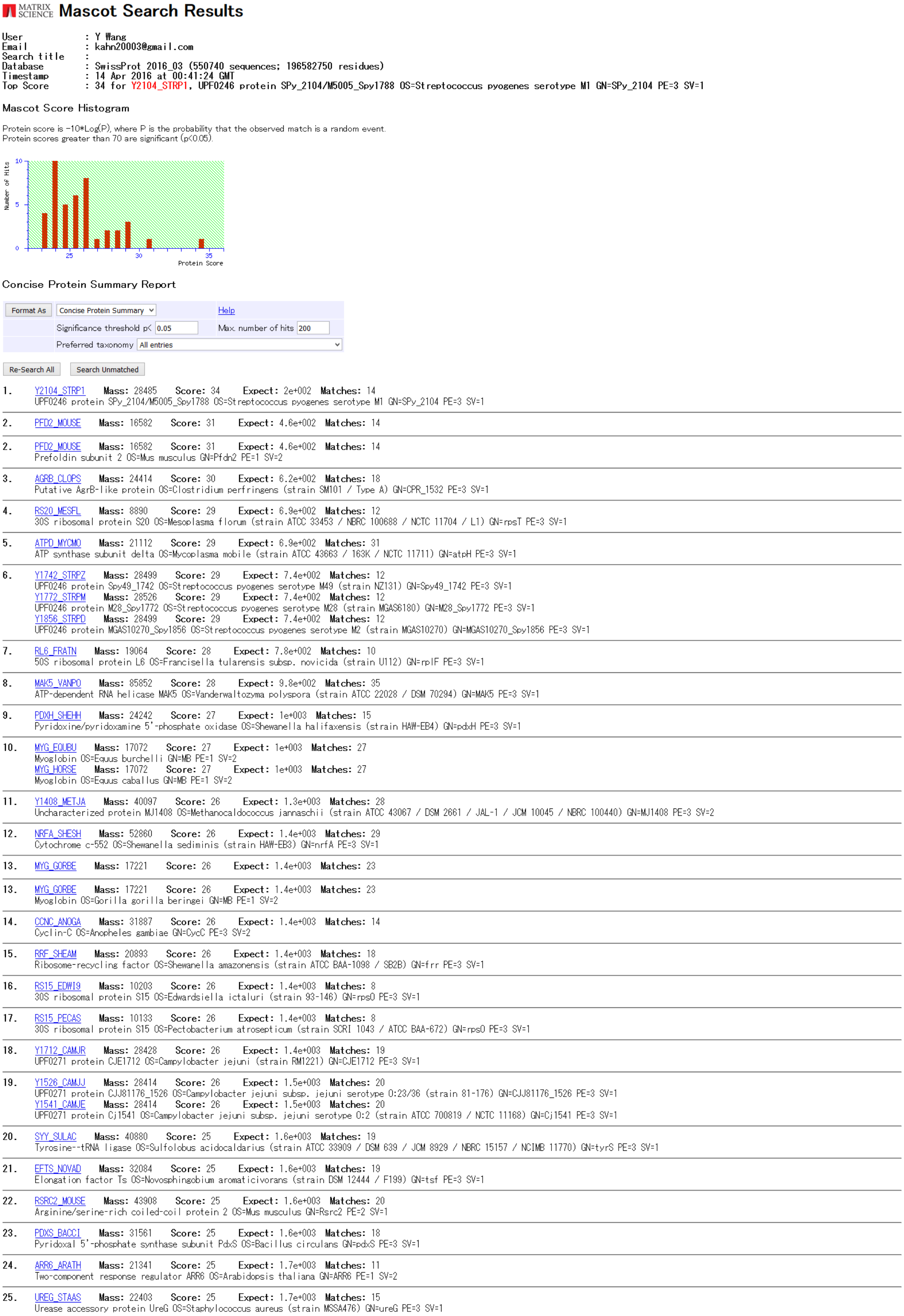
36. SYFB_FRATT **Mass:** 88660 **Score:** 29 **Expect:** 6.9e+002 **Matches:** 2
Phenylalanine--tRNA ligase beta subunit OS=Francisella tularensis subsp. tularensis (strain SCHU S4 / Schu 4) GN=pheT PE=3 SV=1

37. HSLU_TREDE **Mass:** 54034 **Score:** 28 **Expect:** 8e+002 **Matches:** 2
ATP-dependent protease ATPase subunit HslU OS=Treponema denticola (strain ATCC 35405 / CIP 103919 / DSM 14222) GN=hslU PE=3 SV=1

Search Parameters

```
Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (859.7891,1+): <no title>
                       : seq(C-P)
   Query2 (868.7808,1+): <no title>
                       : seq(C-P)
   Query3 (872.7726,1+): <no title>
                       : seq(C-P)
   Query4 (885.6591,1+): <no title>
                       : seq(C-E)
   Query5 (938.7394,1+): <no title>
                       : seq(C-P)
   Query6 (939.7742,1+): <no title>
                       : seq(C-P)
   Query7 (951.7171,1+): <no title>
                       : seq(C-Y)
```

4P mass



Mascot Search Results

```
User          : Y Wang
Email         : kahn20003@gmail.com
Search title  :
Database      : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp     : 14 Apr 2016 at 00:41:24 GMT
Top Score     : 34 for Y2104_STRP1, UPF0246 protein SPy_2104/M5005_Spy1788 OS=Streptococcus pyogenes serotype M1 GN=SPy_2104 PE=3 SV=1
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Concise Protein Summary Report

Format As: Concise Protein Summary
Significance threshold p< 0.05
Max. number of hits 200
Preferred taxonomy All entries

```
1.   Y2104_STRP1   Mass: 28485   Score: 34   Expect: 2e+002  Matches: 14
     UPF0246 protein SPy_2104/M5005_Spy1788 OS=Streptococcus pyogenes serotype M1 GN=SPy_2104 PE=3 SV=1

2.   PFD2_MOUSE   Mass: 16582   Score: 31   Expect: 4.6e+002  Matches: 14

2.   PFD2_MOUSE   Mass: 16582   Score: 31   Expect: 4.6e+002  Matches: 14
     Prefoldin subunit 2 OS=Mus musculus GN=Pfdn2 PE=1 SV=2

3.   AGRB_CLOPS   Mass: 24414   Score: 30   Expect: 6.2e+002  Matches: 18
     Putative AgrB-like protein OS=Clostridium perfringens (strain SM101 / Type A) GN=CPR_1532 PE=3 SV=1

4.   RS20_MESFL   Mass: 8890   Score: 29   Expect: 6.9e+002  Matches: 12
     30S ribosomal protein S20 OS=Mesoplasma florum (strain ATCC 33453 / NBRC 100688 / NCTC 11704 / L1) GN=rpsT PE=3 SV=1

5.   ATPD_MYCMO   Mass: 21112   Score: 29   Expect: 6.9e+002  Matches: 31
     ATP synthase subunit delta OS=Mycoplasma mobile (strain ATCC 43663 / 163K / NCTC 11711) GN=atpH PE=3 SV=1

6.   Y1742_STRPZ   Mass: 28499   Score: 29   Expect: 7.4e+002  Matches: 12
     UPF0246 protein Spy49_1742 OS=Streptococcus pyogenes serotype M49 (strain NZ131) GN=Spy49_1742 PE=3 SV=1
     Y1772_STRPM   Mass: 28526   Score: 29   Expect: 7.4e+002  Matches: 12
     UPF0246 protein M28_Spy1772 OS=Streptococcus pyogenes serotype M28 (strain MGAS6180) GN=M28_Spy1772 PE=3 SV=1
     Y1856_STRPD   Mass: 28499   Score: 29   Expect: 7.4e+002  Matches: 12
     UPF0246 protein MGAS10270_Spy1856 OS=Streptococcus pyogenes serotype M2 (strain MGAS10270) GN=MGAS10270_Spy1856 PE=3 SV=1

7.   RL6_FRATN   Mass: 19064   Score: 28   Expect: 7.8e+002  Matches: 10
     50S ribosomal protein L6 OS=Francisella tularensis subsp. novicida (strain U112) GN=rplF PE=3 SV=1

8.   MAK5_VANPO   Mass: 85852   Score: 28   Expect: 9.8e+002  Matches: 35
     ATP-dependent RNA helicase MAK5 OS=Vanderwaltozyma polyspora (strain ATCC 22028 / DSM 70294) GN=MAK5 PE=3 SV=1

9.   PDXH_SHEHH   Mass: 24242   Score: 27   Expect: 1e+003  Matches: 15
     Pyridoxine/pyridoxamine 5'-phosphate oxidase OS=Shewanella halifaxensis (strain HAW-EB4) GN=pdxH PE=3 SV=1

10.  MYG_EQUBU   Mass: 17072   Score: 27   Expect: 1e+003  Matches: 27
     Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
     MYG_HORSE   Mass: 17072   Score: 27   Expect: 1e+003  Matches: 27
     Myoglobin OS=Equus caballus GN=MB PE=1 SV=2

11.  Y1408_METJA   Mass: 40097   Score: 26   Expect: 1.3e+003  Matches: 28
     Uncharacterized protein MJ1408 OS=Methanocaldococcus jannaschii (strain ATCC 43067 / DSM 2661 / JAL-1 / JCM 10045 / NBRC 100440) GN=MJ1408 PE=3 SV=2

12.  NRFA_SHESH   Mass: 52860   Score: 26   Expect: 1.4e+003  Matches: 29
     Cytochrome c-552 OS=Shewanella sediminis (strain HAW-EB3) GN=nrfA PE=3 SV=1

13.  MYG_GORBE   Mass: 17221   Score: 26   Expect: 1.4e+003  Matches: 23

13.  MYG_GORBE   Mass: 17221   Score: 26   Expect: 1.4e+003  Matches: 23
     Myoglobin OS=Gorilla gorilla beringei GN=MB PE=1 SV=2

14.  CCNC_ANOGA   Mass: 31887   Score: 26   Expect: 1.4e+003  Matches: 14
     Cyclin-C OS=Anopheles gambiae GN=CycC PE=3 SV=2

15.  RRF_SHEAM   Mass: 20893   Score: 26   Expect: 1.4e+003  Matches: 18
     Ribosome-recycling factor OS=Shewanella amazonensis (strain ATCC BAA-1098 / SB2B) GN=frr PE=3 SV=1

16.  RS15_EDWI9   Mass: 10203   Score: 26   Expect: 1.4e+003  Matches: 8
     30S ribosomal protein S15 OS=Edwardsiella ictaluri (strain 93-146) GN=rpsO PE=3 SV=1

17.  RS15_PECAS   Mass: 10133   Score: 26   Expect: 1.4e+003  Matches: 8
     30S ribosomal protein S15 OS=Pectobacterium atrosepticum (strain SCRI 1043 / ATCC BAA-672) GN=rpsO PE=3 SV=1

18.  Y1712_CAMJR   Mass: 28428   Score: 26   Expect: 1.4e+003  Matches: 19
     UPF0271 protein CJE1712 OS=Campylobacter jejuni (strain RM1221) GN=CJE1712 PE=3 SV=1

19.  Y1526_CAMJJ   Mass: 28414   Score: 26   Expect: 1.5e+003  Matches: 20
     UPF0271 protein CJJ81176_1526 OS=Campylobacter jejuni subsp. jejuni serotype O:23/36 (strain 81-176) GN=CJJ81176_1526 PE=3 SV=1
     Y1541_CAMJE   Mass: 28414   Score: 26   Expect: 1.5e+003  Matches: 20
     UPF0271 protein Cj1541 OS=Campylobacter jejuni subsp. jejuni serotype O:2 (strain ATCC 700819 / NCTC 11168) GN=Cj1541 PE=3 SV=1

20.  SYY_SULAC   Mass: 40880   Score: 25   Expect: 1.6e+003  Matches: 19
     Tyrosine--tRNA ligase OS=Sulfolobus acidocaldarius (strain ATCC 33909 / DSM 639 / JCM 8929 / NBRC 15157 / NCIMB 11770) GN=tyrS PE=3 SV=1

21.  EFTS_NOVAD   Mass: 32084   Score: 25   Expect: 1.6e+003  Matches: 19
     Elongation factor Ts OS=Novosphingobium aromaticivorans (strain DSM 12444 / F199) GN=tsf PE=3 SV=1

22.  RSRC2_MOUSE   Mass: 43908   Score: 25   Expect: 1.6e+003  Matches: 20
     Arginine/serine-rich coiled-coil protein 2 OS=Mus musculus GN=Rsrc2 PE=2 SV=1

23.  PDXS_BACCI   Mass: 31561   Score: 25   Expect: 1.6e+003  Matches: 18
     Pyridoxal 5'-phosphate synthase subunit PdxS OS=Bacillus circulans GN=pdxS PE=3 SV=1

24.  ARR6_ARATH   Mass: 21341   Score: 25   Expect: 1.7e+003  Matches: 11
     Two-component response regulator ARR6 OS=Arabidopsis thaliana GN=ARR6 PE=1 SV=2

25.  UREG_STAAS   Mass: 22403   Score: 25   Expect: 1.7e+003  Matches: 15
     Urease accessory protein UreG OS=Staphylococcus aureus (strain MSSA476) GN=ureG PE=3 SV=1
```

```
25.   UREG_STAAS    Mass: 22403    Score: 25     Expect: 1.7e+003  Matches: 15
      Urease accessory protein UreG OS=Staphylococcus aureus (strain MSSA476) GN=ureG PE=3 SV=1
      UREG_STAAW    Mass: 22403    Score: 25     Expect: 1.7e+003  Matches: 15
      Urease accessory protein UreG OS=Staphylococcus aureus (strain MW2) GN=ureG PE=3 SV=1

26.   PFD2_RAT    Mass: 16628    Score: 25     Expect: 1.7e+003  Matches: 10
      Prefoldin subunit 2 OS=Rattus norvegicus GN=Pfdn2 PE=2 SV=1

27.   RIMM_THEMA    Mass: 20580    Score: 25     Expect: 1.8e+003  Matches: 10
      Ribosome maturation factor RimM OS=Thermotoga maritima (strain ATCC 43589 / MSB8 / DSM 3109 / JCM 10099) GN=rimM PE=3 SV=1
      RIMM_THEP1    Mass: 20580    Score: 25     Expect: 1.8e+003  Matches: 10
      Ribosome maturation factor RimM OS=Thermotoga petrophila (strain RKU-1 / ATCC BAA-488 / DSM 13995) GN=rimM PE=3 SV=1

28.   DAPL1_BOVIN    Mass: 11872    Score: 25     Expect: 1.8e+003  Matches: 8
      Death-associated protein-like 1 OS=Bos taurus GN=DAPL1 PE=2 SV=1

29.   RL6_FRAP2    Mass: 19192    Score: 24     Expect: 2e+003  Matches: 11
      50S ribosomal protein L6 OS=Francisella philomiragia subsp. philomiragia (strain ATCC 25017) GN=rplF PE=3 SV=1

30.   RRF_METCA    Mass: 21129    Score: 24     Expect: 2.1e+003  Matches: 9
      Ribosome-recycling factor OS=Methylococcus capsulatus (strain ATCC 33009 / NCIMB 11132 / Bath) GN=frr PE=3 SV=1

31.   GMPR1_MOUSE    Mass: 37980    Score: 24     Expect: 2.1e+003  Matches: 14
      GMP reductase 1 OS=Mus musculus GN=Gmpr PE=1 SV=1

32.   DAPD_BUCBP    Mass: 30924    Score: 24     Expect: 2.1e+003  Matches: 17
      2,3,4,5-tetrahydropyridine-2,6-dicarboxylate N-succinyltransferase OS=Buchnera aphidicola subsp. Baizongia pistaciae (strain Bp) GN=dapD PE=3 SV=1

33.   IBBC2_SOYBN    Mass: 10007    Score: 24     Expect: 2.1e+003  Matches: 8
      Bowman-Birk type proteinase inhibitor C-II OS=Glycine max PE=1 SV=2

34.   RUVA_PROM1    Mass: 25712    Score: 24     Expect: 2.1e+003  Matches: 16
      Holliday junction ATP-dependent DNA helicase RuvA OS=Prochlorococcus marinus (strain NATL1A) GN=ruvA PE=3 SV=1

35.   Y1790_STRP3    Mass: 28453    Score: 24     Expect: 2.2e+003  Matches: 13
      UPF0246 protein SpyM3_1790 OS=Streptococcus pyogenes serotype M3 (strain ATCC BAA-595 / MGAS315) GN=SpyM3_1790 PE=3 SV=1
      Y1790_STRPQ    Mass: 28453    Score: 24     Expect: 2.2e+003  Matches: 13
      UPF0246 protein SPs1787 OS=Streptococcus pyogenes serotype M3 (strain SSI-1) GN=SPs1787 PE=3 SV=1

36.   MYX_CRODR    Mass: 5234    Score: 24     Expect: 2.3e+003  Matches: 7
      Crotamine Ile-19 OS=Crotalus durissus ruruima PE=1 SV=2
36.   MYX_CRODR    Mass: 5234    Score: 24     Expect: 2.3e+003  Matches: 7
      Crotamine Ile-19 OS=Crotalus durissus ruruima PE=1 SV=2

37.   YDDF_BACSU    Mass: 13109    Score: 24     Expect: 2.3e+003  Matches: 13
      Uncharacterized protein YddF OS=Bacillus subtilis (strain 168) GN=yddF PE=4 SV=1

38.   Y088_MYCPN    Mass: 11588    Score: 24     Expect: 2.4e+003  Matches: 8
      Uncharacterized protein MPN_088 OS=Mycoplasma pneumoniae (strain ATCC 29342 / M129) GN=MPN_088 PE=4 SV=1

39.   RS5_STAMF    Mass: 23668    Score: 24     Expect: 2.4e+003  Matches: 13
      30S ribosomal protein S5 OS=Staphylothermus marinus (strain ATCC 43588 / DSM 3639 / JCM 9404 / F1) GN=rps5 PE=3 SV=1

40.   UBIE_LARHH    Mass: 27238    Score: 24     Expect: 2.5e+003  Matches: 17
      Ubiquinone/menaquinone biosynthesis C-methyltransferase UbiE OS=Laribacter hongkongensis (strain HLHK9) GN=ubiE PE=3 SV=1

41.   PAN_METTH    Mass: 46140    Score: 24     Expect: 2.5e+003  Matches: 15
      Proteasome-activating nucleotidase OS=Methanothermobacter thermautotrophicus (strain ATCC 29096 / DSM 1053 / JCM 10044 / NBRC 100330 / Delta H) GN=pan PE=3 SV=1

42.   GLMU_CARHZ    Mass: 48583    Score: 23     Expect: 2.6e+003  Matches: 19
      Bifunctional protein GlmU OS=Carboxydothermus hydrogenoformans (strain ATCC BAA-161 / DSM 6008 / Z-2901) GN=glmU PE=3 SV=1

43.   RL22_DINSH    Mass: 14224    Score: 23     Expect: 2.6e+003  Matches: 8
      50S ribosomal protein L22 OS=Dinoroseobacter shibae (strain DSM 16493 / NCIMB 14021 / DFL 12) GN=rplV PE=3 SV=1

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (514.6834,1+): <no title>
   Query2 (514.6894,1+): <no title>
   Query3 (515.6702,1+): <no title>
   Query4 (516.6091,1+): <no title>
   Query5 (516.6136,1+): <no title>
   Query6 (516.6147,1+): <no title>
   Query7 (516.6308,1+): <no title>
```

]

4P mass/mass+c

Mascot Search Results

```
User          : kahn20002
Email         : kahn20003@gmail.com
Search title  : 4P 0.5 1miss
Database      : SwissProt 2016_04 (550960 sequences; 196692942 residues)
Timestamp     : 17 Apr 2016 at 06:25:46 GMT
Top Score     : 91 for MYG_GORBE, Myoglobin OS=Gorilla gorilla beringei GN=MB PE=1 SV=2
```

Mascot Score Histogram

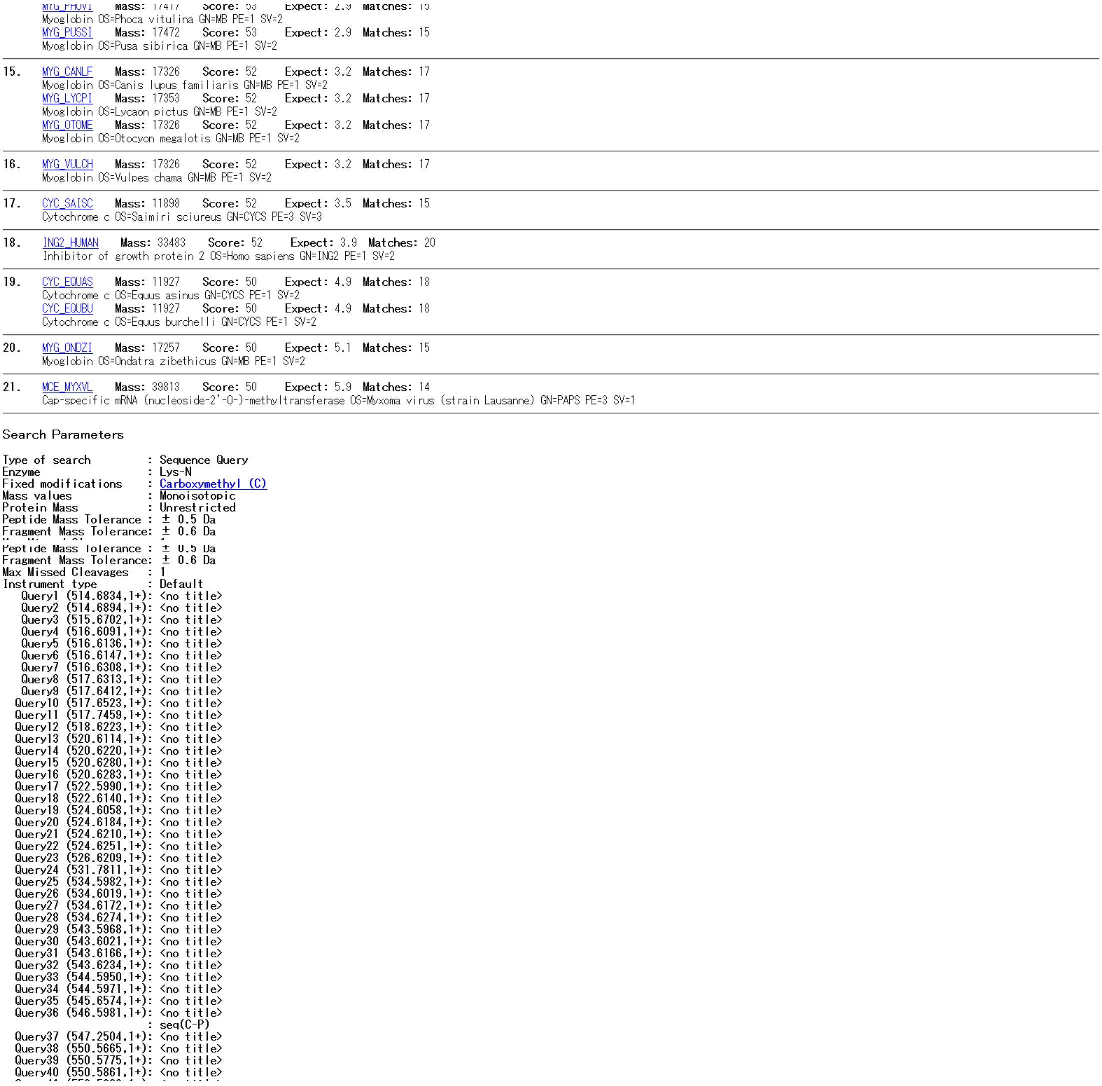
Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Score Distribution

Concise Protein Summary Report

Concise Protein Summary
Significance threshold p< 0.05
Max. number of hits 50
Preferred taxonomy All entries

```
1.    MYG_GORBE   Mass: 17221   Score: 91   Expect: 0.0004  Matches: 21
      Myoglobin OS=Gorilla gorilla beringei GN=MB PE=1 SV=2
      MYG_HUMAN   Mass: 17231   Score: 85   Expect: 0.0016  Matches: 20
      Myoglobin OS=Homo sapiens GN=MB PE=1 SV=2
      MYG_HYLAG   Mass: 17261   Score: 85   Expect: 0.0016  Matches: 20
      Myoglobin OS=Hylobates agilis GN=MB PE=1 SV=2
      MYG_SYMSY   Mass: 17261   Score: 85   Expect: 0.0016  Matches: 20
      Myoglobin OS=Symphalangus syndactylus GN=MB PE=1 SV=2
      MYG_PANTR   Mass: 17240   Score: 81   Expect: 0.0048  Matches: 19
      Myoglobin OS=Pan troglodytes GN=MB PE=1 SV=2
      MYG_PONPY   Mass: 17187   Score: 81   Expect: 0.0048  Matches: 19
      Myoglobin OS=Pongo pygmaeus GN=MB PE=1 SV=2
      MYG_ERYPA   Mass: 17137   Score: 80   Expect: 0.0062  Matches: 18
      Myoglobin OS=Erythrocebus patas GN=MB PE=1 SV=2
      MYG_ERYPA   Mass: 17137   Score: 80   Expect: 0.0062  Matches: 18
      Myoglobin OS=Erythrocebus patas GN=MB PE=1 SV=2
      MYG_PAPAN   Mass: 17137   Score: 80   Expect: 0.0062  Matches: 18
      Myoglobin OS=Papio anubis GN=MB PE=1 SV=2
      MYG_SEMEN   Mass: 17137   Score: 80   Expect: 0.0062  Matches: 18
      Myoglobin OS=Semnopithecus entellus GN=MB PE=1 SV=2
      MYG_MACFA   Mass: 17165   Score: 63   Expect: 0.28  Matches: 15
      Myoglobin OS=Macaca fascicularis GN=MB PE=1 SV=2

2.    MYG_EQUBU   Mass: 17072   Score: 90   Expect: 0.00062  Matches: 23
      Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
      MYG_HORSE   Mass: 17072   Score: 90   Expect: 0.00062  Matches: 23
      Myoglobin OS=Equus caballus GN=MB PE=1 SV=2

3.    MYG_BALBO   Mass: 17238   Score: 83   Expect: 0.0025  Matches: 22
      Myoglobin OS=Balaenoptera borealis GN=MB PE=2 SV=3
      MYG_BALED   Mass: 17238   Score: 83   Expect: 0.0025  Matches: 22
      Myoglobin OS=Balaenoptera edeni GN=MB PE=2 SV=3
      MYG_ESCRO   Mass: 17238   Score: 83   Expect: 0.0025  Matches: 22
      Myoglobin OS=Eschrichtius robustus GN=MB PE=1 SV=2
      MYG_MEGNO   Mass: 17252   Score: 83   Expect: 0.0025  Matches: 22
      Myoglobin OS=Megaptera novaeangliae GN=MB PE=1 SV=2
      MYG_BALAC   Mass: 17275   Score: 79   Expect: 0.0071  Matches: 21
      Myoglobin OS=Balaenoptera acutorostrata GN=MB PE=1 SV=2
      MYG_BALPH   Mass: 17206   Score: 79   Expect: 0.0071  Matches: 21
      Myoglobin OS=Balaenoptera physalus GN=MB PE=1 SV=2
      MYG_DELLE   Mass: 17165   Score: 61   Expect: 0.46  Matches: 17
      Myoglobin OS=Delphinapterus leucas GN=MB PE=1 SV=2
      MYG_GLOME   Mass: 17205   Score: 61   Expect: 0.46  Matches: 17
      Myoglobin OS=Globicephala melas GN=MB PE=1 SV=2
      MYG_ZIPCA   Mass: 17298   Score: 61   Expect: 0.46  Matches: 17
      Myoglobin OS=Ziphius cavirostris GN=MB PE=1 SV=2
      MYG_INDPC   Mass: 17270   Score: 59   Expect: 0.68  Matches: 17
      Myoglobin OS=Indopacetus pacificus GN=MB PE=2 SV=3
      MYG_MESCA   Mass: 17256   Score: 59   Expect: 0.68  Matches: 17
      Myoglobin OS=Mesoplodon carlhubbsi GN=MB PE=1 SV=2
      MYG_MESST   Mass: 17256   Score: 59   Expect: 0.68  Matches: 17
      Myoglobin OS=Mesoplodon stejnegeri GN=MB PE=2 SV=3

