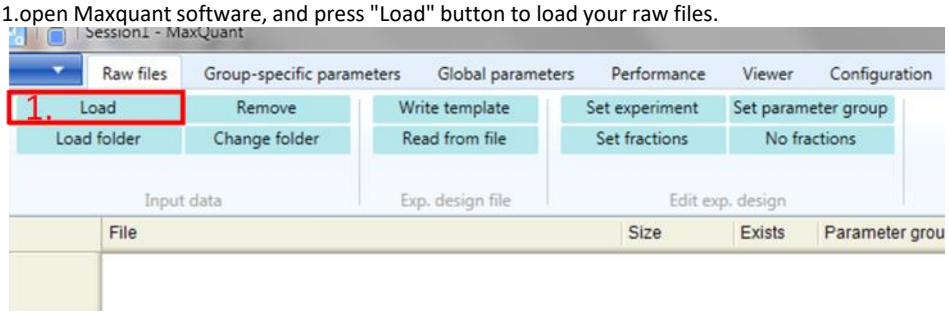
MaxQuant Search