4.    MYG_INIGE   Mass: 17191   Score: 82   Expect: 0.0036  Matches: 19

4.    MYG_INIGE   Mass: 17191   Score: 82   Expect: 0.0036  Matches: 19
      Myoglobin OS=Inia geoffrensis GN=MB PE=1 SV=2

5.    MYG_PHYCD   Mass: 17320   Score: 81   Expect: 0.0045  Matches: 16
      Myoglobin OS=Physeter catodon GN=MB PE=1 SV=2
      MYG_KOGBR   Mass: 17357   Score: 62   Expect: 0.31  Matches: 14
      Myoglobin OS=Kogia breviceps GN=MB PE=2 SV=3
      MYG_KOGSI   Mass: 17357   Score: 62   Expect: 0.31  Matches: 14
      Myoglobin OS=Kogia sima GN=MB PE=1 SV=2

6.    ALBU_BOVIN   Mass: 71279   Score: 74   Expect: 0.02  Matches: 31
      Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4

7.    MYG_RABIT   Mass: 17210   Score: 70   Expect: 0.056  Matches: 18
      Myoglobin OS=Oryctolagus cuniculus GN=MB PE=1 SV=2

8.    MYG_CASFI   Mass: 17140   Score: 70   Expect: 0.059  Matches: 21
      Myoglobin OS=Castor fiber GN=MB PE=1 SV=2

9.    MYG_PERPO   Mass: 17052   Score: 67   Expect: 0.11  Matches: 20
      Myoglobin OS=Perodicticus potto edwarsi GN=MB PE=1 SV=2

10.   MYG_MACRU   Mass: 17258   Score: 66   Expect: 0.13  Matches: 17
      Myoglobin OS=Macropus rufus GN=MB PE=1 SV=2

11.   CYC_HORSE   Mass: 11941   Score: 64   Expect: 0.21  Matches: 20
      Cytochrome c OS=Equus caballus GN=CYCS PE=1 SV=2

12.   MYG_PHODA   Mass: 17221   Score: 58   Expect: 0.87  Matches: 12
      Myoglobin OS=Phocoenoides dalli dalli GN=MB PE=1 SV=2
      MYG_PHOPH   Mass: 17221   Score: 58   Expect: 0.87  Matches: 12
      Myoglobin OS=Phocoena phocoena GN=MB PE=1 SV=2

13.   MYG_ROUAE   Mass: 17068   Score: 55   Expect: 1.9  Matches: 16
      Myoglobin OS=Rousettus aegyptiacus GN=MB PE=1 SV=2

14.   MYG_HALGR   Mass: 17417   Score: 53   Expect: 2.9  Matches: 15
      Myoglobin OS=Halichoerus grypus GN=MB PE=1 SV=2
      MYG_PHOVI   Mass: 17417   Score: 53   Expect: 2.9  Matches: 15
      Myoglobin OS=Phoca vitulina GN=MB PE=1 SV=2
```



```
    MYG_PHOVI    Mass: 17417    Score: 53    Expect: 2.9  Matches: 15
    Myoglobin OS=Phoca vitulina GN=MB PE=1 SV=2
    MYG_PUSSI    Mass: 17472    Score: 53    Expect: 2.9  Matches: 15
    Myoglobin OS=Pusa sibirica GN=MB PE=1 SV=2
________________________________________________________________________________
15. MYG_CANLF    Mass: 17326    Score: 52    Expect: 3.2  Matches: 17
    Myoglobin OS=Canis lupus familiaris GN=MB PE=1 SV=2
    MYG_LYCPI    Mass: 17353    Score: 52    Expect: 3.2  Matches: 17
    Myoglobin OS=Lycaon pictus GN=MB PE=1 SV=2
    MYG_OTOME    Mass: 17326    Score: 52    Expect: 3.2  Matches: 17
    Myoglobin OS=Otocyon megalotis GN=MB PE=1 SV=2
________________________________________________________________________________
16. MYG_VULCH    Mass: 17326    Score: 52    Expect: 3.2  Matches: 17
    Myoglobin OS=Vulpes chama GN=MB PE=1 SV=2
________________________________________________________________________________
17. CYC_SAISC    Mass: 11898    Score: 52    Expect: 3.5  Matches: 15
    Cytochrome c OS=Saimiri sciureus GN=CYCS PE=3 SV=3
________________________________________________________________________________
18. ING2_HUMAN    Mass: 33483    Score: 52    Expect: 3.9  Matches: 20
    Inhibitor of growth protein 2 OS=Homo sapiens GN=ING2 PE=1 SV=2
________________________________________________________________________________
19. CYC_EQUAS    Mass: 11927    Score: 50    Expect: 4.9  Matches: 18
    Cytochrome c OS=Equus asinus GN=CYCS PE=1 SV=2
    CYC_EQUBU    Mass: 11927    Score: 50    Expect: 4.9  Matches: 18
    Cytochrome c OS=Equus burchelli GN=CYCS PE=1 SV=2
________________________________________________________________________________
20. MYG_ONDZI    Mass: 17257    Score: 50    Expect: 5.1  Matches: 15
    Myoglobin OS=Ondatra zibethicus GN=MB PE=1 SV=2
________________________________________________________________________________
21. MCE_MYXVL    Mass: 39813    Score: 50    Expect: 5.9  Matches: 14
    Cap-specific mRNA (nucleoside-2'-O-)-methyltransferase OS=Myxoma virus (strain Lausanne) GN=PAPS PE=3 SV=1
________________________________________________________________________________

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (514.6834,1+): <no title>
   Query2 (514.6894,1+): <no title>
   Query3 (515.6702,1+): <no title>
   Query4 (516.6091,1+): <no title>
   Query5 (516.6136,1+): <no title>
   Query6 (516.6147,1+): <no title>
   Query7 (516.6308,1+): <no title>
   Query8 (517.6313,1+): <no title>
   Query9 (517.6412,1+): <no title>
  Query10 (517.6523,1+): <no title>
  Query11 (517.7459,1+): <no title>
  Query12 (518.6223,1+): <no title>
  Query13 (520.6114,1+): <no title>
  Query14 (520.6220,1+): <no title>
  Query15 (520.6280,1+): <no title>
  Query16 (520.6283,1+): <no title>
  Query17 (522.5990,1+): <no title>
  Query18 (522.6140,1+): <no title>
  Query19 (524.6058,1+): <no title>
  Query20 (524.6184,1+): <no title>
  Query21 (524.6210,1+): <no title>
  Query22 (524.6251,1+): <no title>
  Query23 (526.6209,1+): <no title>
  Query24 (531.7811,1+): <no title>
  Query25 (534.5982,1+): <no title>
  Query26 (534.6019,1+): <no title>
  Query27 (534.6172,1+): <no title>
  Query28 (534.6274,1+): <no title>
  Query29 (543.5968,1+): <no title>
  Query30 (543.6021,1+): <no title>
  Query31 (543.6166,1+): <no title>
  Query32 (543.6234,1+): <no title>
  Query33 (544.5950,1+): <no title>
  Query34 (544.5971,1+): <no title>
  Query35 (545.6574,1+): <no title>
  Query36 (546.5981,1+): <no title>
                       : seq(C-P)
  Query37 (547.2504,1+): <no title>
  Query38 (550.5665,1+): <no title>
  Query39 (550.5775,1+): <no title>
  Query40 (550.5861,1+): <no title>
```

4P mass+c

Mascot Search Results

```
User          : kahn20002
Email         : kahn20003@gmail.com
Search title  : 4P 0.5 1miss
Database      : SwissProt 2016_03 (550740 sequences; 196582750 residues)
Timestamp     : 17 Apr 2016 at 04:44:57 GMT
Top Score     : 111 for MYG_EQUBU, Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
```

Mascot Score Histogram

Protein score is -10*Log(P), where P is the probability that the observed match is a random event.
Protein scores greater than 70 are significant (p<0.05).

Score Distribution

Concise Protein Summary Report

Format As: Concise Protein Summary
Significance threshold p< 0.05
Max. number of hits 50
Preferred taxonomy: All entries

```
1.      MYG_EQUBU    Mass: 17072    Score: 111    Expect: 4.4e-06  Matches: 7
        Myoglobin OS=Equus burchelli GN=MB PE=1 SV=2
        MYG_HORSE    Mass: 17072    Score: 111    Expect: 4.4e-06  Matches: 7
        Myoglobin OS=Equus caballus GN=MB PE=1 SV=2
        MYG_PHYCD    Mass: 17320    Score: 93     Expect: 0.00029  Matches: 6
        Myoglobin OS=Physeter catodon GN=MB PE=1 SV=2
        MYG_BALAC    Mass: 17275    Score: 90     Expect: 0.0005  Matches: 6
        Myoglobin OS=Balaenoptera acutorostrata GN=MB PE=1 SV=2
        MYG_BALBO    Mass: 17238    Score: 90     Expect: 0.0005  Matches: 6
        Myoglobin OS=Balaenoptera borealis GN=MB PE=2 SV=3
        MYG_BALED    Mass: 17238    Score: 90     Expect: 0.0005  Matches: 6
        Myoglobin OS=Balaenoptera edeni GN=MB PE=2 SV=3
        MYG_BALPH    Mass: 17206    Score: 90     Expect: 0.0005  Matches: 6
        Myoglobin OS=Balaenoptera physalus GN=MB PE=1 SV=2
        MYG_BALPH    Mass: 17206    Score: 90     Expect: 0.0005  Matches: 6
        Myoglobin OS=Balaenoptera physalus GN=MB PE=1 SV=2
        MYG_ESCRO    Mass: 17238    Score: 90     Expect: 0.0005  Matches: 6
        Myoglobin OS=Eschrichtius robustus GN=MB PE=1 SV=2
        MYG_INIGE    Mass: 17191    Score: 90     Expect: 0.0005  Matches: 6
        Myoglobin OS=Inia geoffrensis GN=MB PE=1 SV=2
        MYG_MEGNO    Mass: 17252    Score: 90     Expect: 0.0005  Matches: 6
        Myoglobin OS=Megaptera novaeangliae GN=MB PE=1 SV=2
        MYG_KOGBR    Mass: 17357    Score: 72     Expect: 0.038  Matches: 5
        Myoglobin OS=Kogia breviceps GN=MB PE=2 SV=3
        MYG_KOGSI    Mass: 17357    Score: 72     Expect: 0.038  Matches: 5
        Myoglobin OS=Kogia sima GN=MB PE=1 SV=2
        MYG_DELLE    Mass: 17165    Score: 70     Expect: 0.059  Matches: 5
        Myoglobin OS=Delphinapterus leucas GN=MB PE=1 SV=2
        MYG_GLOME    Mass: 17205    Score: 70     Expect: 0.059  Matches: 5
        Myoglobin OS=Globicephala melas GN=MB PE=1 SV=2
        MYG_ZIPCA    Mass: 17298    Score: 70     Expect: 0.059  Matches: 5
        Myoglobin OS=Ziphius cavirostris GN=MB PE=1 SV=2
        MYG_INDPC    Mass: 17270    Score: 68     Expect: 0.089  Matches: 5
        Myoglobin OS=Indopacetus pacificus GN=MB PE=2 SV=3
        MYG_MACRU    Mass: 17258    Score: 68     Expect: 0.089  Matches: 5
        Myoglobin OS=Macropus rufus GN=MB PE=1 SV=2
        MYG_MESCA    Mass: 17256    Score: 68     Expect: 0.089  Matches: 5
        Myoglobin OS=Mesoplodon carlhubbsi GN=MB PE=1 SV=2
        MYG_MESST    Mass: 17256    Score: 68     Expect: 0.089  Matches: 5
        Myoglobin OS=Mesoplodon stejnegeri GN=MB PE=2 SV=3
        MYG_PHODA    Mass: 17221    Score: 60     Expect: 0.51  Matches: 4
        Myoglobin OS=Phocoenoides dalli dalli GN=MB PE=1 SV=2
        MYG_PHOPH    Mass: 17221    Score: 60     Expect: 0.51  Matches: 4
        Myoglobin OS=Phocoena phocoena GN=MB PE=1 SV=2
        MYG_HALGR    Mass: 17417    Score: 55     Expect: 1.7  Matches: 4
        Myoglobin OS=Halichoerus grypus GN=MB PE=1 SV=2
        MYG_PHOVI    Mass: 17417    Score: 55     Expect: 1.7  Matches: 4
        Myoglobin OS=Phoca vitulina GN=MB PE=1 SV=2
        MYG_PUSSI    Mass: 17472    Score: 55     Expect: 1.7  Matches: 4
        Myoglobin OS=Pusa sibirica GN=MB PE=1 SV=2
        MYG_CANLF    Mass: 17326    Score: 52     Expect: 3.3  Matches: 4
        Myoglobin OS=Canis lupus familiaris GN=MB PE=1 SV=2
        MYG_LYCPI    Mass: 17353    Score: 52     Expect: 3.3  Matches: 4
        Myoglobin OS=Lycaon pictus GN=MB PE=1 SV=2
        MYG_LYCPI    Mass: 17353    Score: 52     Expect: 3.3  Matches: 4
        Myoglobin OS=Lycaon pictus GN=MB PE=1 SV=2
        MYG_OTOME    Mass: 17326    Score: 52     Expect: 3.3  Matches: 4
        Myoglobin OS=Otocyon megalotis GN=MB PE=1 SV=2
        MYG_VULCH    Mass: 17326    Score: 52     Expect: 3.3  Matches: 4
        Myoglobin OS=Vulpes chama GN=MB PE=1 SV=2
        MYG_AOTTR    Mass: 17150    Score: 51     Expect: 4.5  Matches: 4
        Myoglobin OS=Aotus trivirgatus GN=MB PE=1 SV=2
        MYG_CALJA    Mass: 17164    Score: 51     Expect: 4.5  Matches: 4
        Myoglobin OS=Callithrix jacchus GN=MB PE=1 SV=2
```

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2.      MYG_ERYPA    Mass: 17137    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Erythrocebus patas GN=MB PE=1 SV=2
        MYG_GORBE    Mass: 17221    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Gorilla gorilla beringei GN=MB PE=1 SV=2
        MYG_HUMAN    Mass: 17231    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Homo sapiens GN=MB PE=1 SV=2
        MYG_HYLAG    Mass: 17261    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Hylobates agilis GN=MB PE=1 SV=2
        MYG_PANTR    Mass: 17240    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Pan troglodytes GN=MB PE=1 SV=2
        MYG_PAPAN    Mass: 17137    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Papio anubis GN=MB PE=1 SV=2
        MYG_PONPY    Mass: 17187    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Pongo pygmaeus GN=MB PE=1 SV=2
        MYG_SEMEN    Mass: 17137    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Semnopithecus entellus GN=MB PE=1 SV=2
        MYG_SYMSY    Mass: 17261    Score: 90     Expect: 0.0006  Matches: 6
        Myoglobin OS=Symphalangus syndactylus GN=MB PE=1 SV=2
        MYG_PERPO    Mass: 17052    Score: 74     Expect: 0.021  Matches: 5
        Myoglobin OS=Perodicticus potto edwarsi GN=MB PE=1 SV=2
        MYG_RABIT    Mass: 17210    Score: 74     Expect: 0.021  Matches: 5
        Myoglobin OS=Oryctolagus cuniculus GN=MB PE=1 SV=2
        MYG_CASFI    Mass: 17140    Score: 72     Expect: 0.032  Matches: 5
        Myoglobin OS=Castor fiber GN=MB PE=1 SV=2
        MYG_MACFA    Mass: 17165    Score: 72     Expect: 0.032  Matches: 5
        Myoglobin OS=Macaca fascicularis GN=MB PE=1 SV=2
        MYG_ONDZI    Mass: 17257    Score: 56     Expect: 1.2  Matches: 4
        Myoglobin OS=Ondatra zibethicus GN=MB PE=1 SV=2
        MYG_ROUAE    Mass: 17068    Score: 56     Expect: 1.5  Matches: 4
        Myoglobin OS=Rousettus aegyptiacus GN=MB PE=1 SV=2
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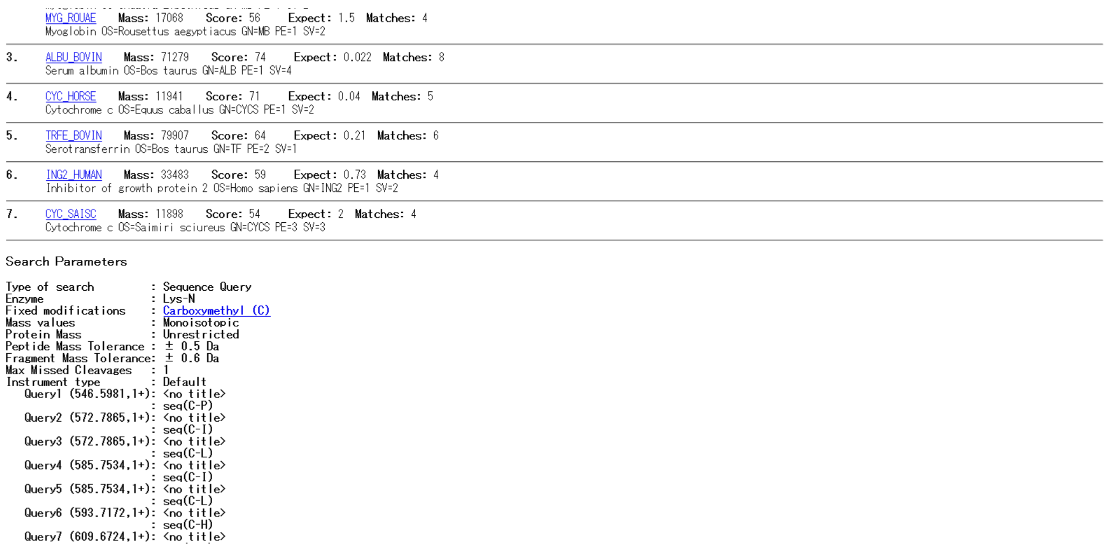

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        MYG_ROUAE    Mass: 17068    Score: 56     Expect: 1.5  Matches: 4
        Myoglobin OS=Rousettus aegyptiacus GN=MB PE=1 SV=2
3.      ALBU_BOVIN    Mass: 71279    Score: 74     Expect: 0.022  Matches: 8
        Serum albumin OS=Bos taurus GN=ALB PE=1 SV=4
4.      CYC_HORSE    Mass: 11941    Score: 71     Expect: 0.04  Matches: 5
        Cytochrome c OS=Equus caballus GN=CYCS PE=1 SV=2
5.      TRFE_BOVIN    Mass: 79907    Score: 64     Expect: 0.21  Matches: 6
        Serotransferrin OS=Bos taurus GN=TF PE=2 SV=1
6.      ING2_HUMAN    Mass: 33483    Score: 59     Expect: 0.73  Matches: 4
        Inhibitor of growth protein 2 OS=Homo sapiens GN=ING2 PE=1 SV=2
7.      CYC_SAISC    Mass: 11898    Score: 54     Expect: 2  Matches: 4
        Cytochrome c OS=Saimiri sciureus GN=CYCS PE=3 SV=3

Search Parameters

Type of search         : Sequence Query
Enzyme                 : Lys-N
Fixed modifications    : Carboxymethyl (C)
Mass values            : Monoisotopic
Protein Mass           : Unrestricted
Peptide Mass Tolerance : ± 0.5 Da
Fragment Mass Tolerance: ± 0.6 Da
Max Missed Cleavages   : 1
Instrument type        : Default
   Query1 (546.5981,1+): <no title>
                       : seq(C-P)
   Query2 (572.7865,1+): <no title>
                       : seq(C-I)
   Query3 (572.7865,1+): <no title>
                       : seq(C-L)
   Query4 (585.7534,1+): <no title>
                       : seq(C-I)
   Query5 (585.7534,1+): <no title>
                       : seq(C-L)
   Query6 (593.7172,1+): <no title>
                       : seq(C-H)
   Query7 (609.6724,1+): <no title>
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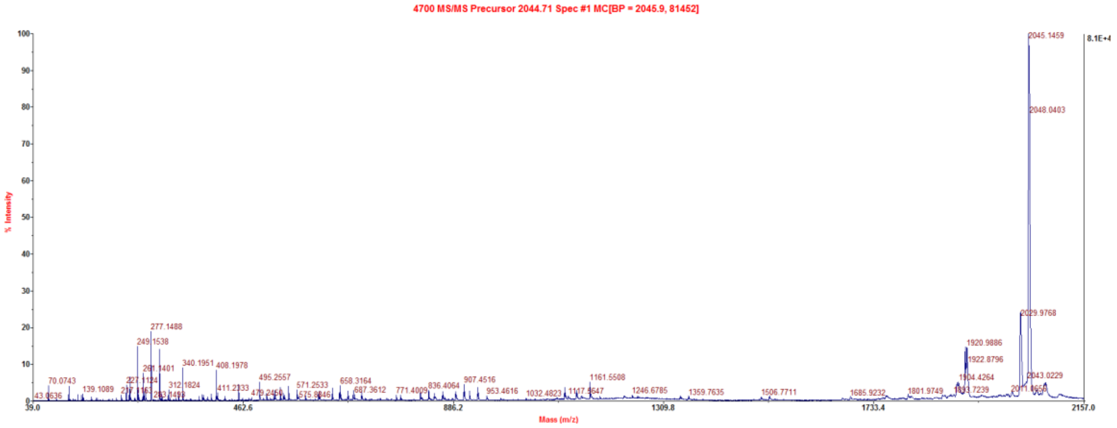
Spectra of peptides derived from BSA digested by different proteases

Trypsin digest

2044.71 RHPYFYAPELLYYANK



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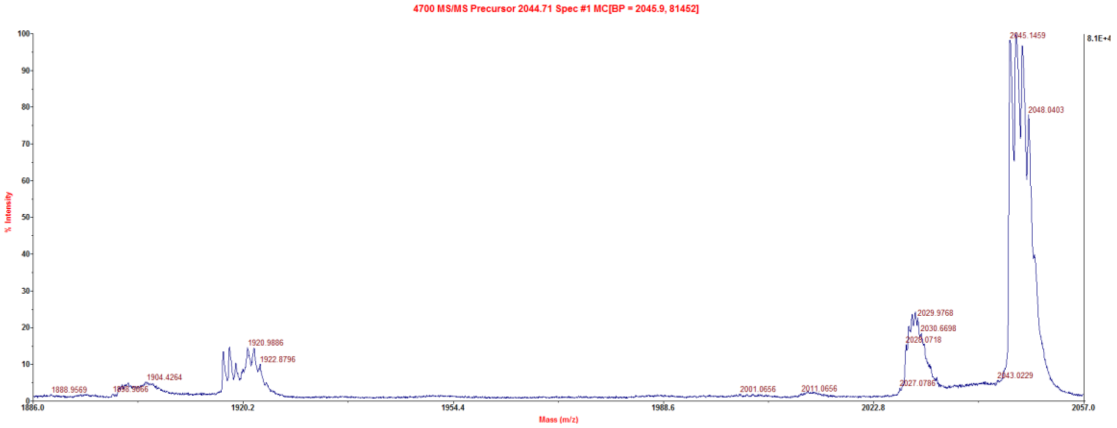
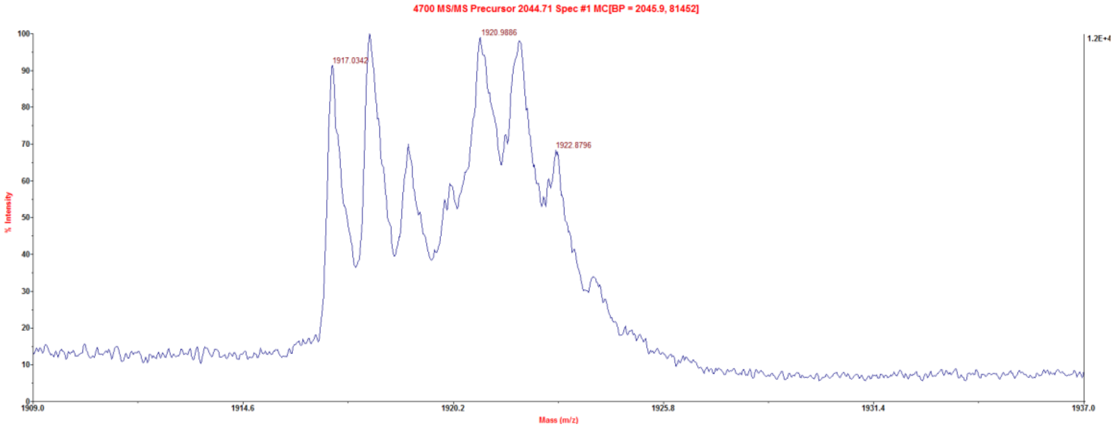
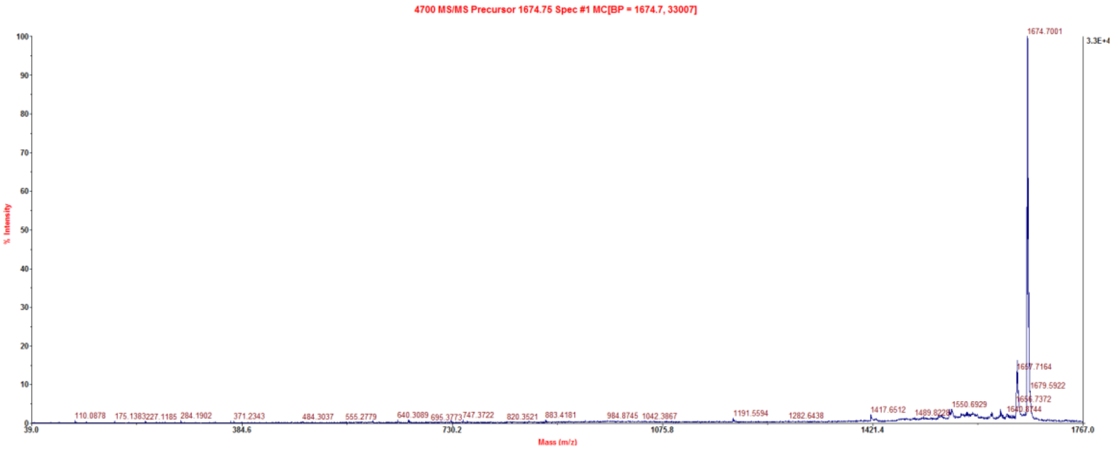


RHPYFYAPELLYYAN b+18 ion



1674.75 RHPYFYAPELLYYAN



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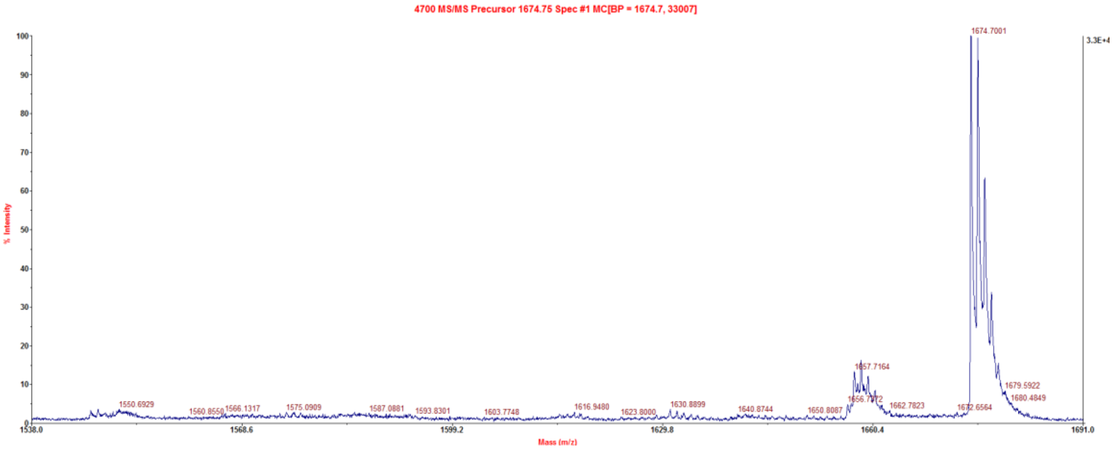
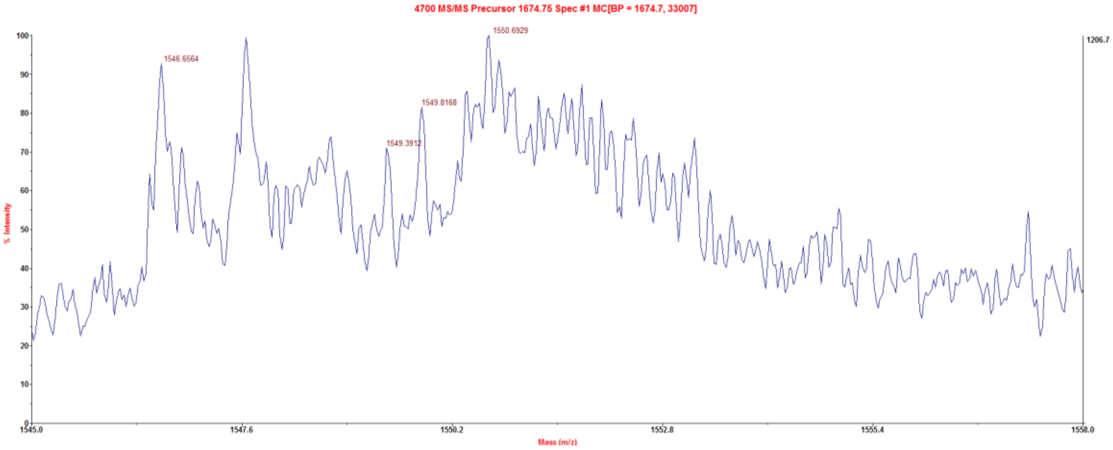
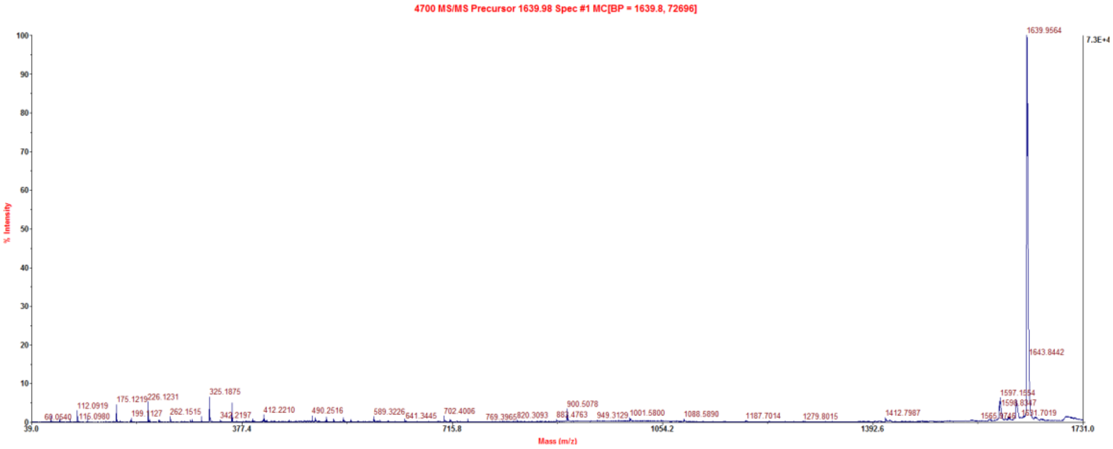


RHPYFYAPELLYYA b+18 ion



1534.83   LKECmcCmcDKPLLEK



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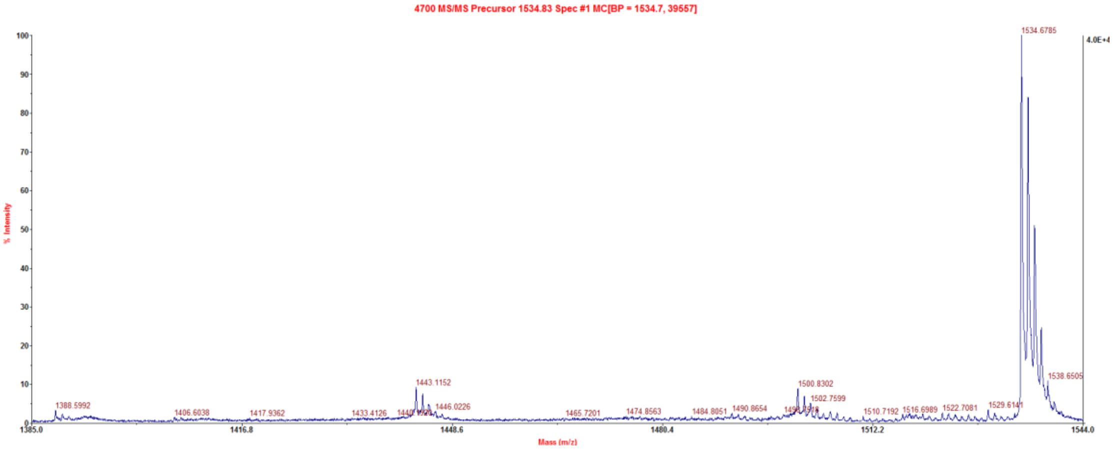
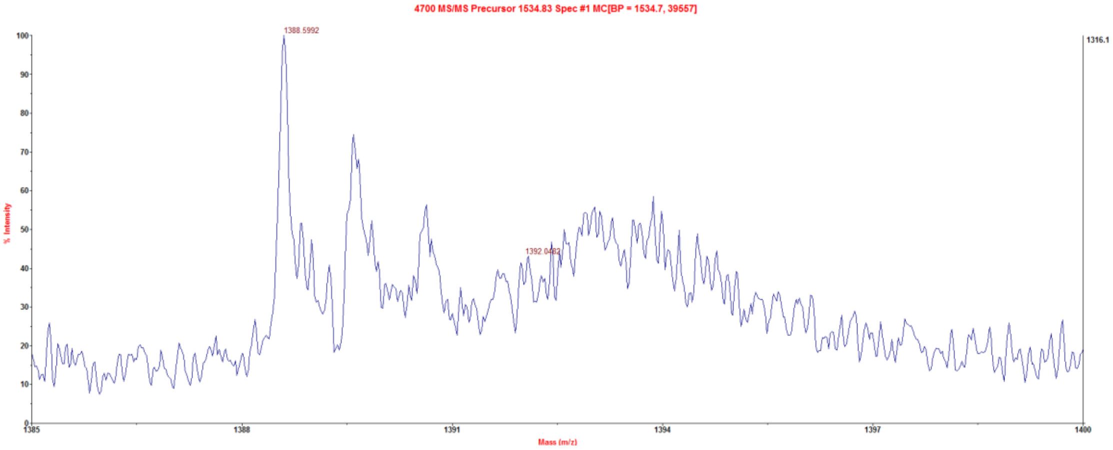
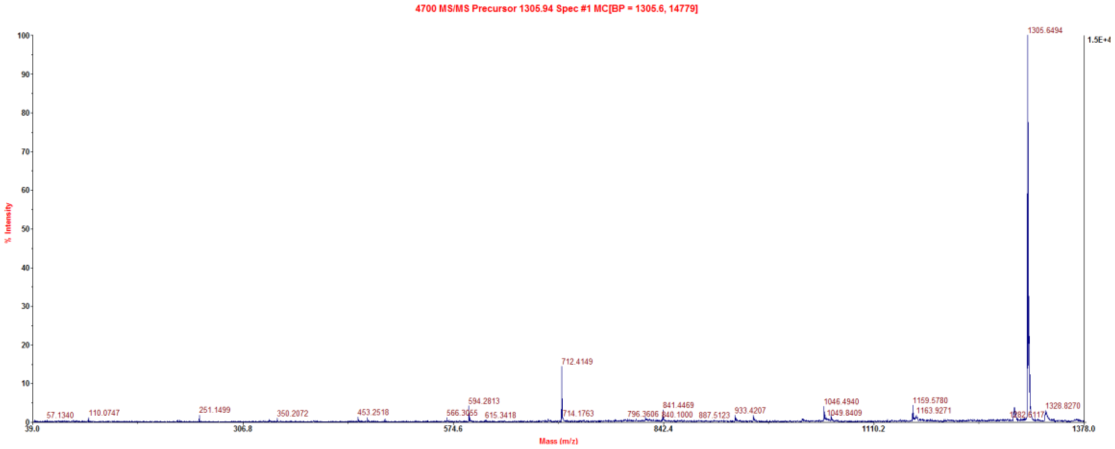


LKECmcCmcDKPLLE   b ion



1305.94 HLVDEPQNLIK



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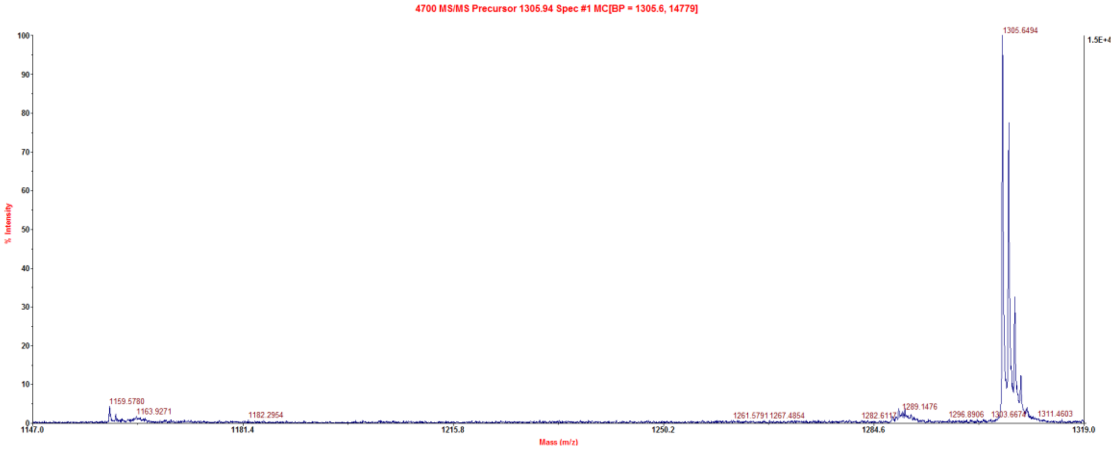
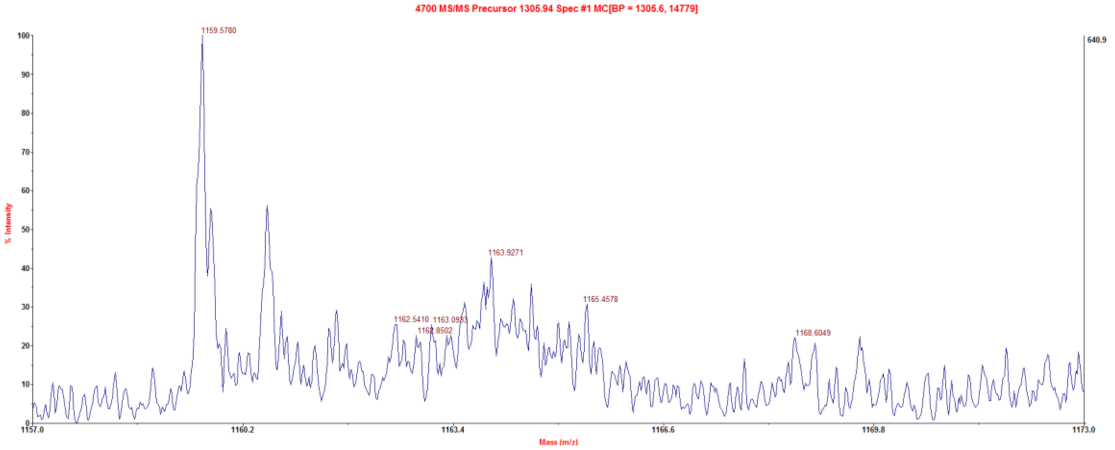
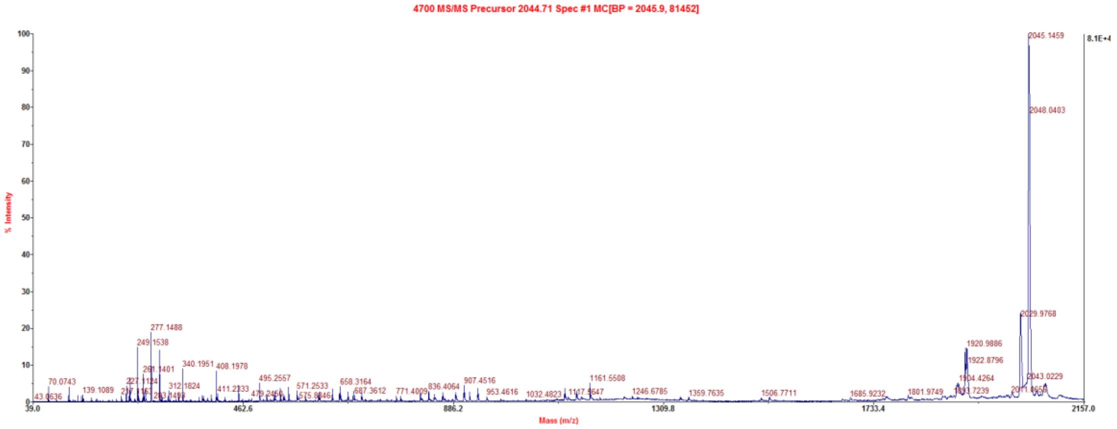


HLVDEPQNLI b ion

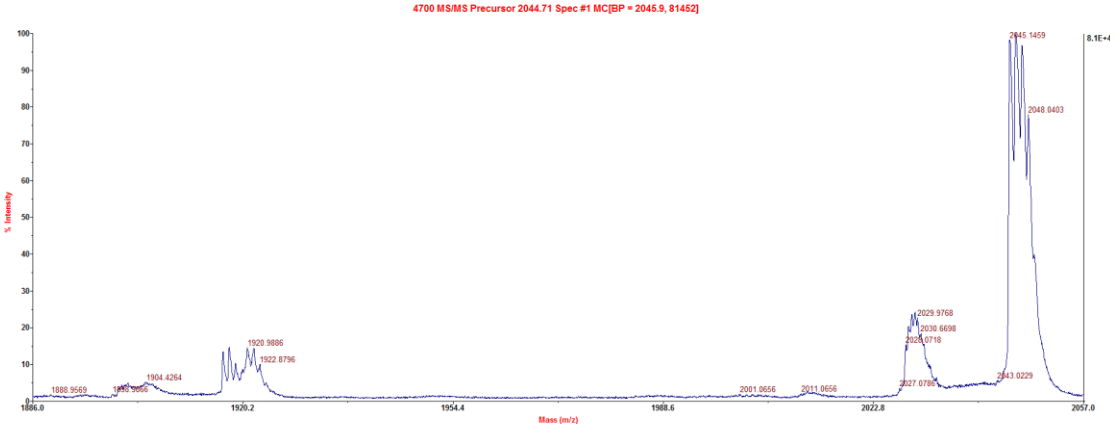


AspN digest

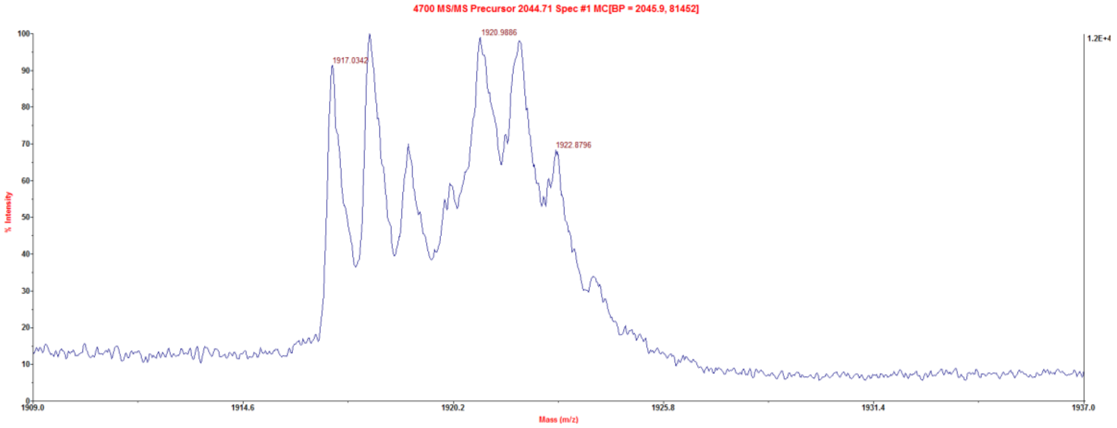
2044.71 RHPYFYAPELLYYANK



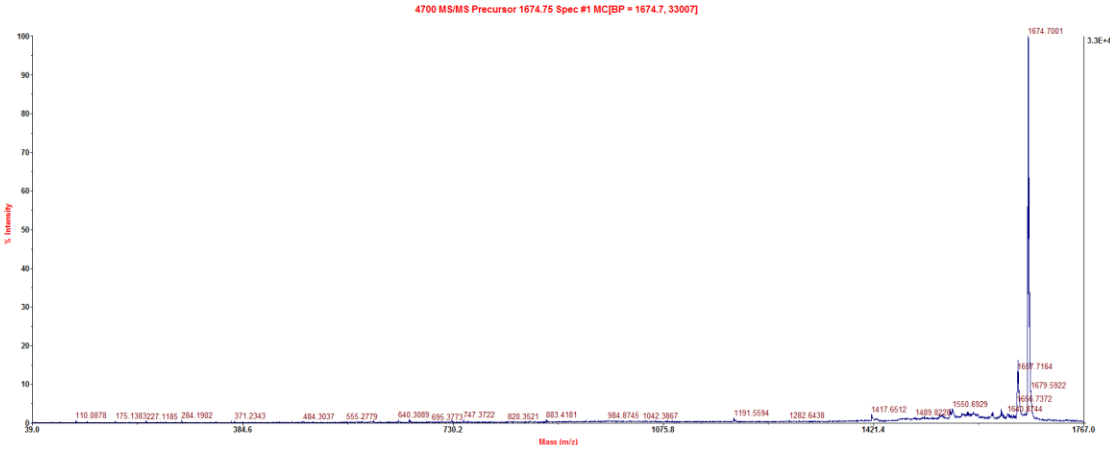
Precursor



RHPYFYAPELLYYAN b+18 ion



1674.75 RHPYFYAPELLYYAN



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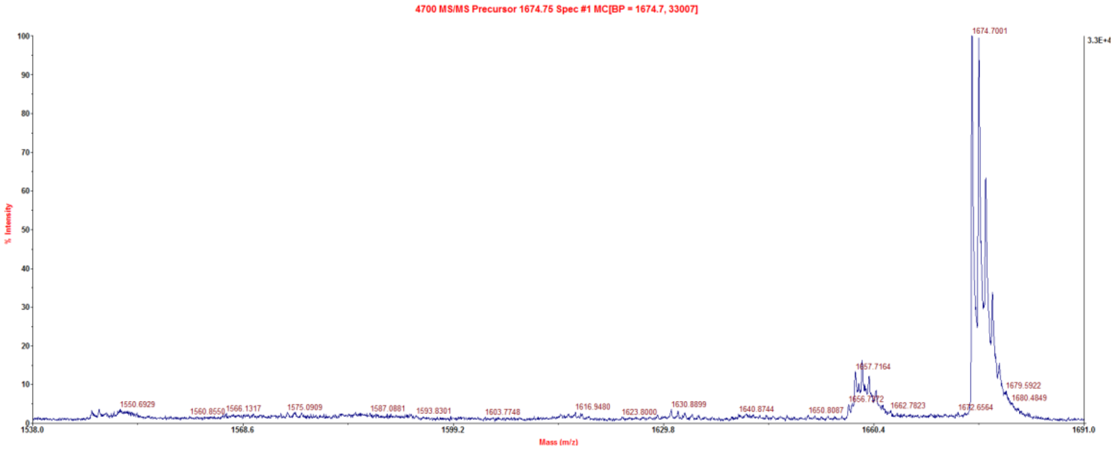
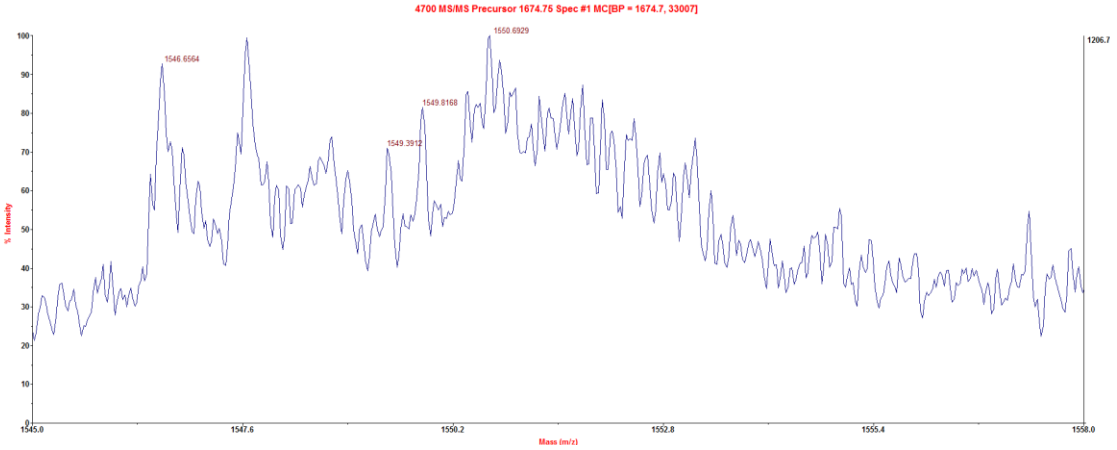
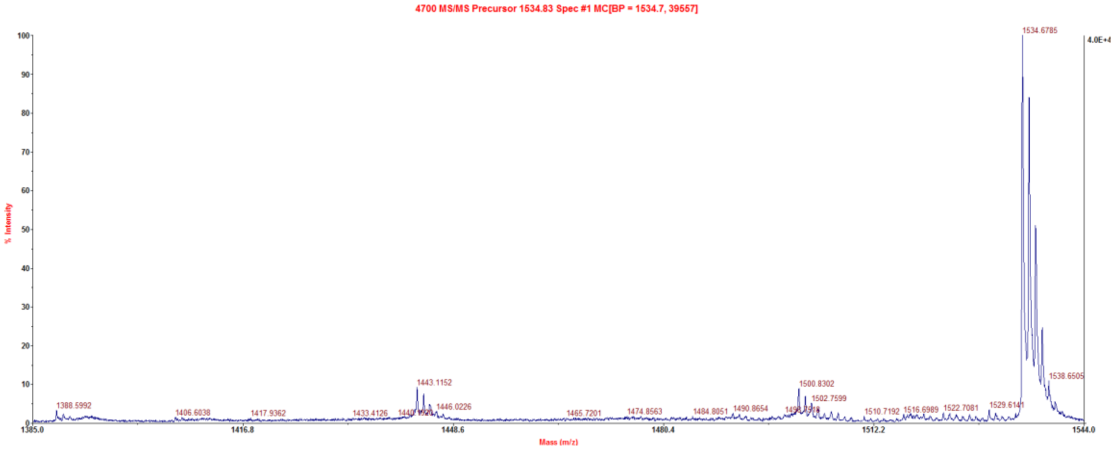


RHPYFYAPELLYYA b+18 ion

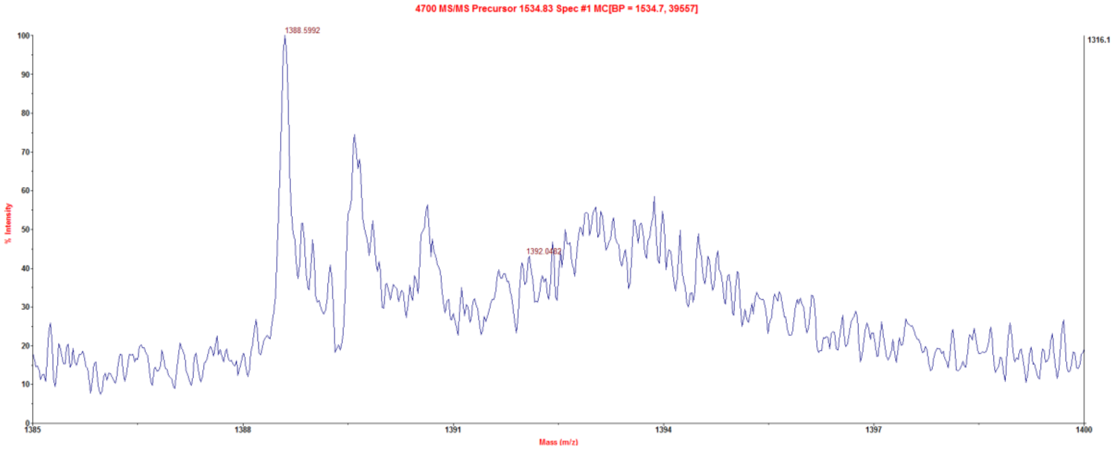


1534.83   LKECmcCmcDKPLLEK

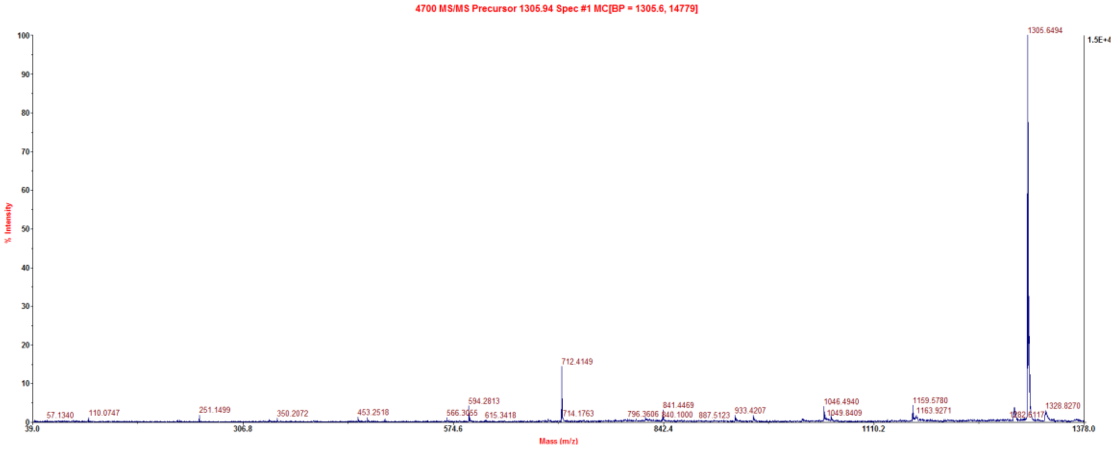
Precursor



LKECmcCmcDKPLLE   b ion



1305.94 HLVDEPQNLIK



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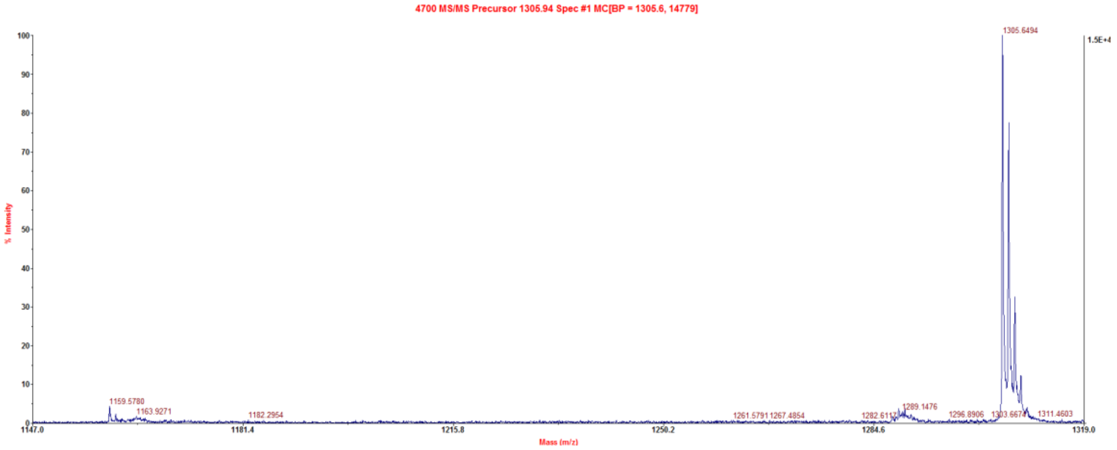
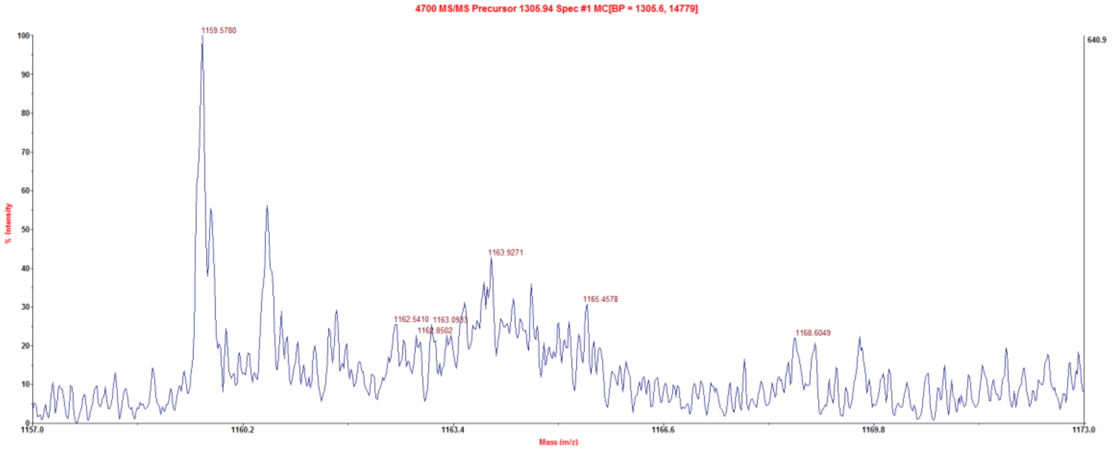
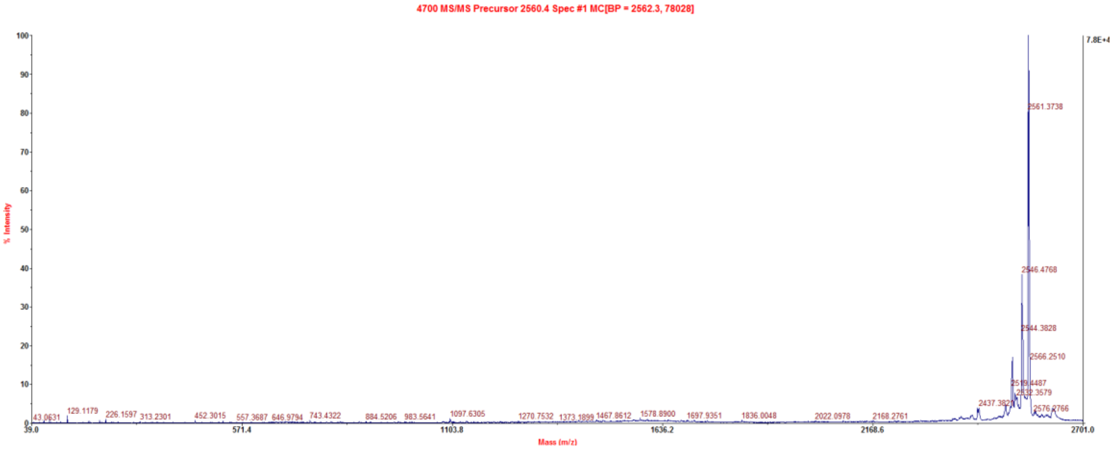


HLVDEPQNLI b ion

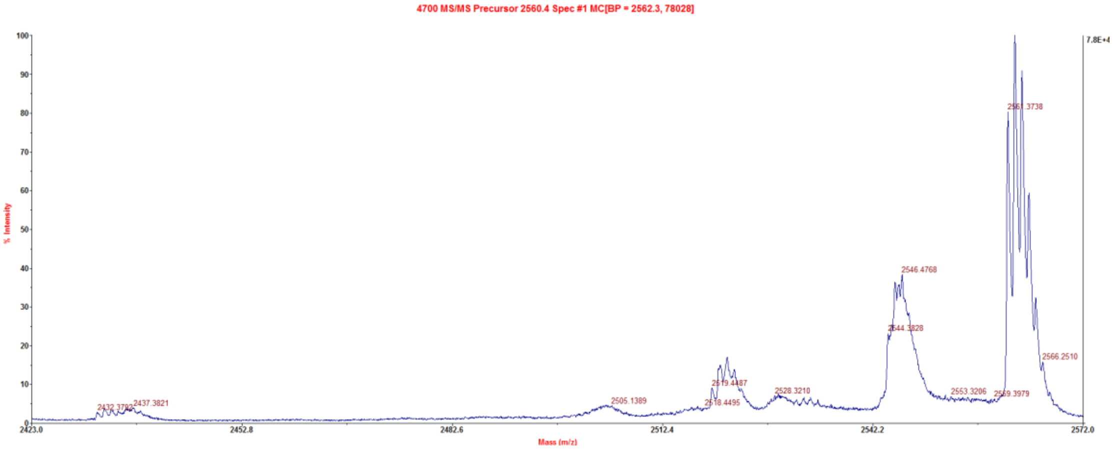


GluC digest

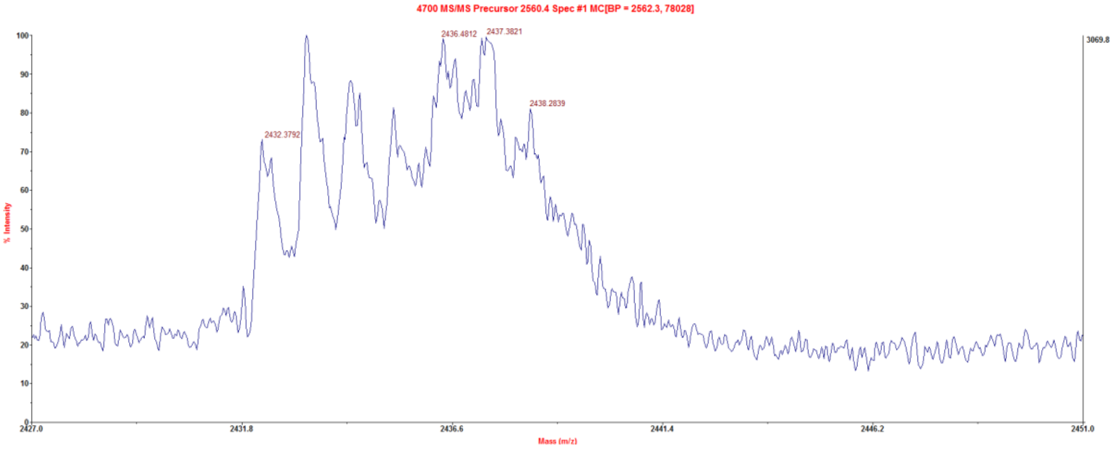
2560.40 RALKAWSVARLSQKFPKAEFVE



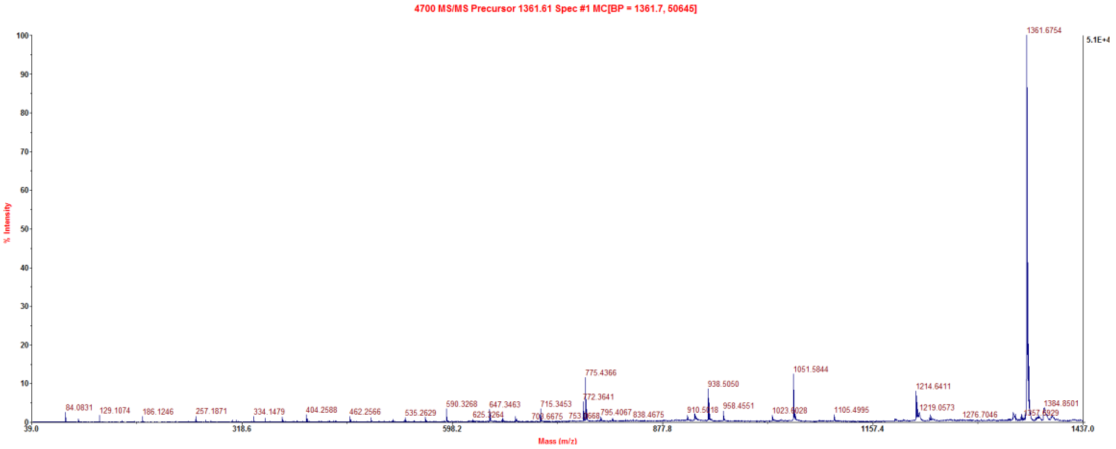
Precursor



RALKAWSVARLSQKFPKAEFV b+18 ion



1361.61 KKFWGKYLYE



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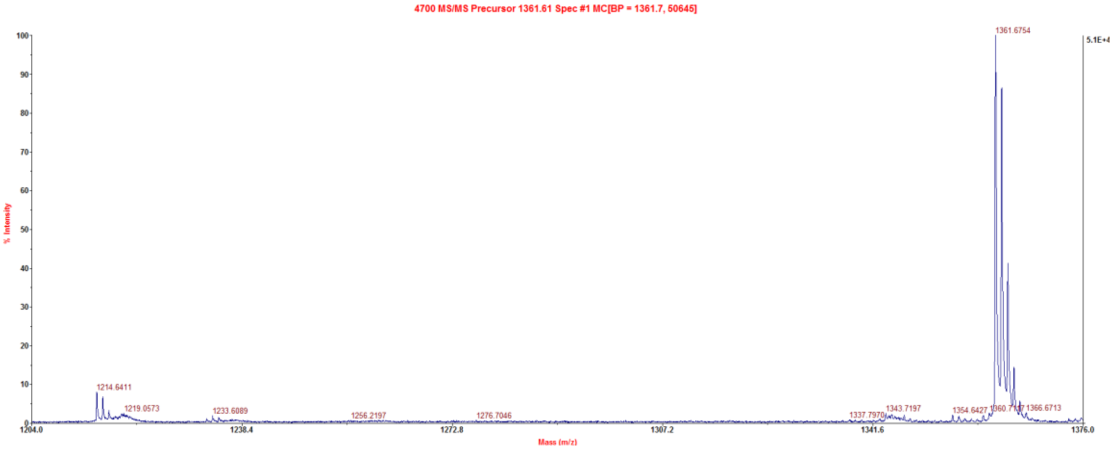
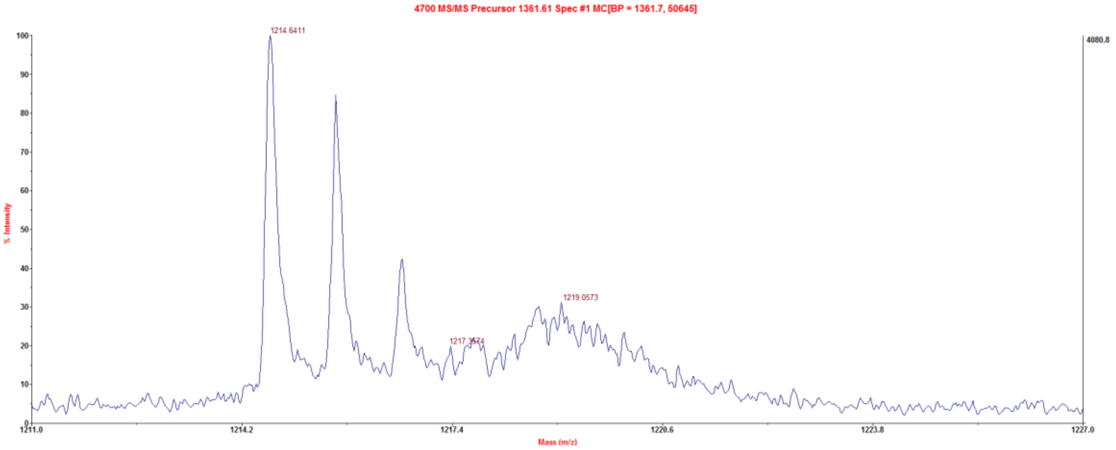
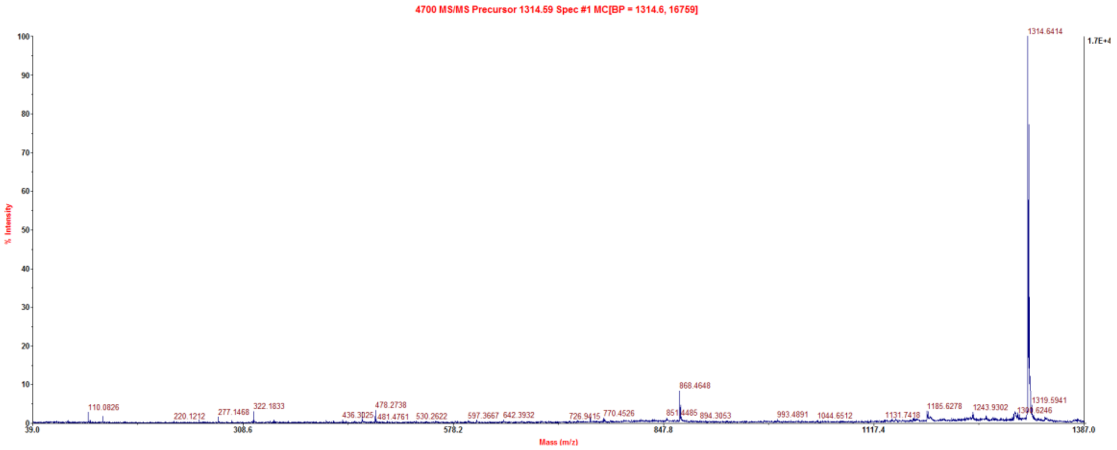


KKFWGKYLY b ion



1314.59 IAHRFKDLGEE



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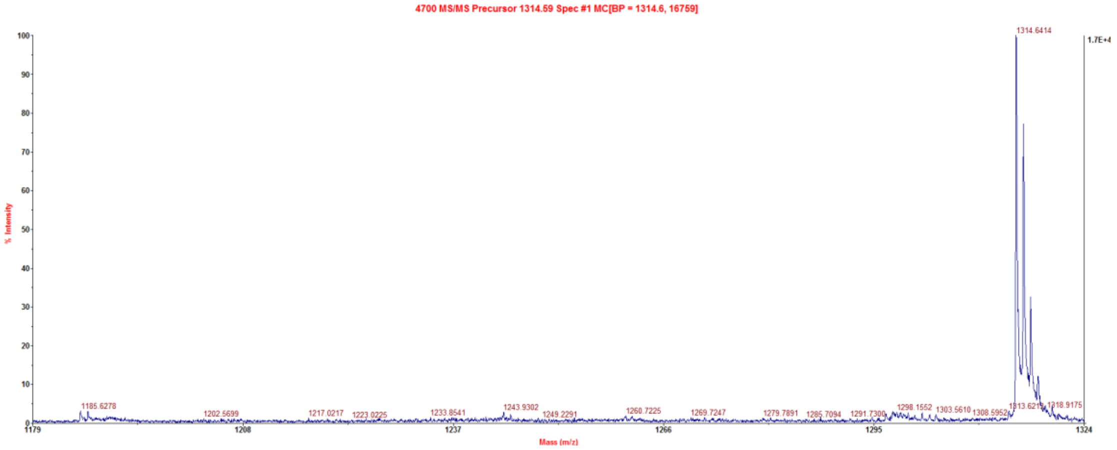
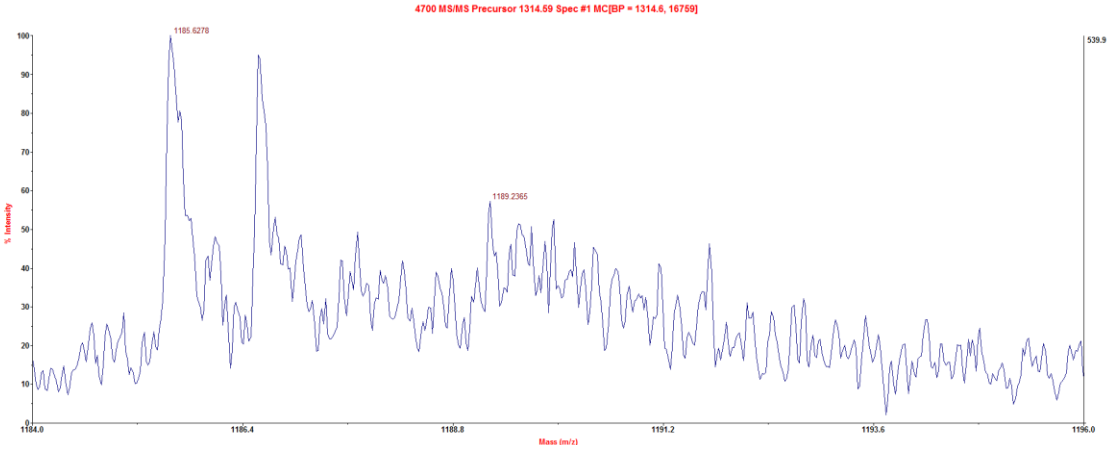
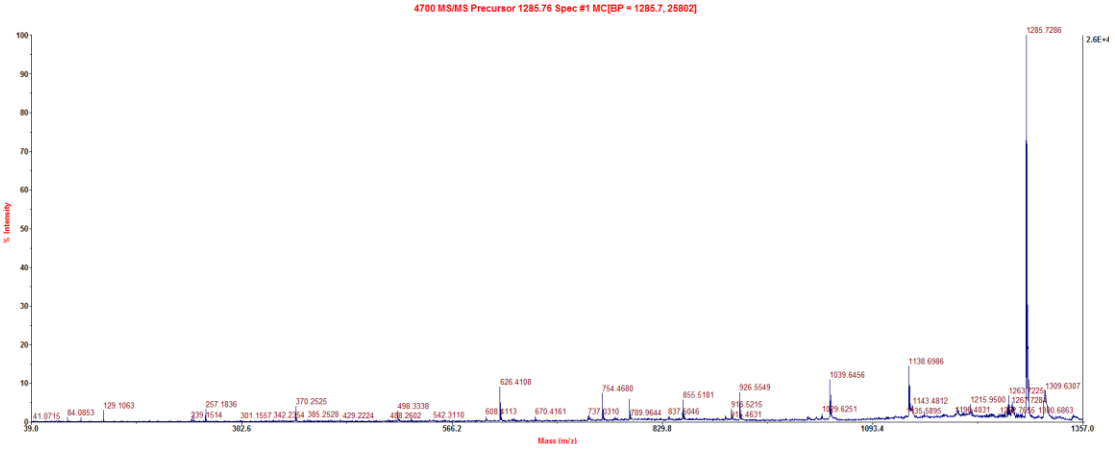


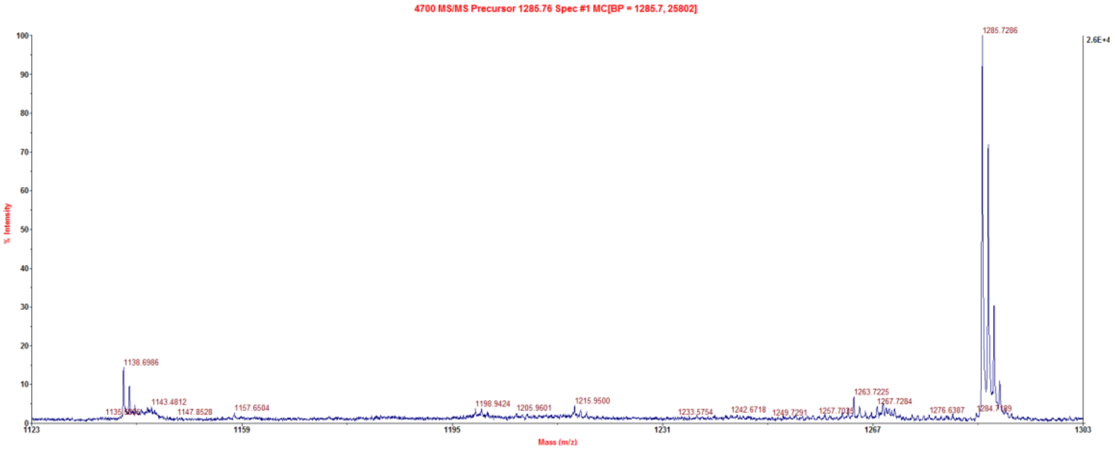
IAHRFKDLGE b+18 ion



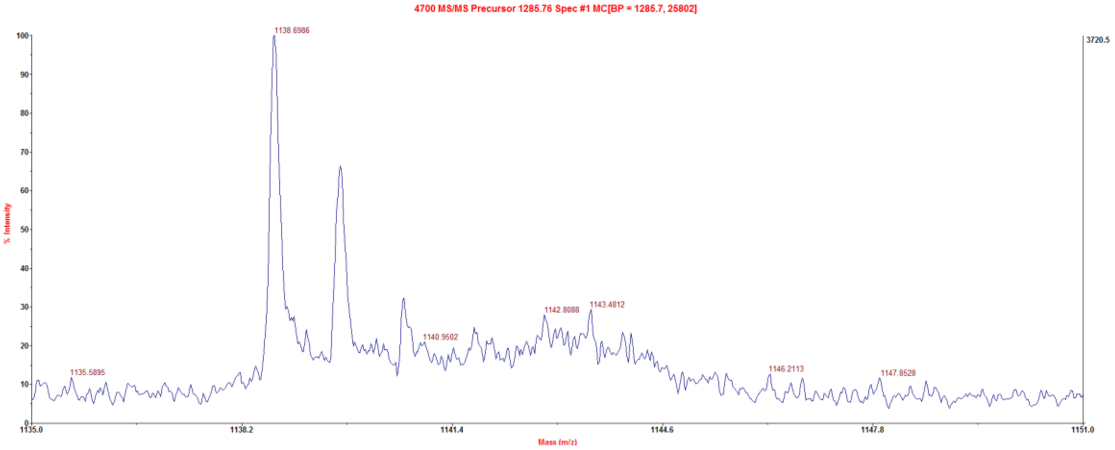
1285.76 KQIKKQTALVE



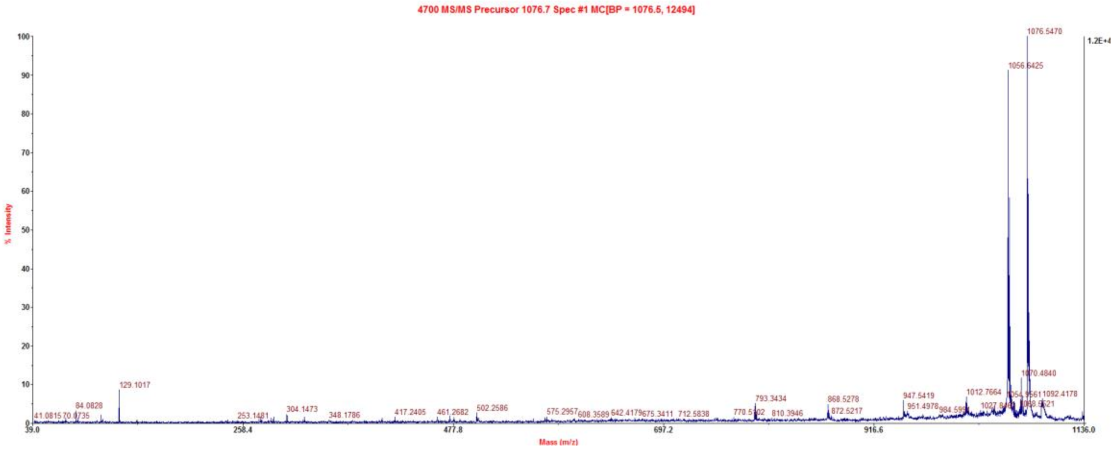
Precursor



KQIKKQTALV b ion



1076.70 LCmcKVASLRE



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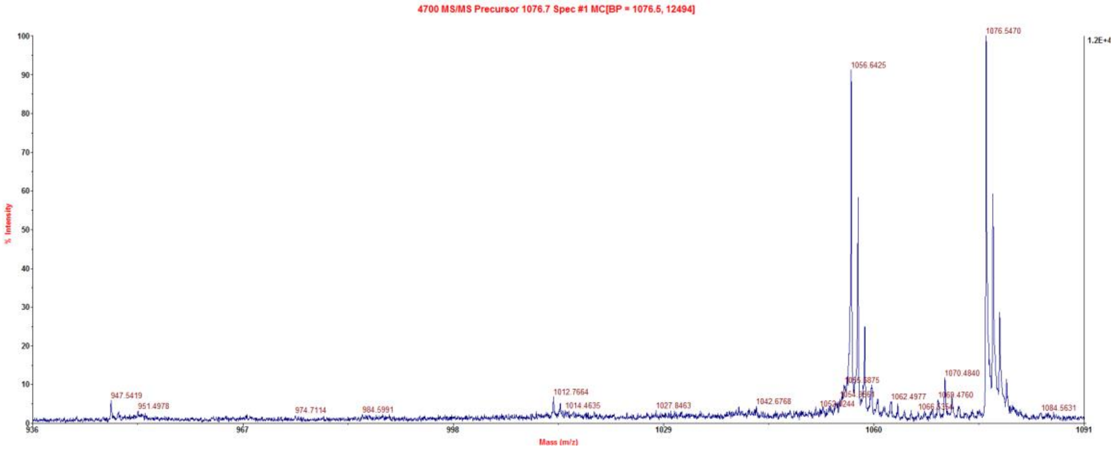
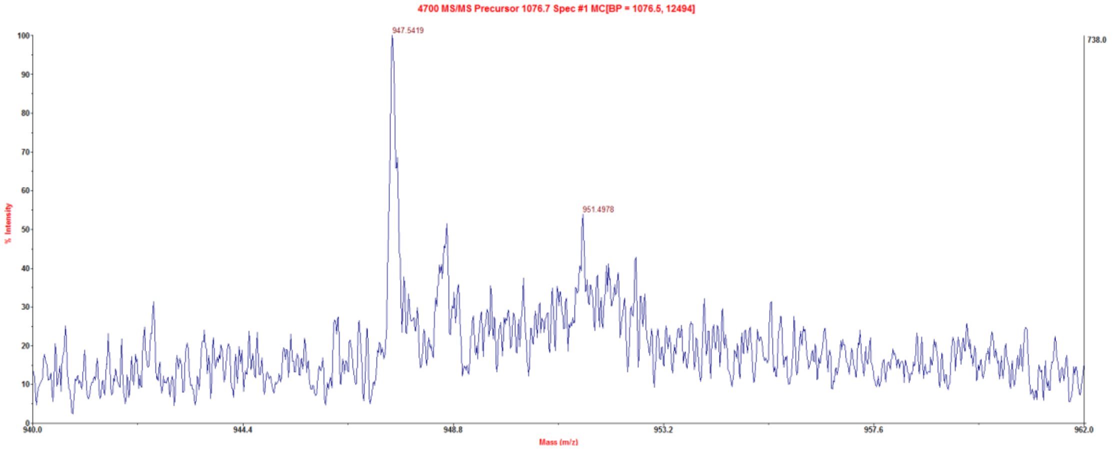
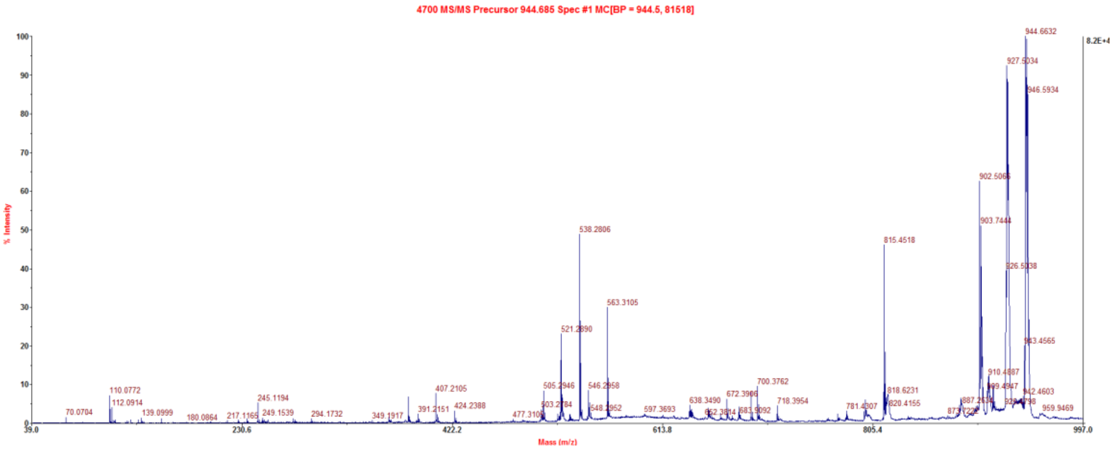


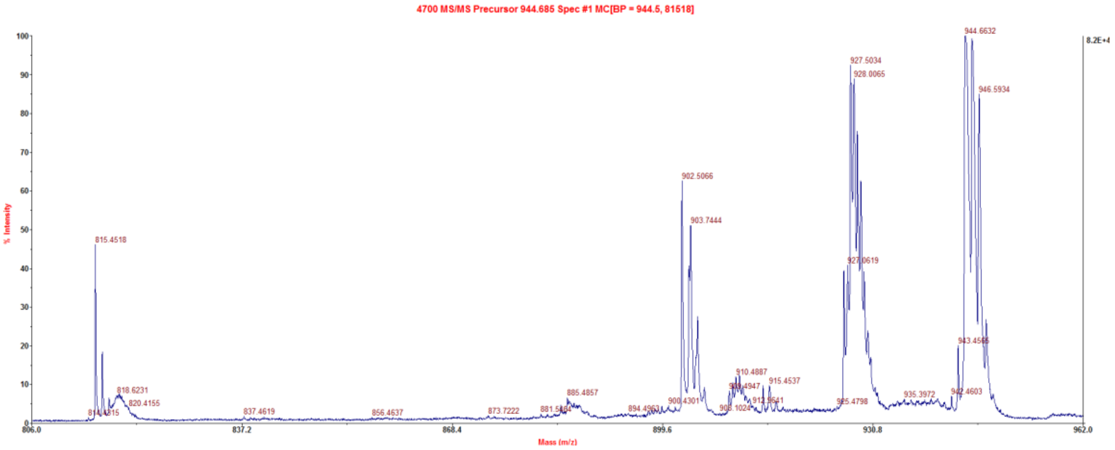
LCmcKVASLR b+18 ion



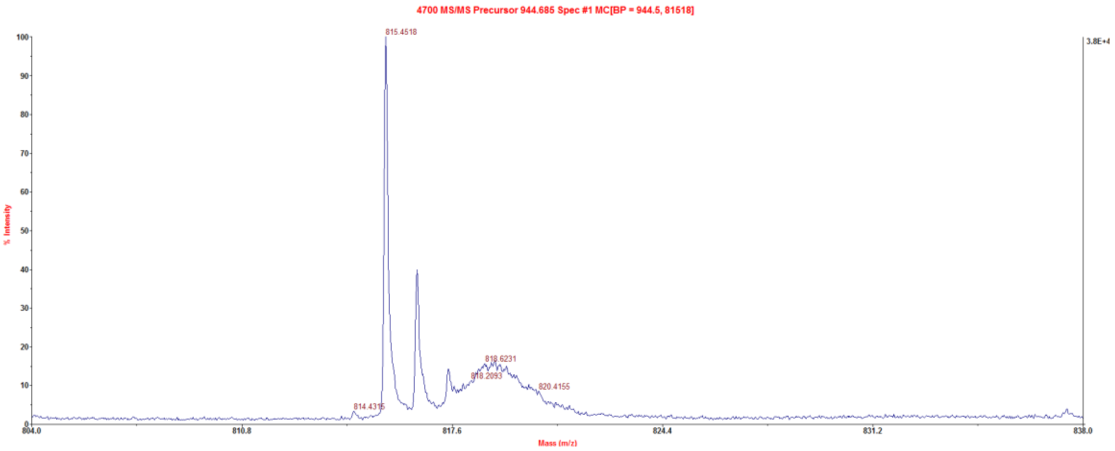
944.69 YSRRHPE



Precursor

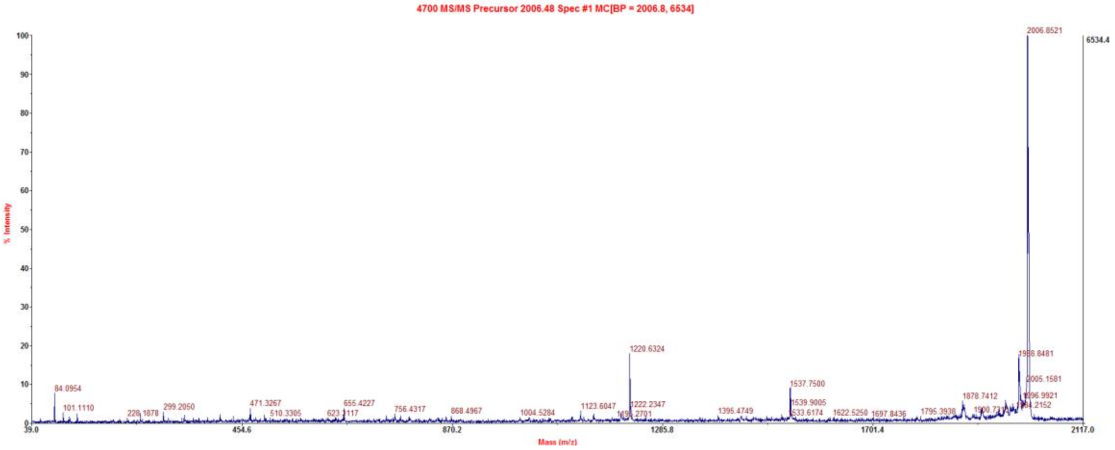


YSRRHP b+18 ion



LysN digest

2006.48 KVASLRETYGDMADCmcCmcE



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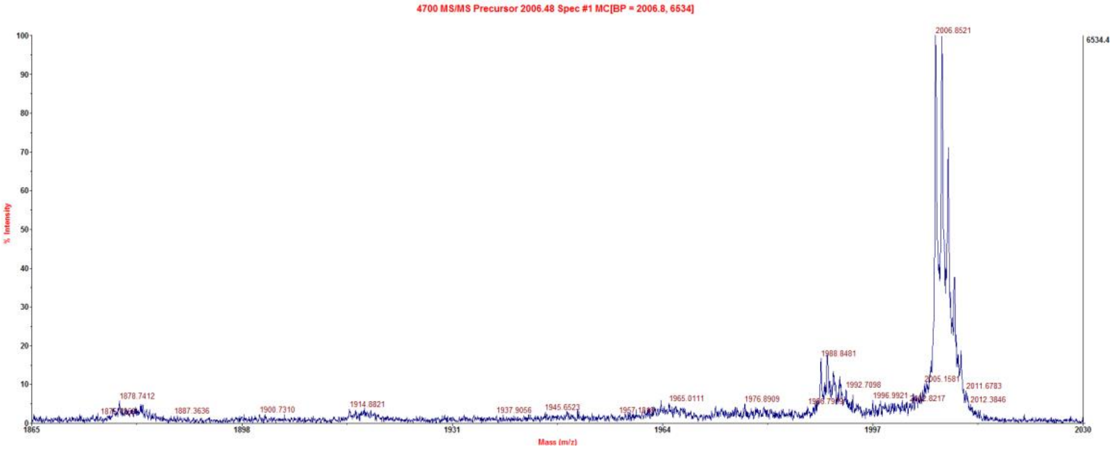
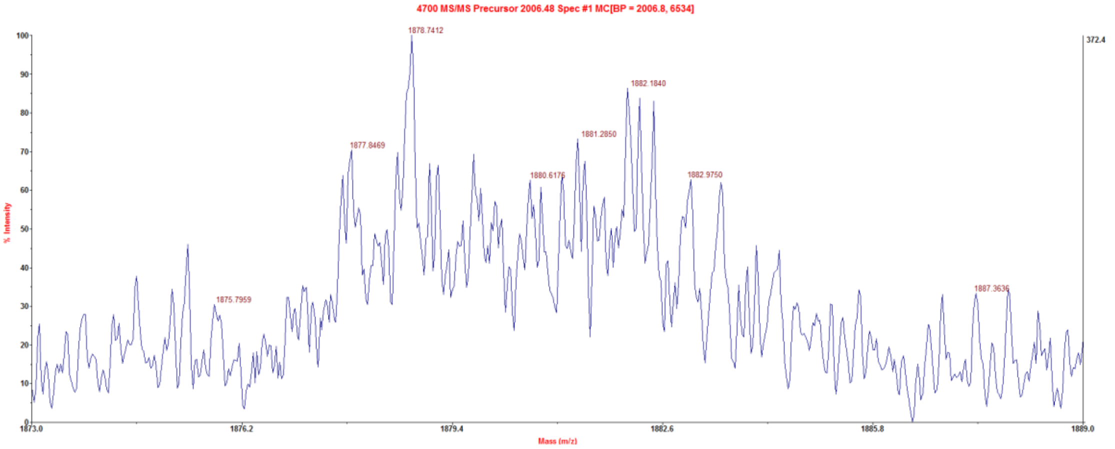
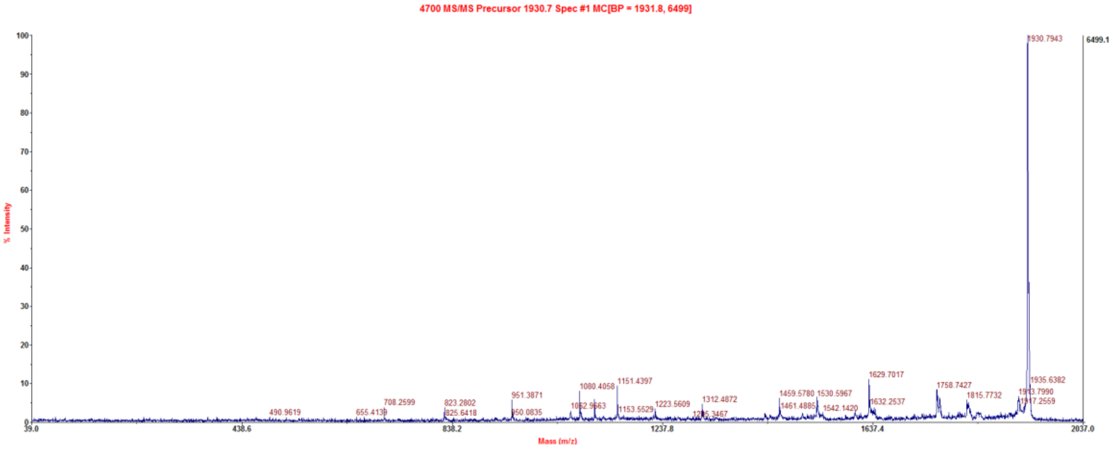


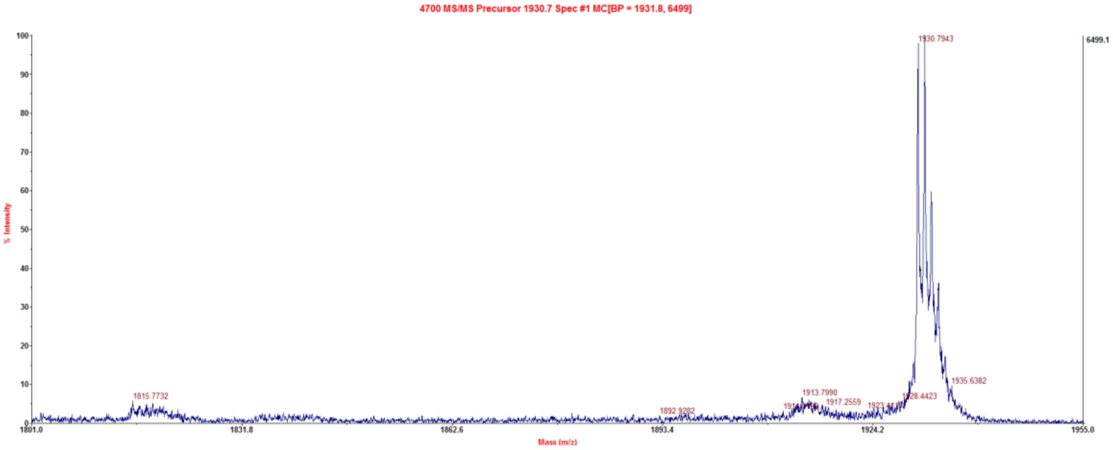
KVASLRETYGDMADCmcCmc b+18 ion



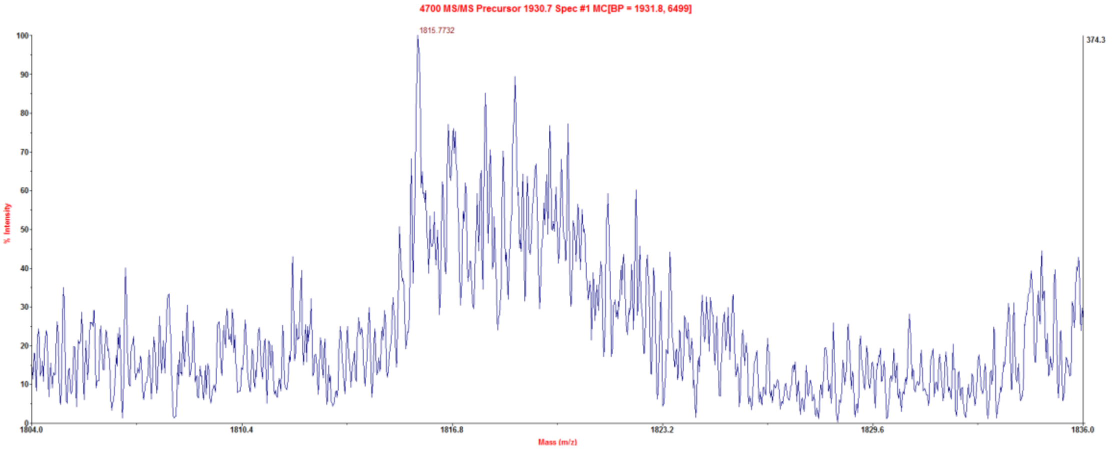
1930.70 KCmcCmcAADDKEACmcFAVEGP



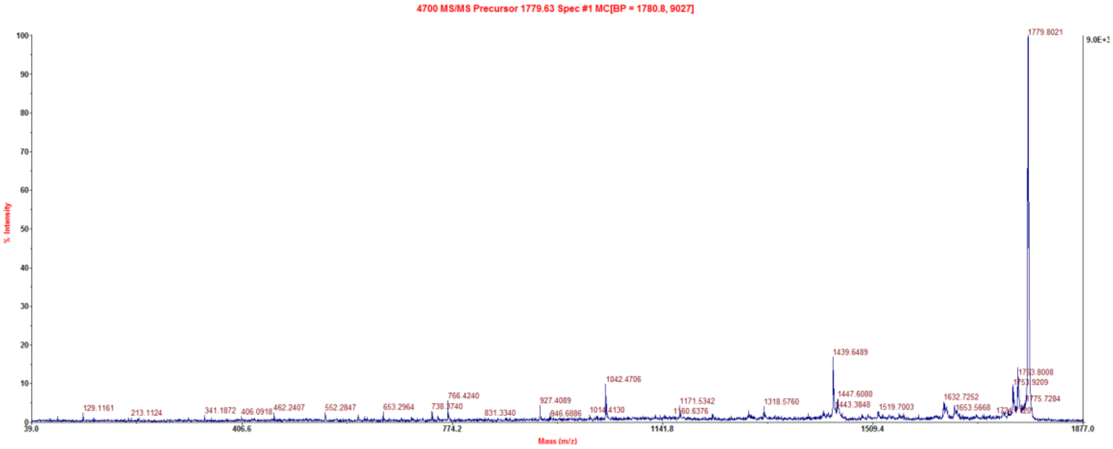
Precursor



KCmcCmcAADDKEACmcFAVEG b ion



1779.63 KPDPNTLCmcDEFKADE



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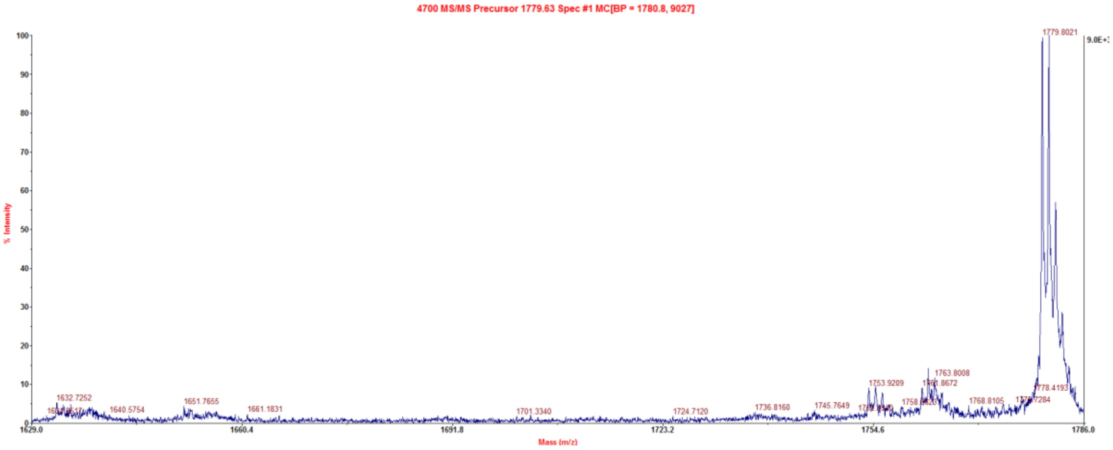
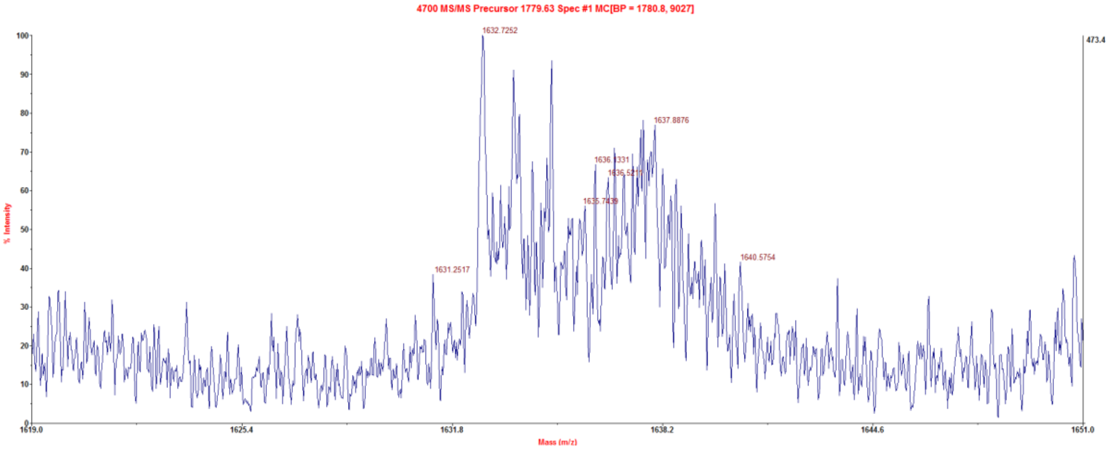
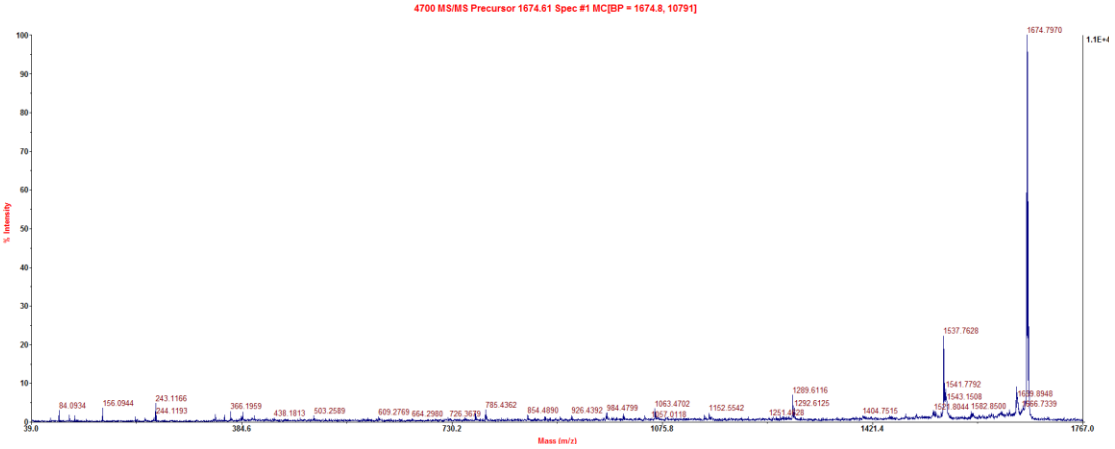


KPDPNTLCmcDEFKAD b ion



1764.61 KQEPERNECmcFLSH



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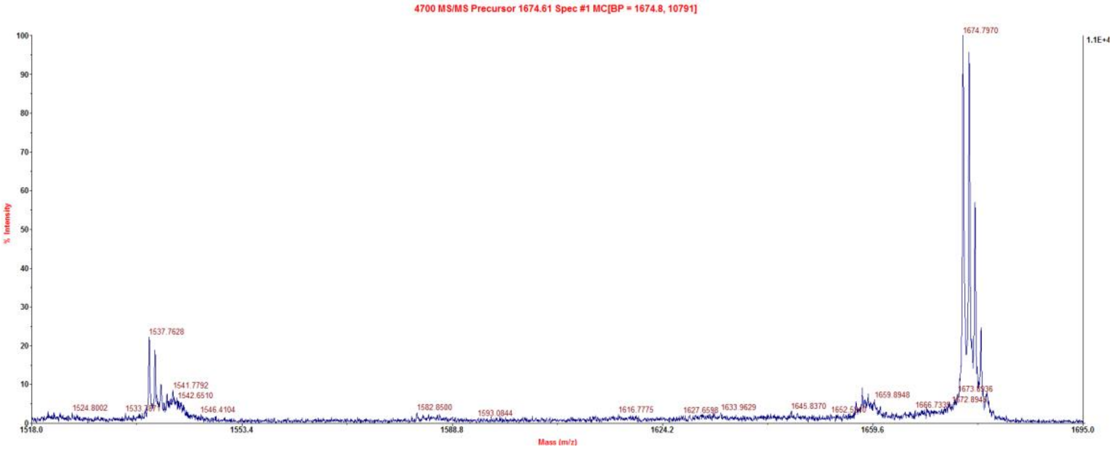
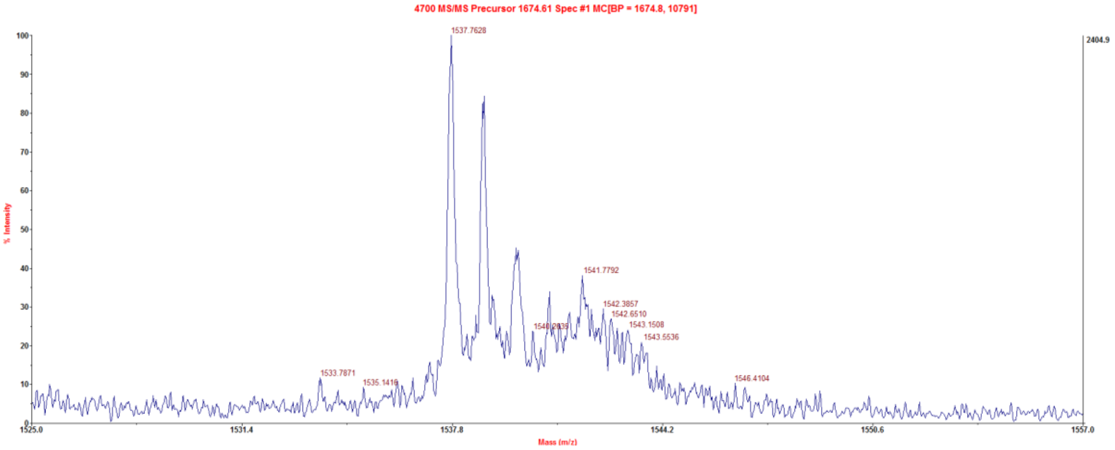
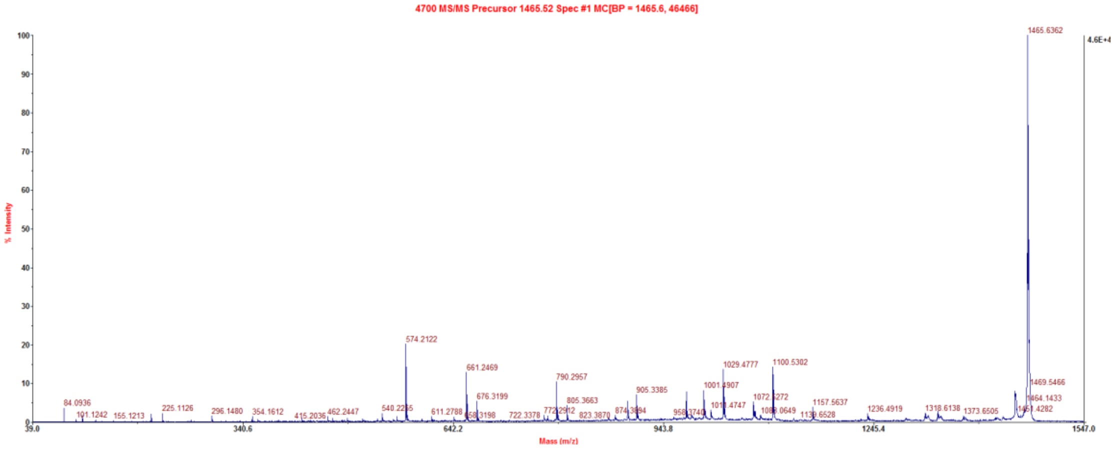


KQEPERNECmcFLS b+18 ion



1465.52 KTCmcVADESHAGCmcE



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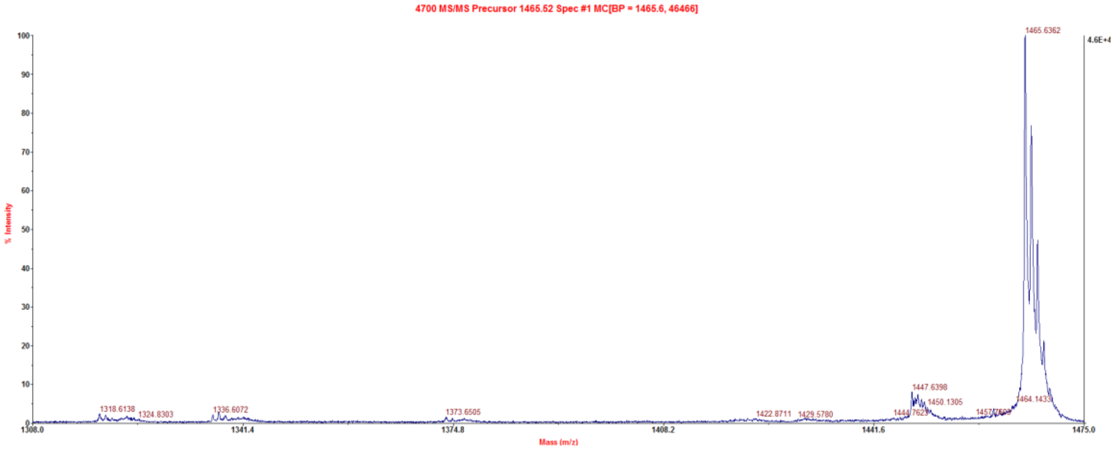
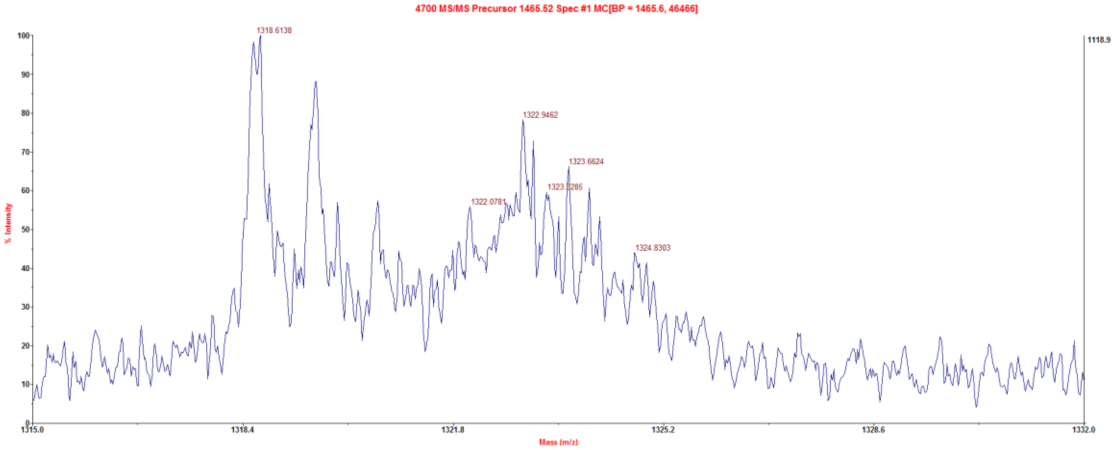
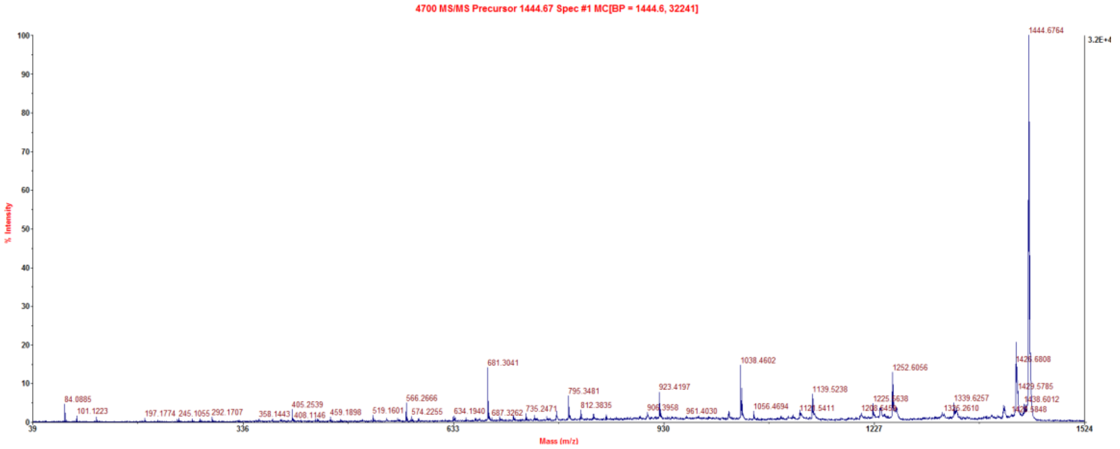


KTCmcVADESHAGCmc b ion



1443.67 KYICmcDNQDTISS



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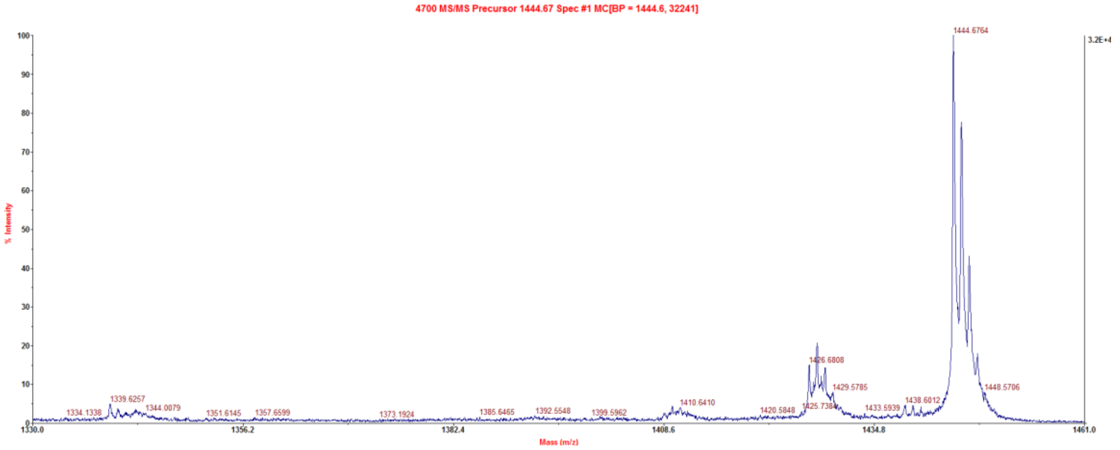
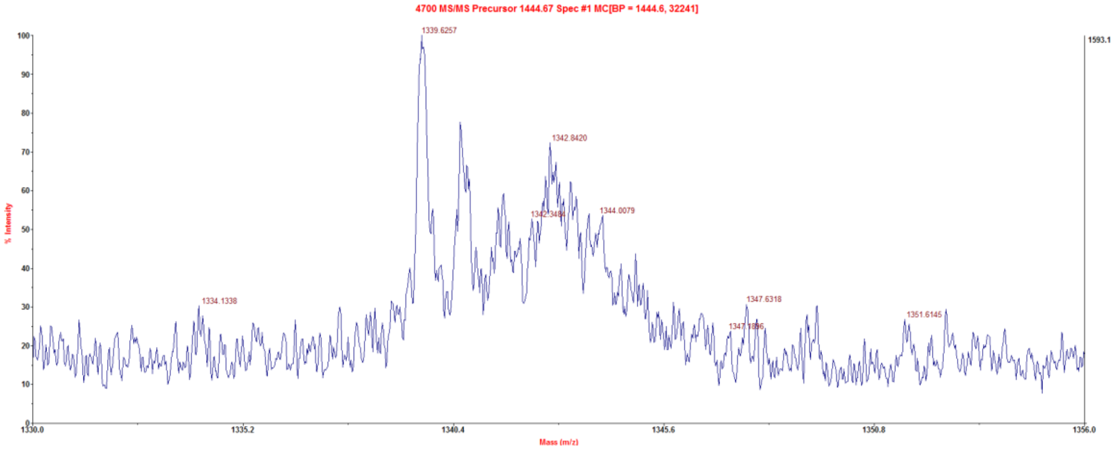
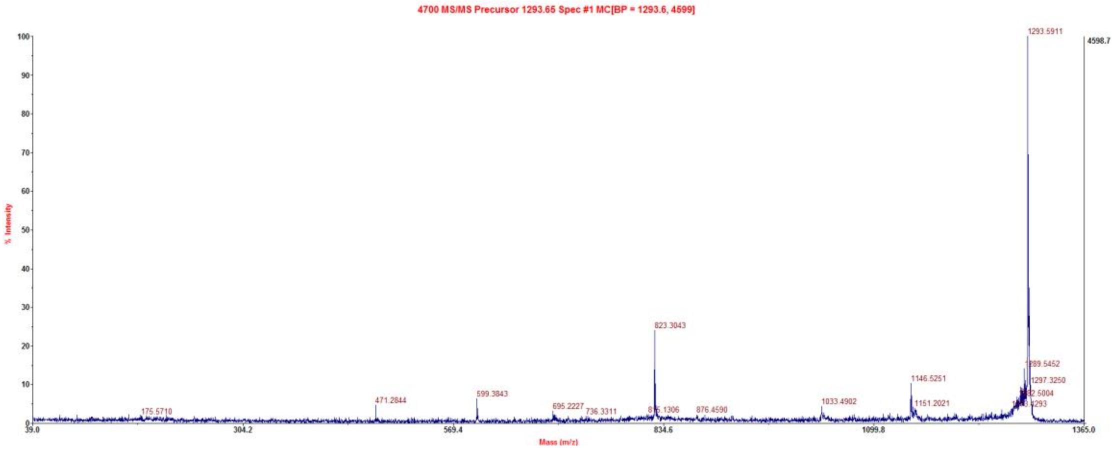


KYICmcDNQDTIS b ion



1293.65 KECmcCmcDKPLLE



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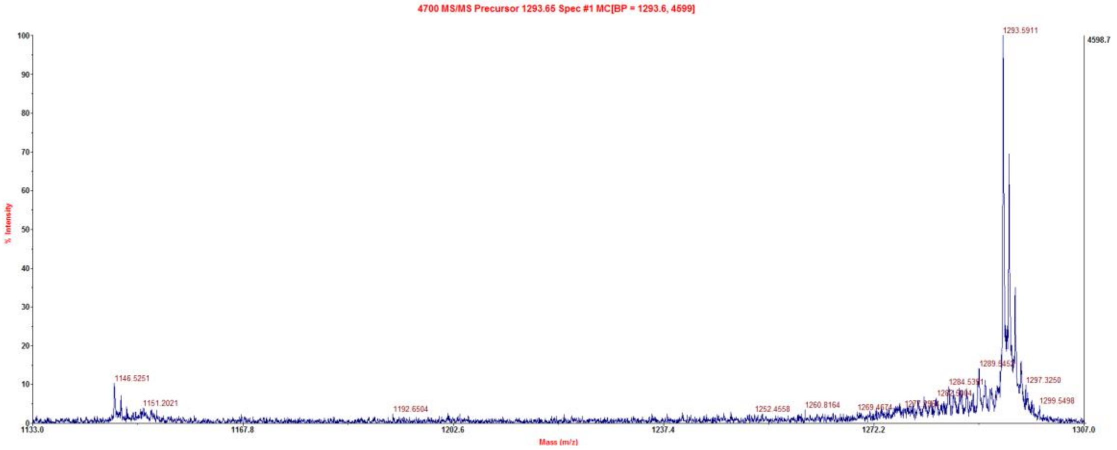
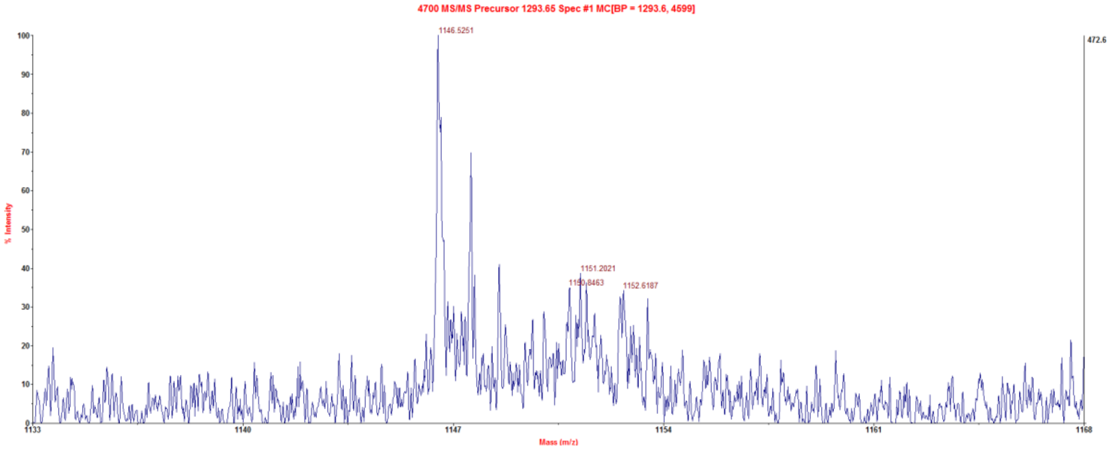
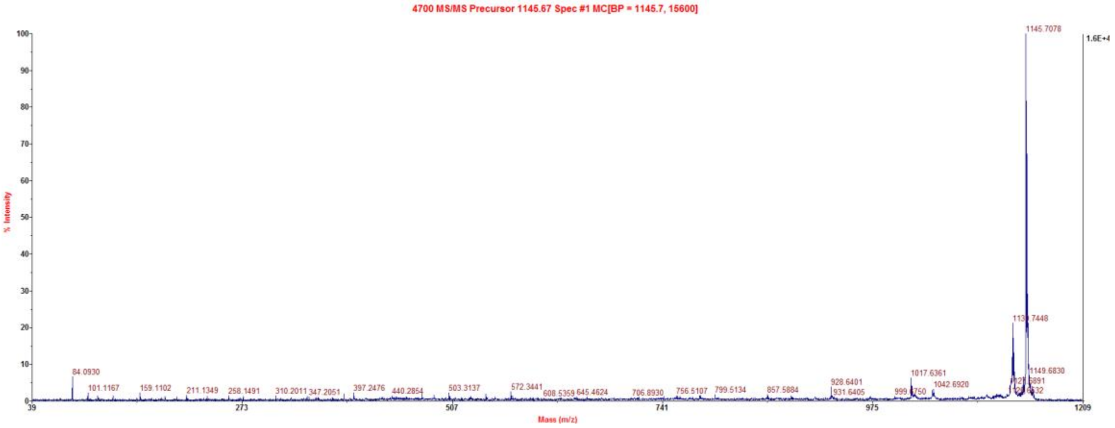


KECmcCmcDKPLL b ion



1145.67 KAWSVARLSQ



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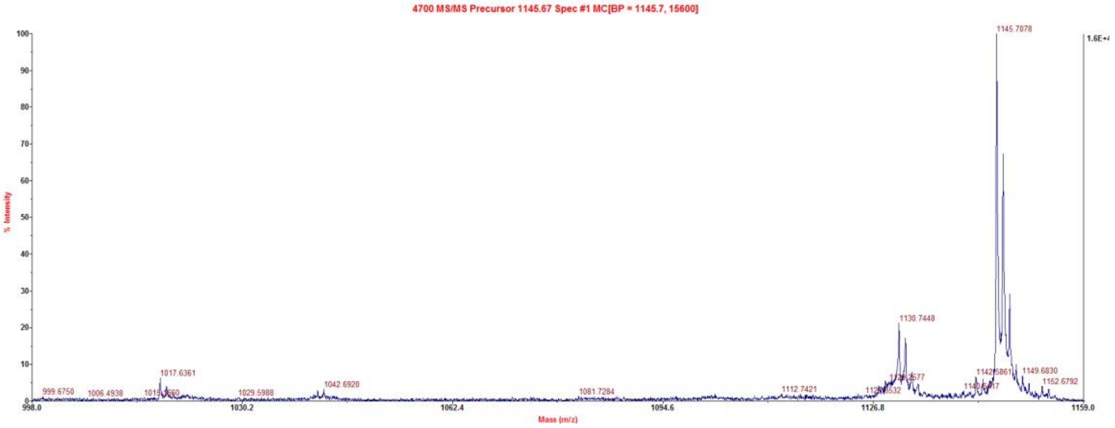
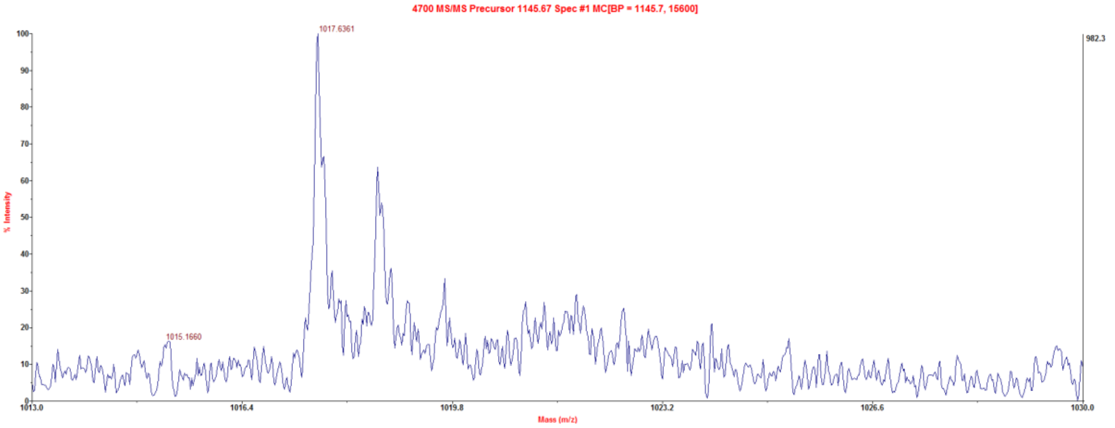
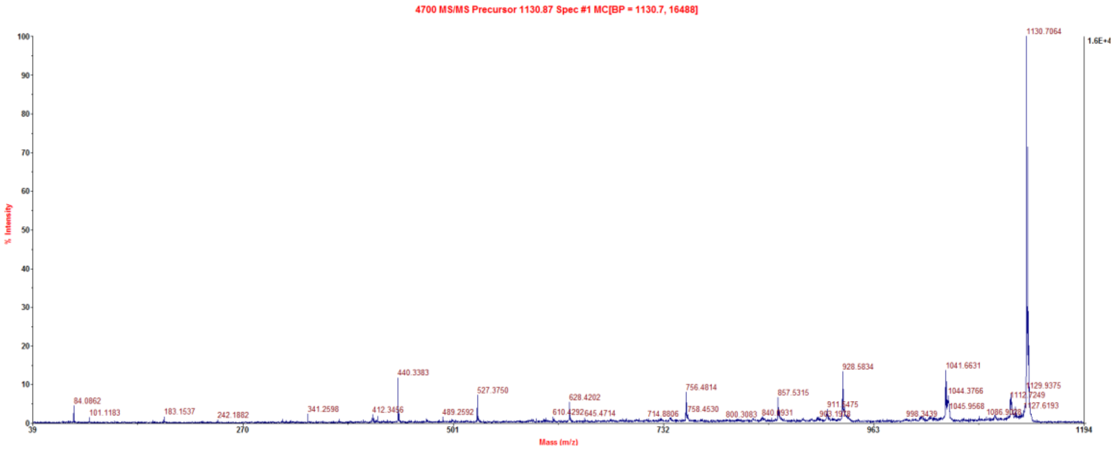


KAWSVARLS   b+18 ion



1130.87 KLVVSTQTALA



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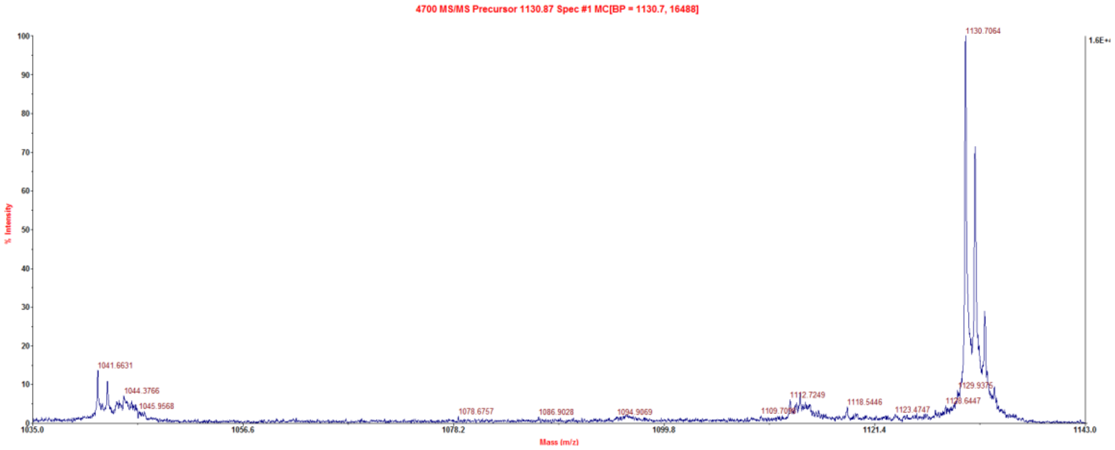
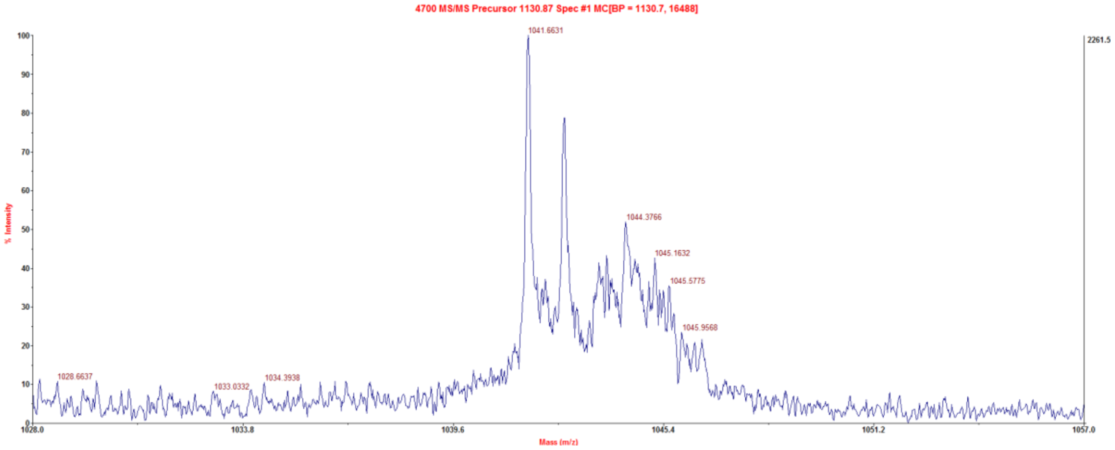
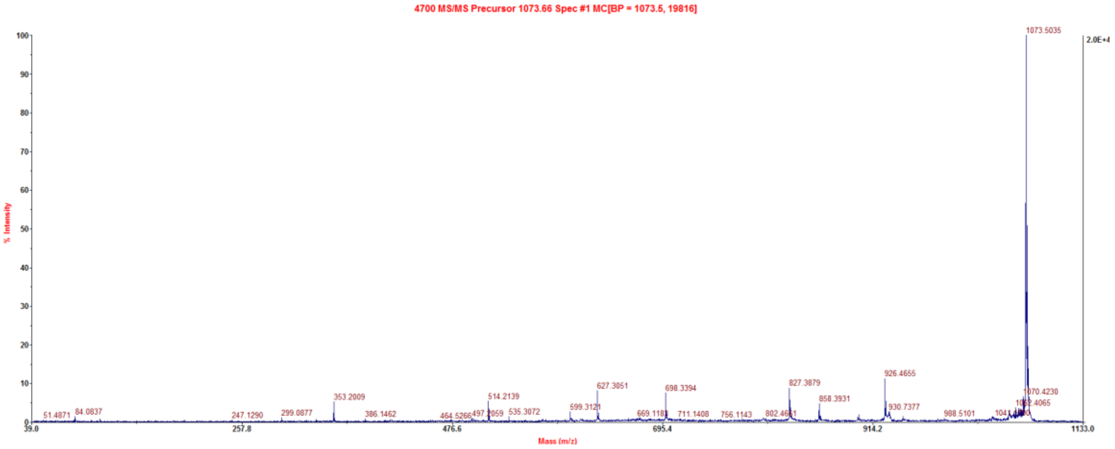


KLVVSTQTAL b ion



1073.66 KSHCmcIAEVE



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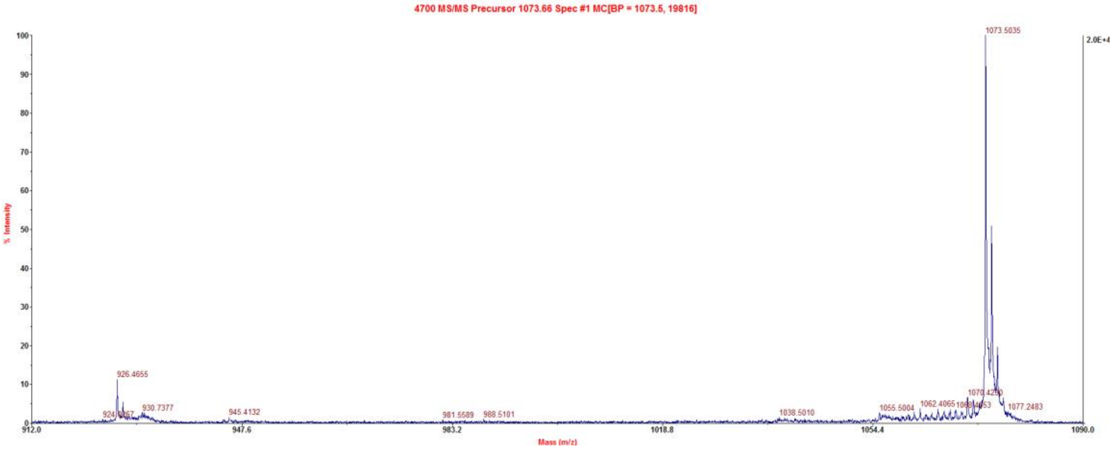
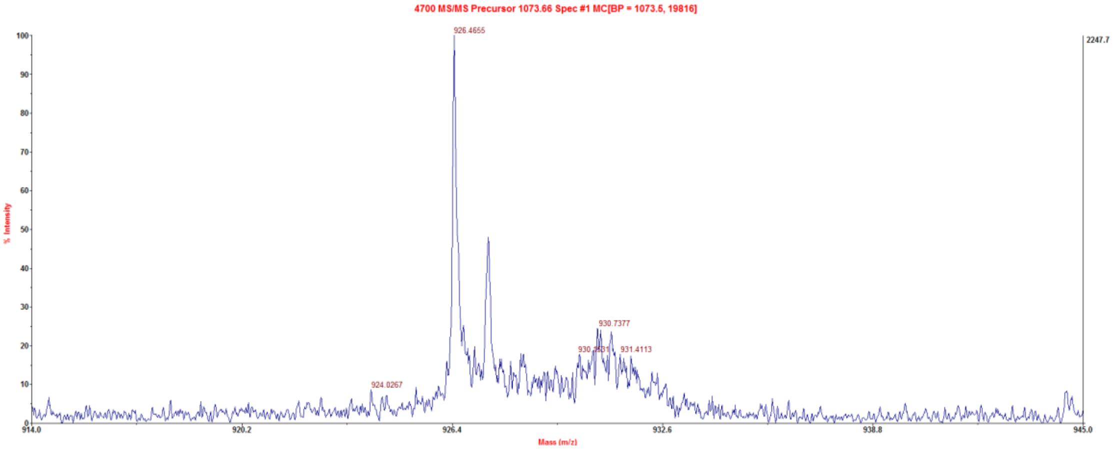
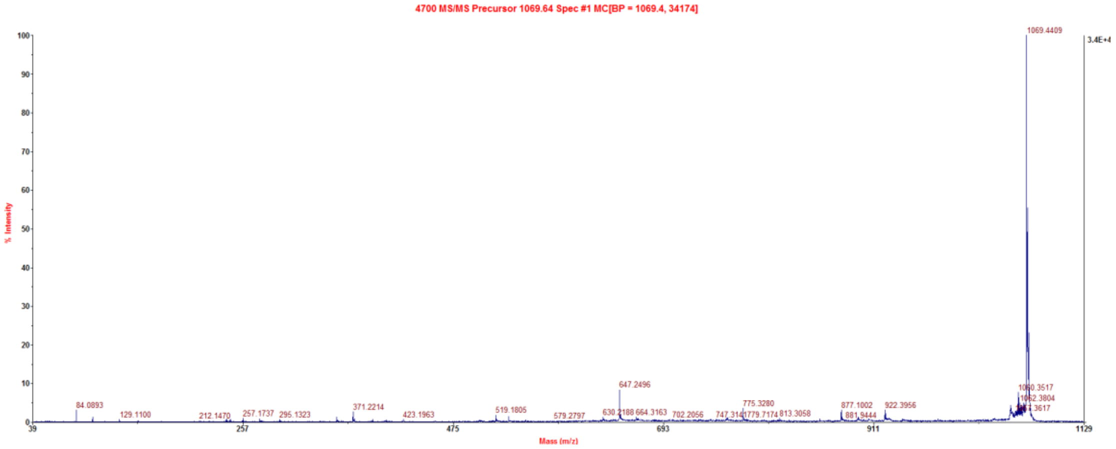


KSHCmcIAEV b ion



1069.64 KQNCmcDQFE



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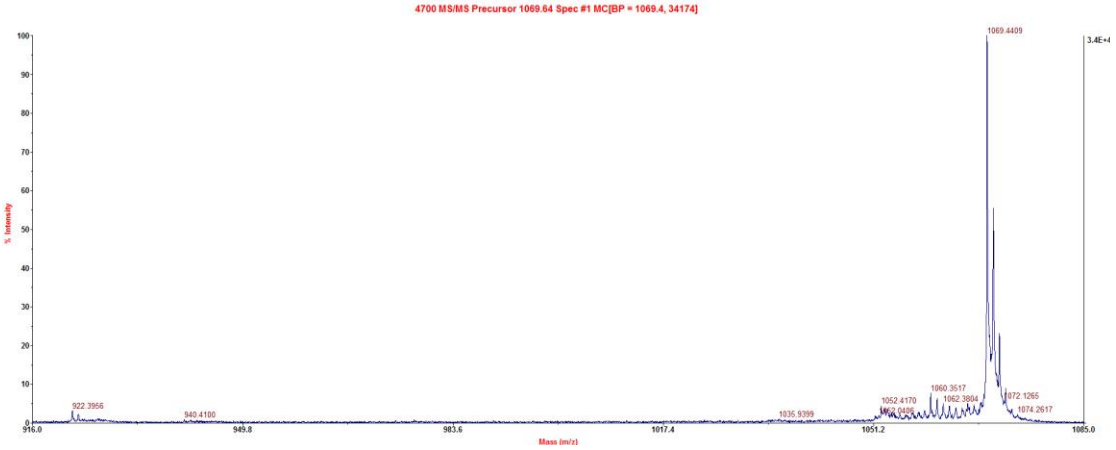
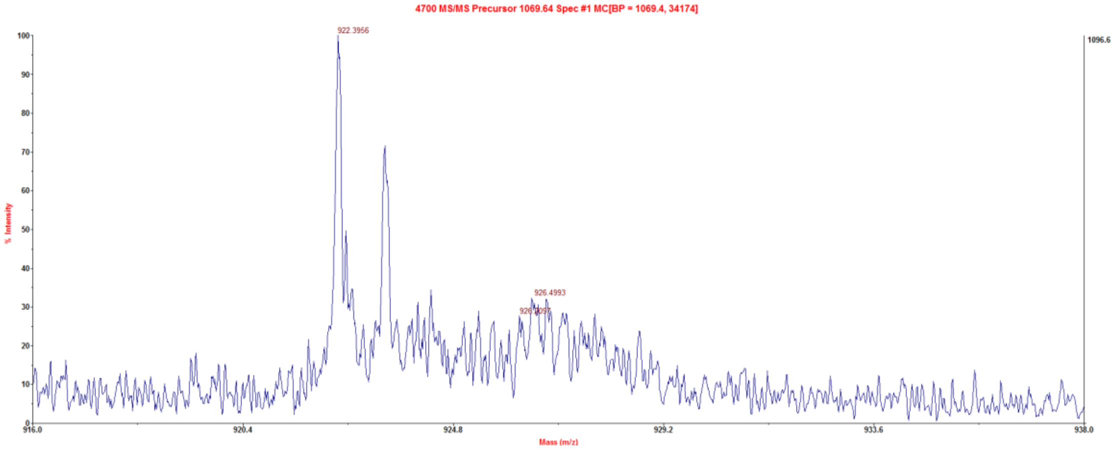
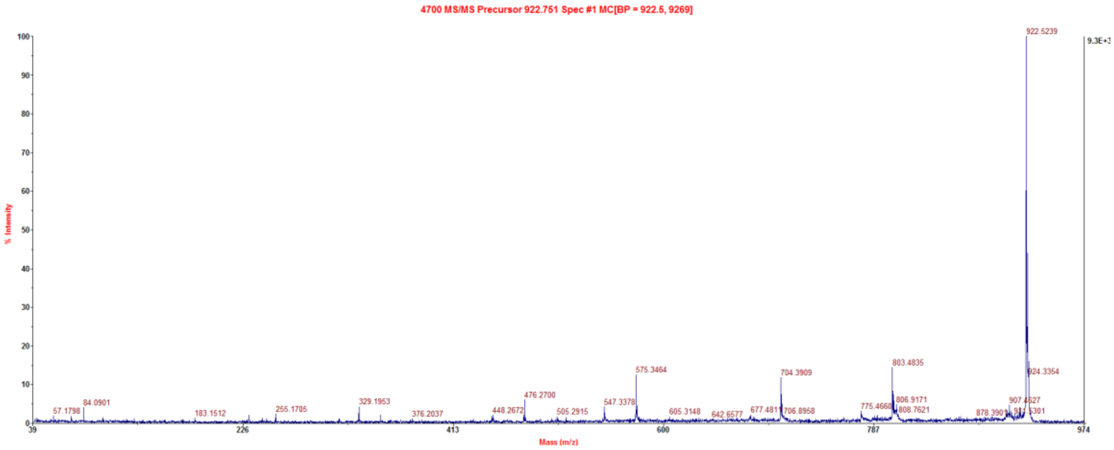


KQNCmcDQF b ion



922.75 KAEFVEVT



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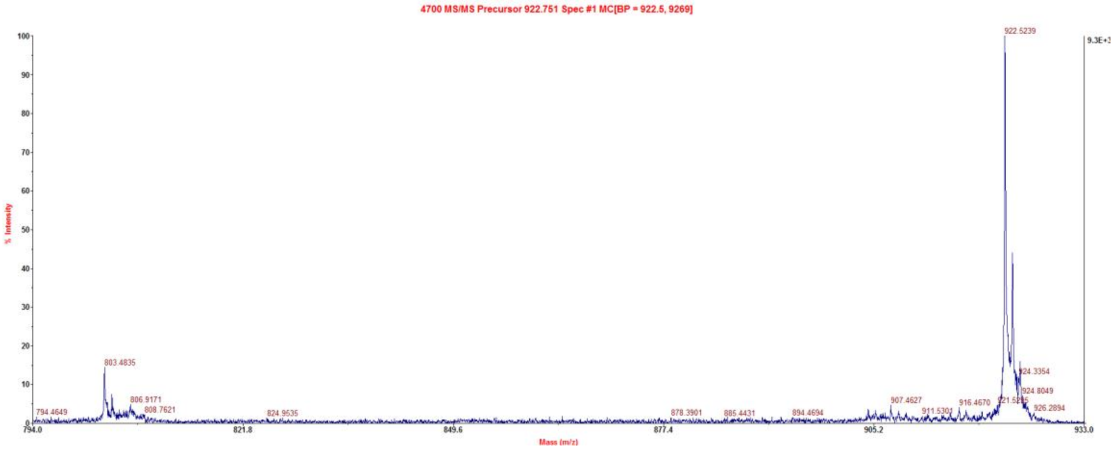
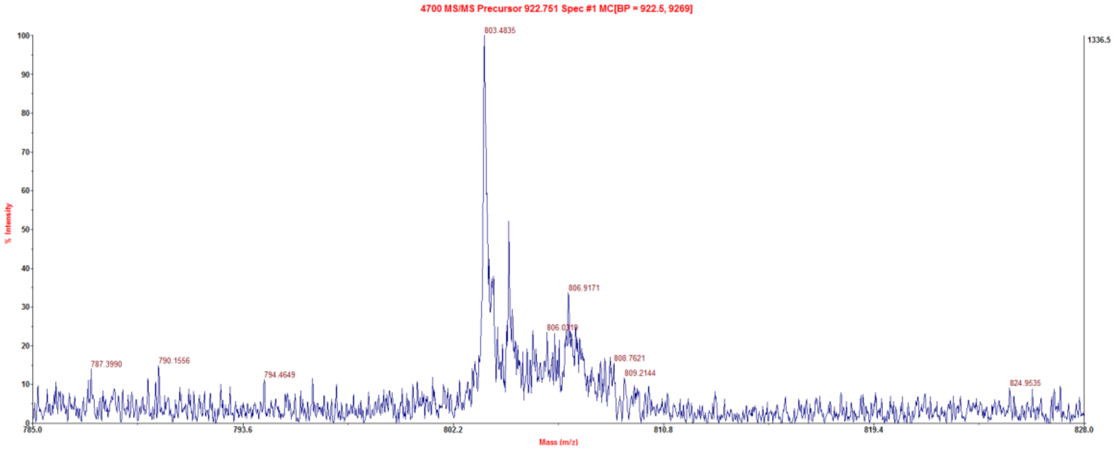
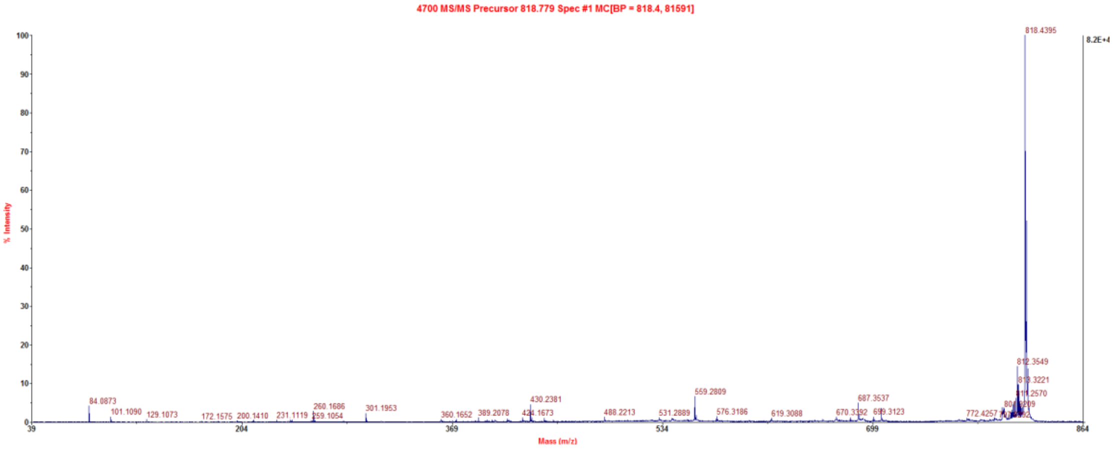


KAEFVEV b ion



818.78 KATEEQL



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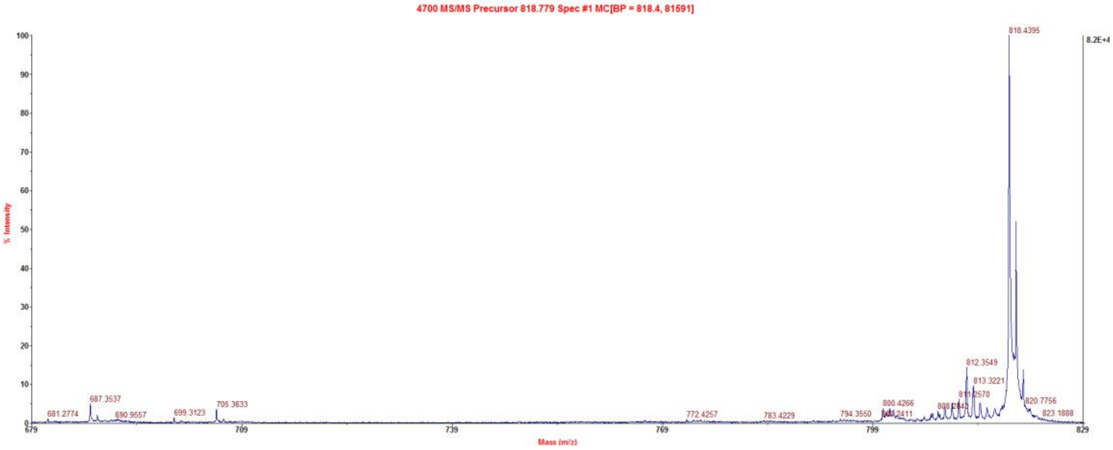
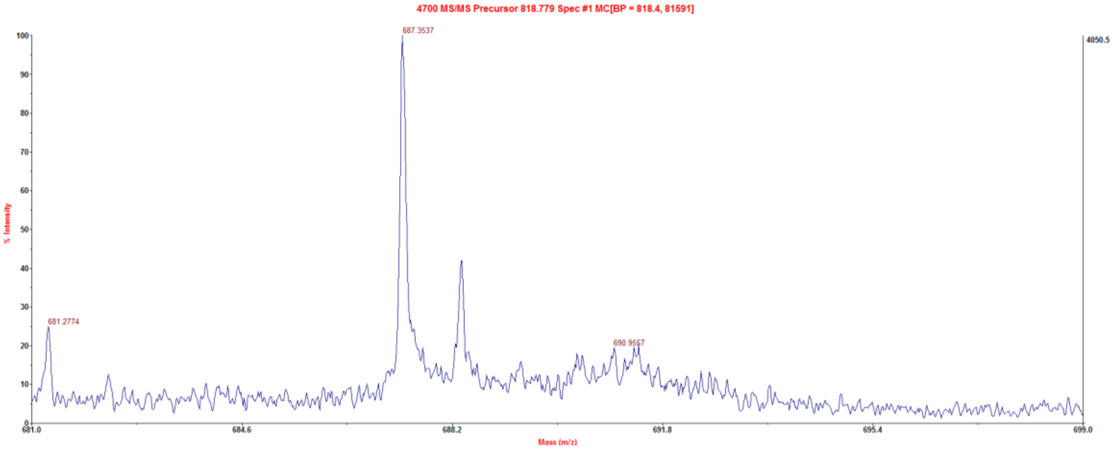
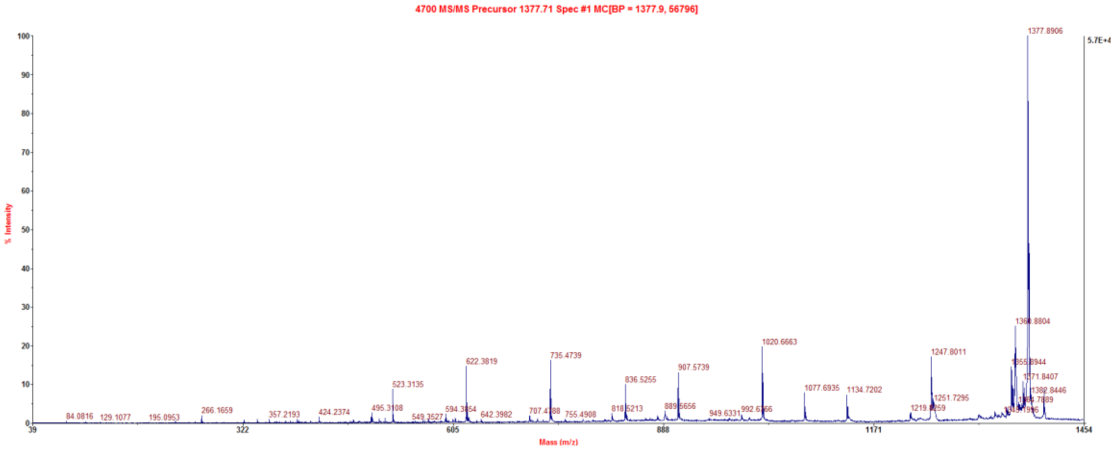


KATEEQ b ion



Spectra of peptides with C-terminal amide

KHGTVVLTALGGIL-NH_2 1377.71



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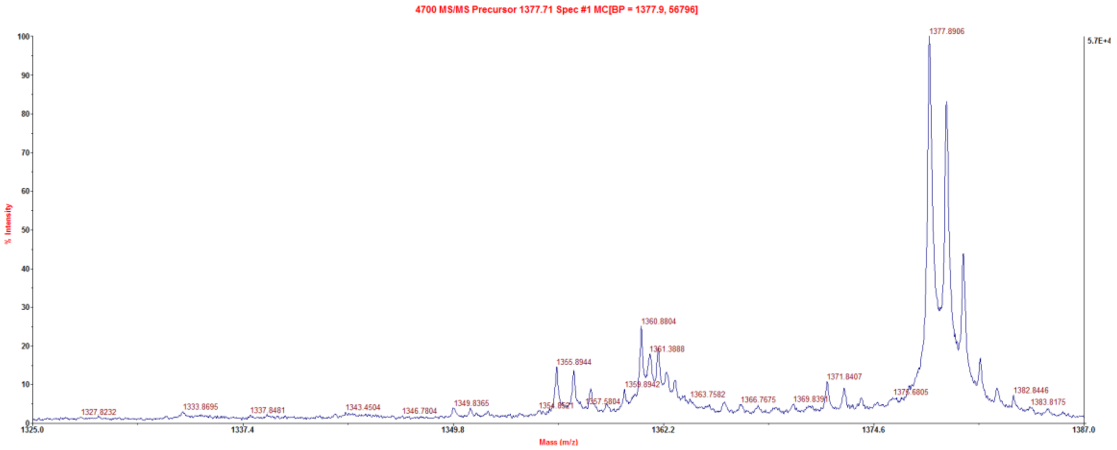
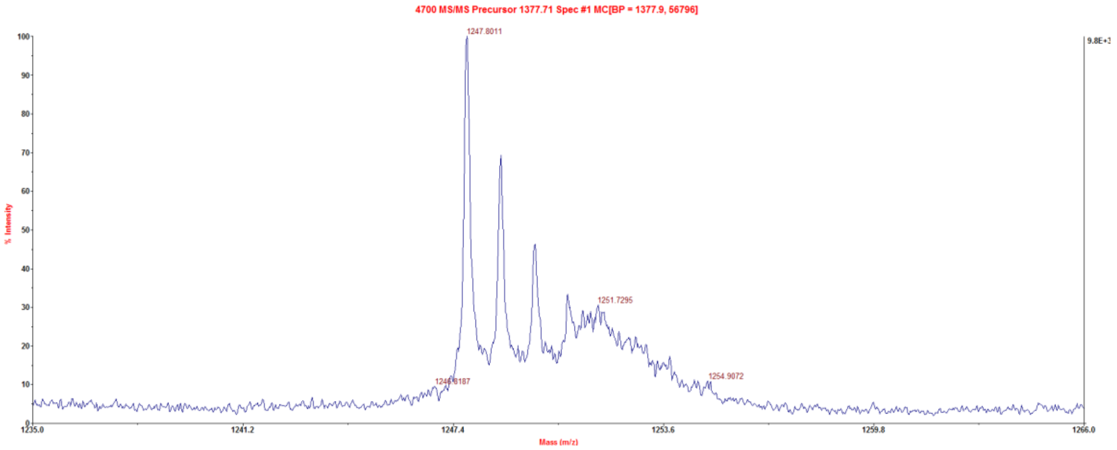
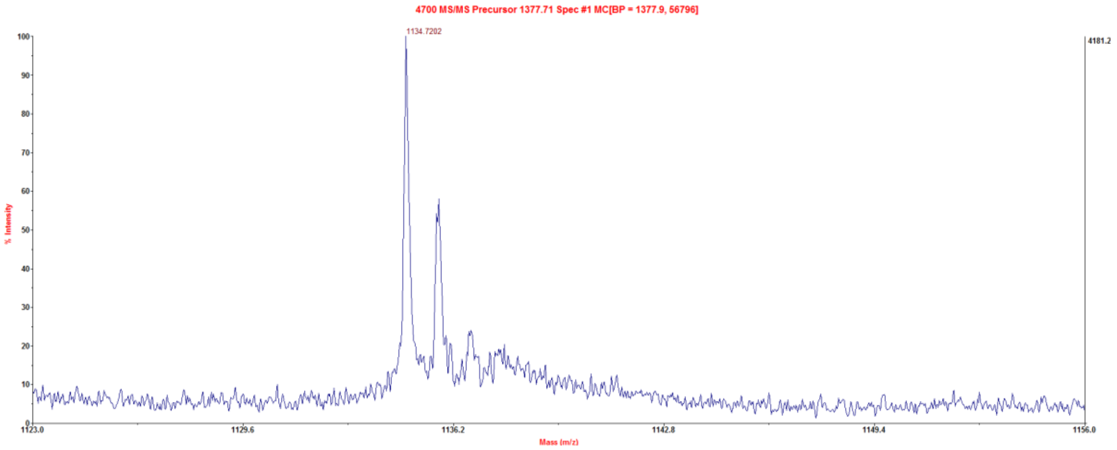


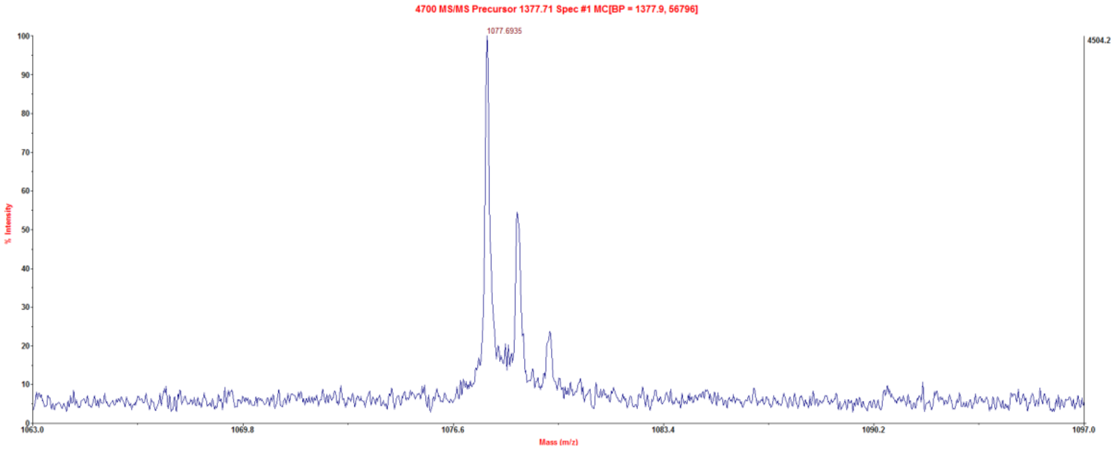
KHGTVVLTALGGI 1247.80 b-ion



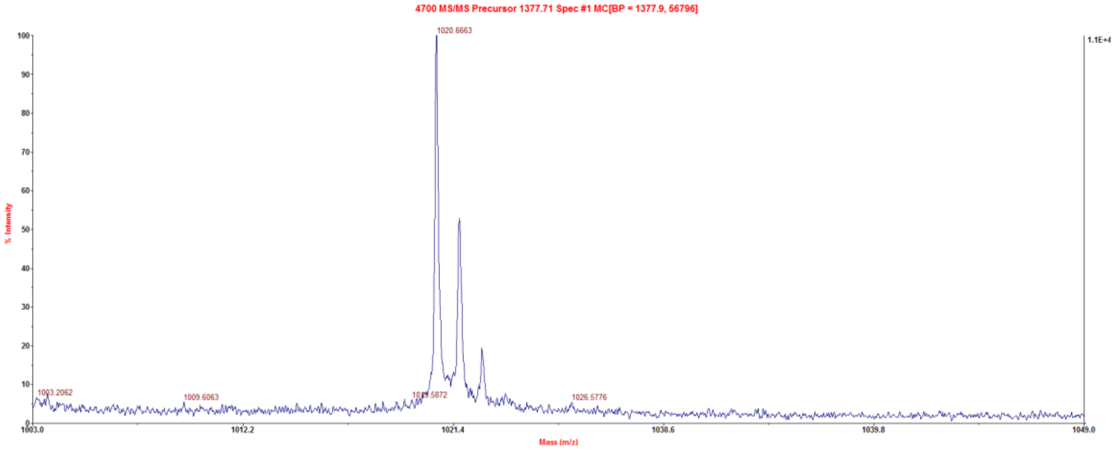
KHGTVVLTALGG   1134.72



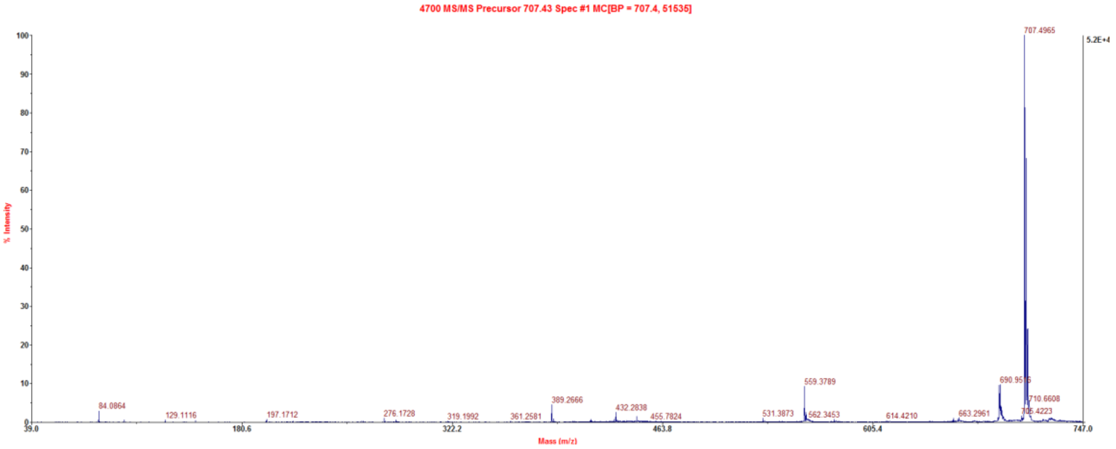
KHGTVVLTALG 1077.69



KHGTVVLTAL 1020.663



Eledoisin related peptide KFIGLM-NH_2 707.43



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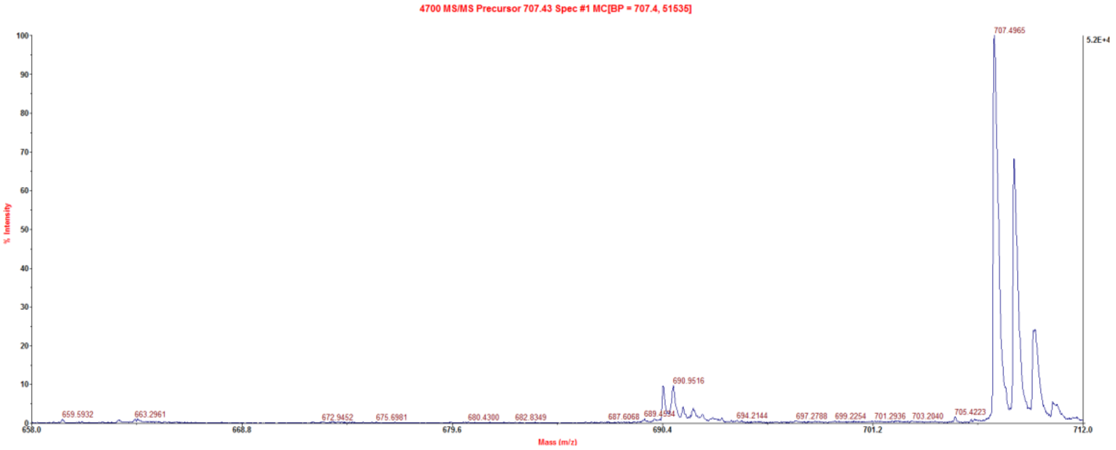
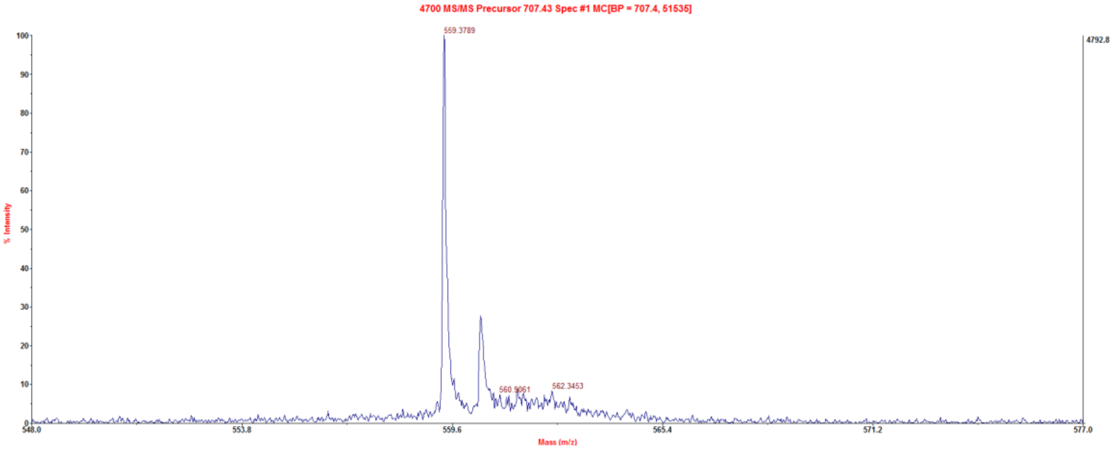
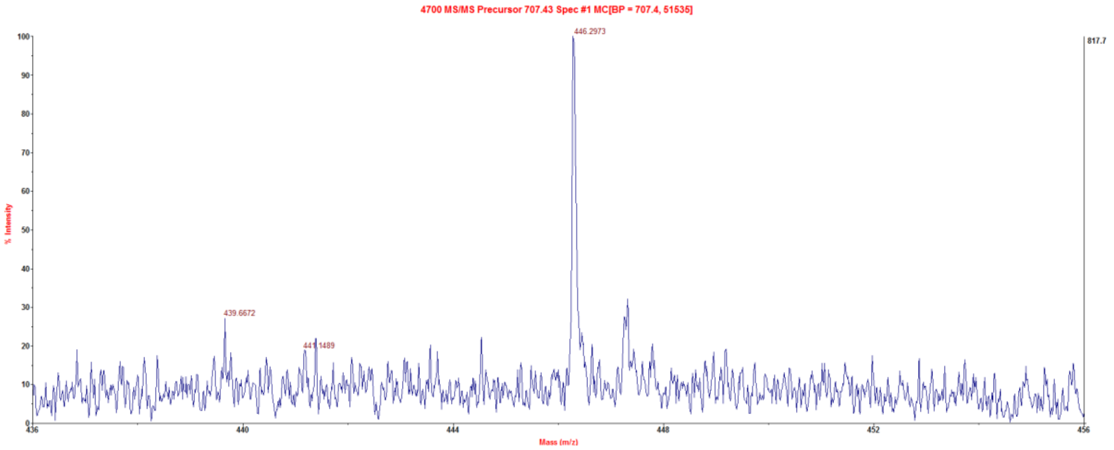


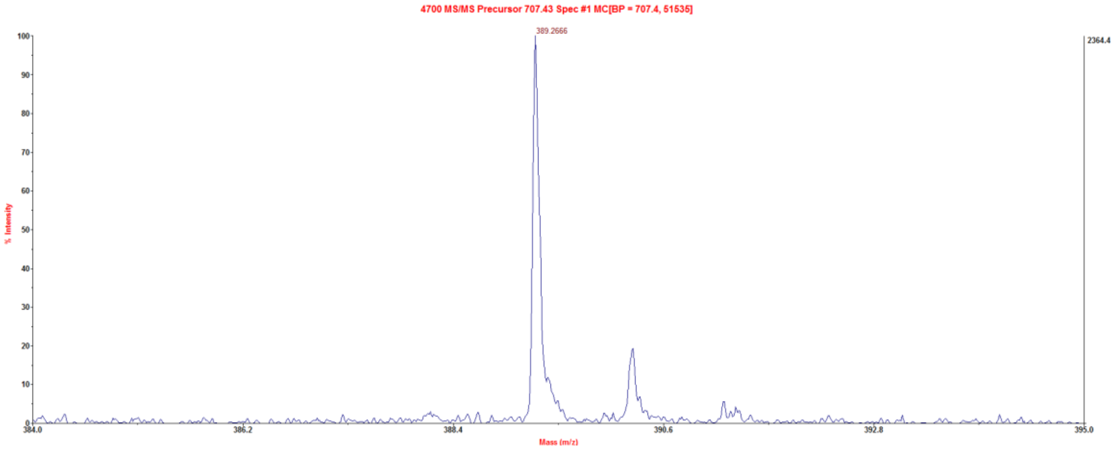
KFIGL 559.37 b-ion



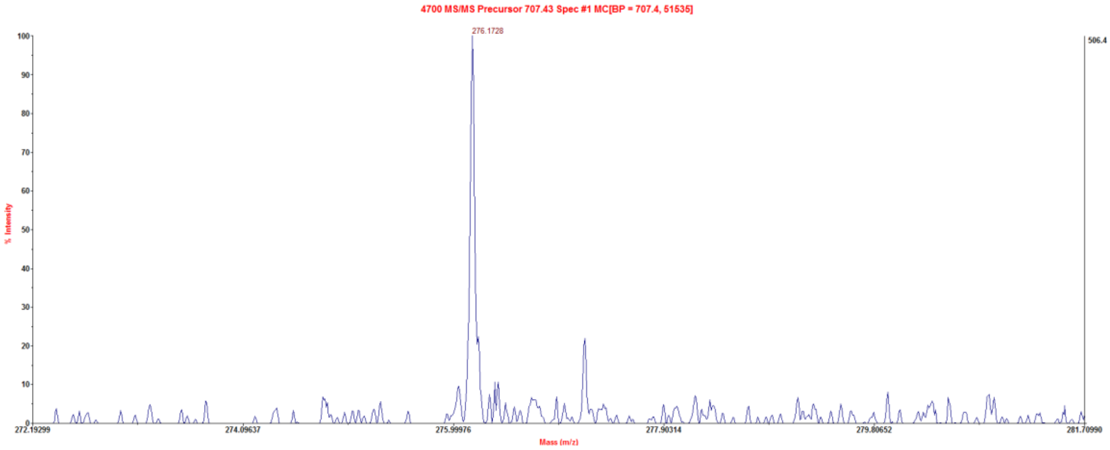
KFIG 446.30



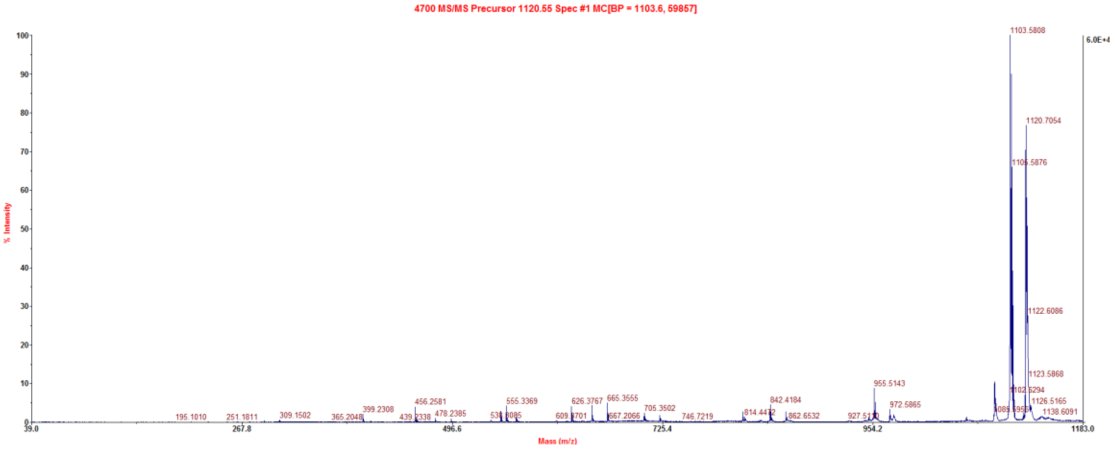
KFI 389.27



KF 276.37



Neuromedin C GNHWAVGHLM-NH$_2$ 1120.55



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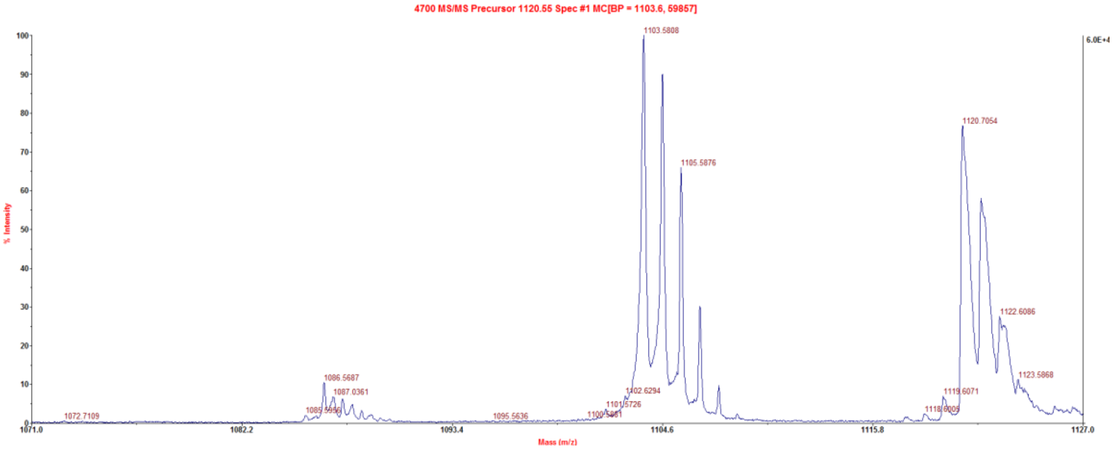
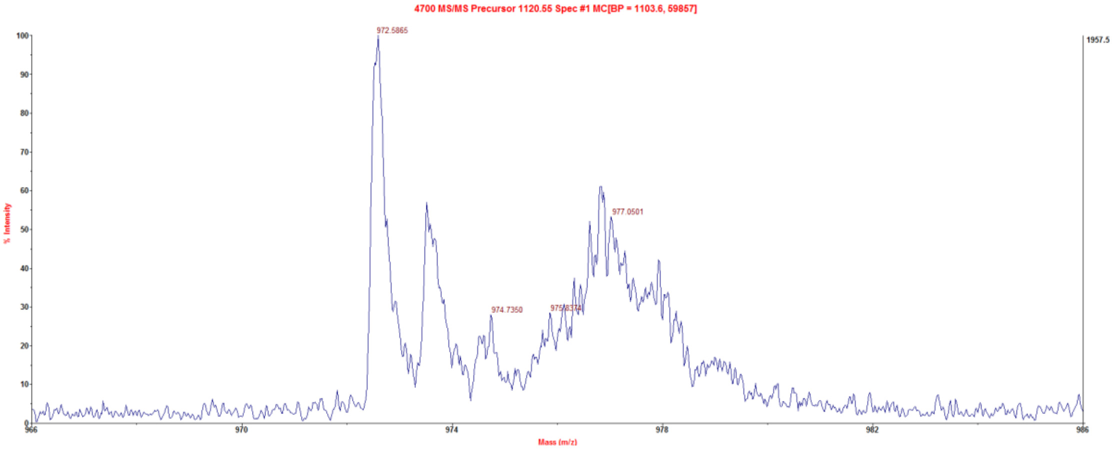
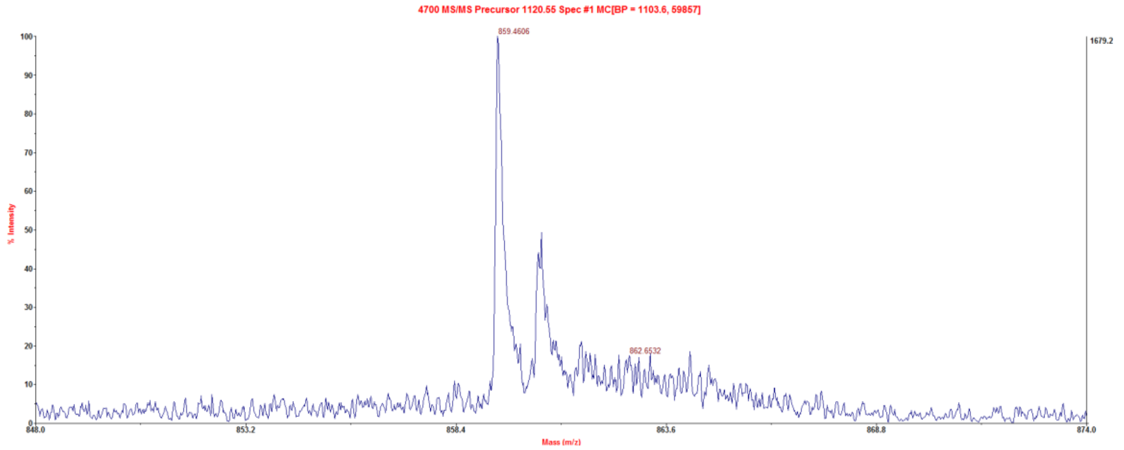


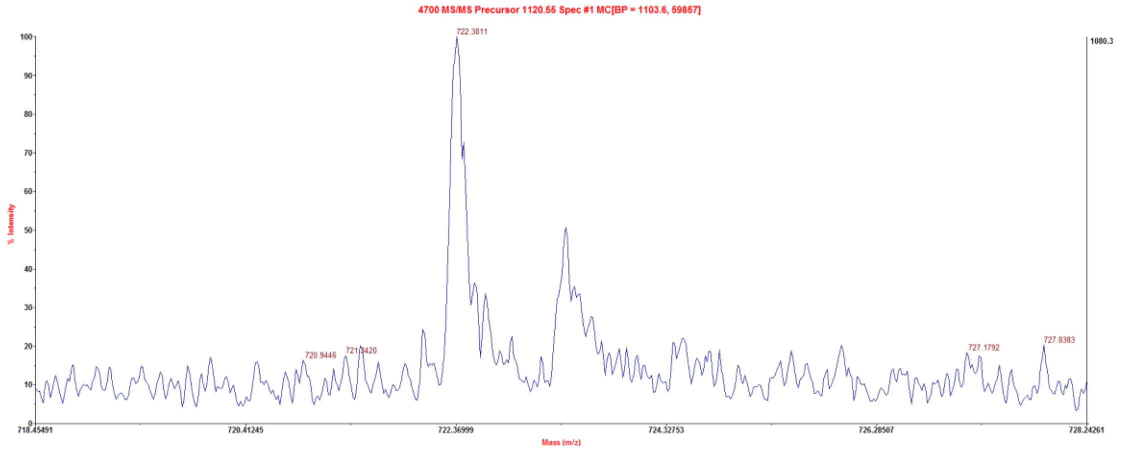
GNHWAVGHL 972.59 b-ion



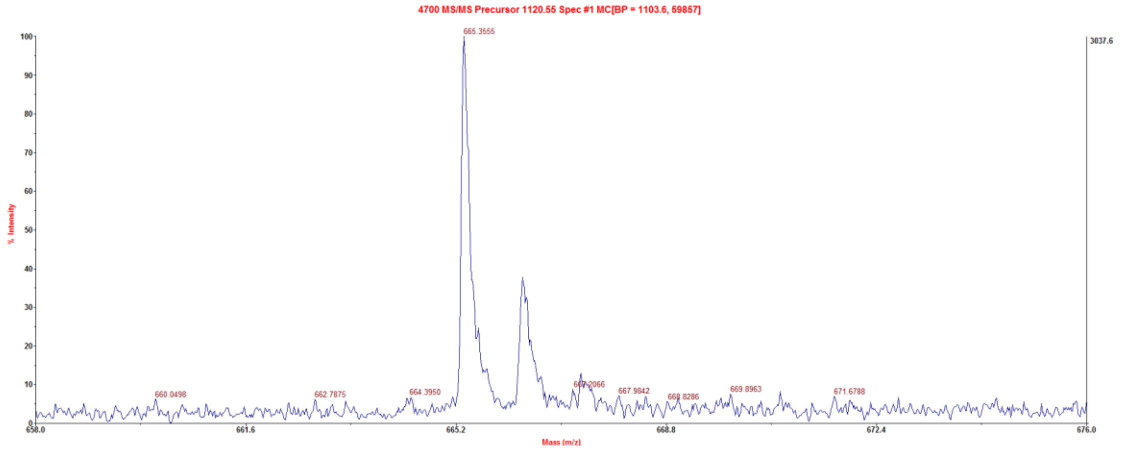
GNHWAVGH 859.46



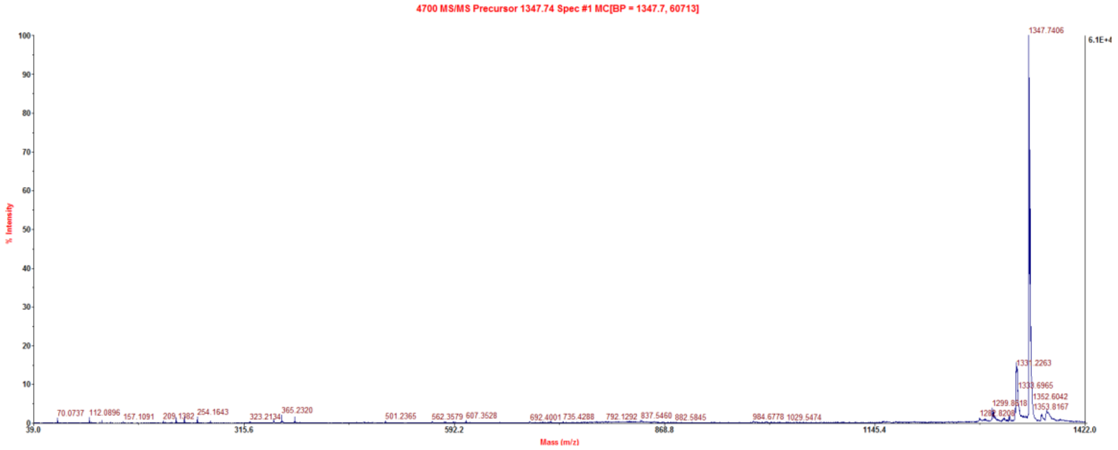
GNHWAVG 722.38



GNHWAV 665.36



Substance P RPKPQQFFGLM-NH₂ 1347.74



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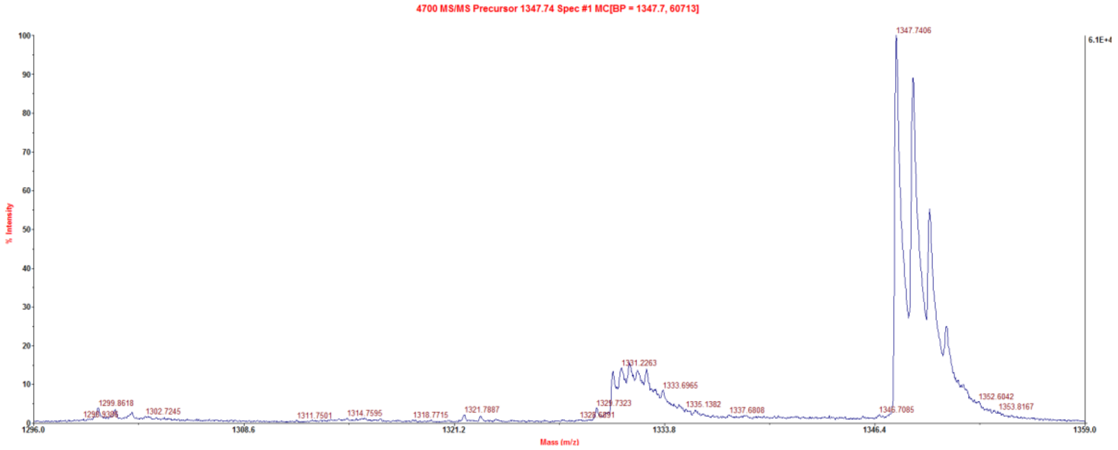
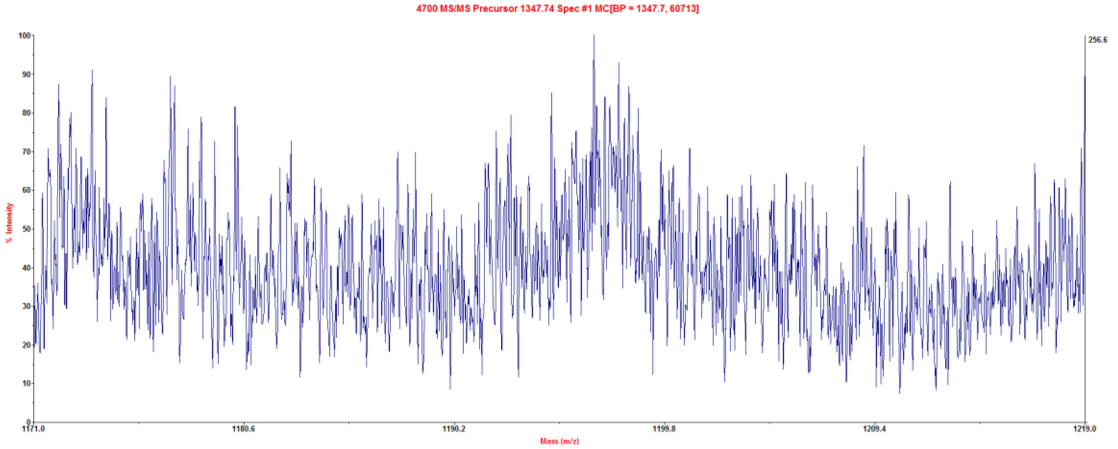
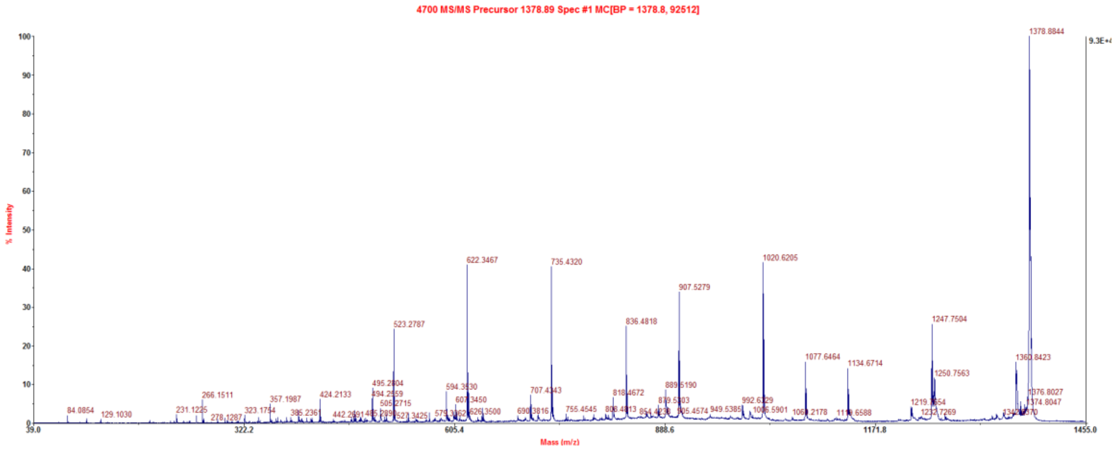


RPKPQQFFGL 1199 x



KHGTVVLTALGGIL 1378.89



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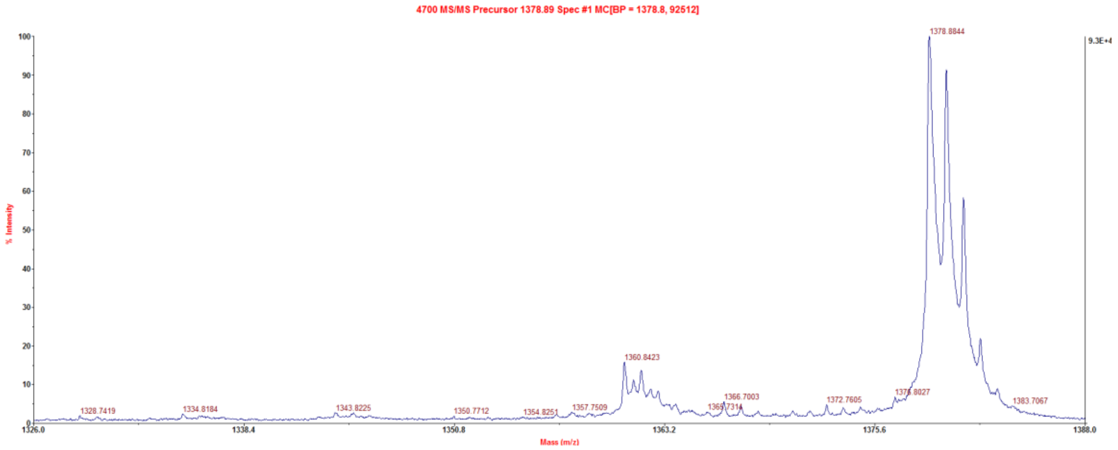

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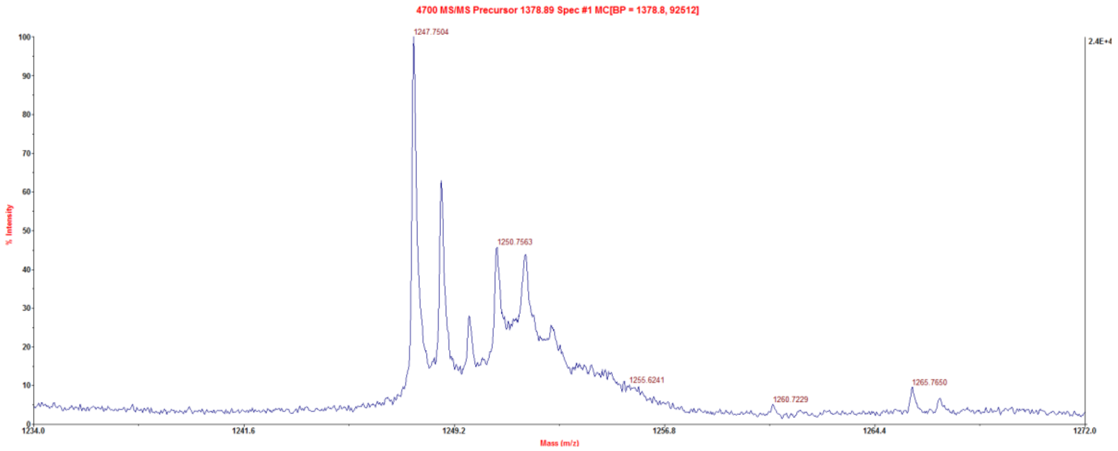

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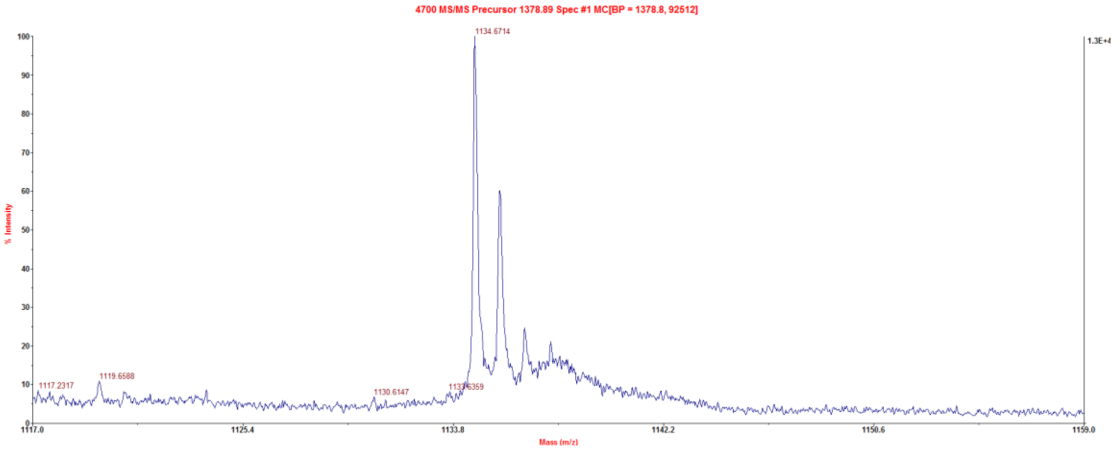
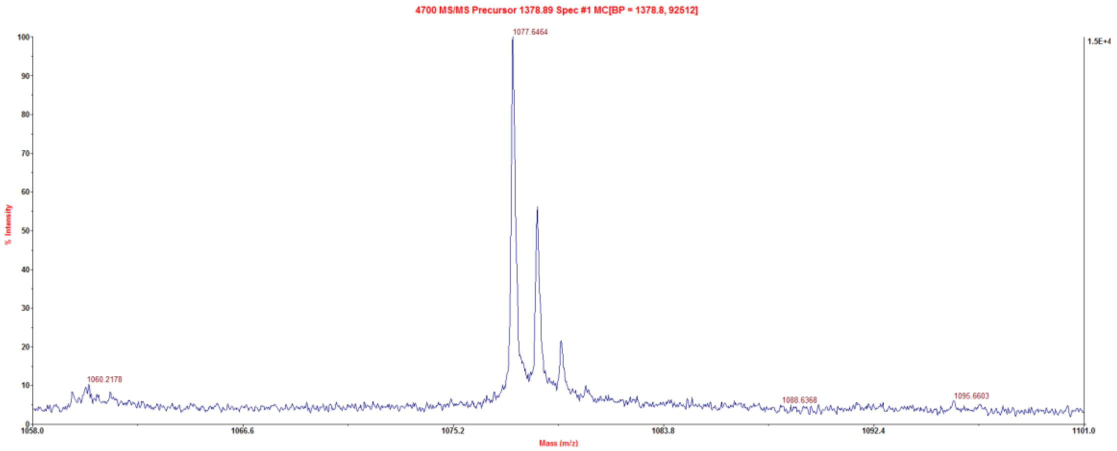


KHGTVVLTALG 1077.65



KHGTVVLTAL 1020.62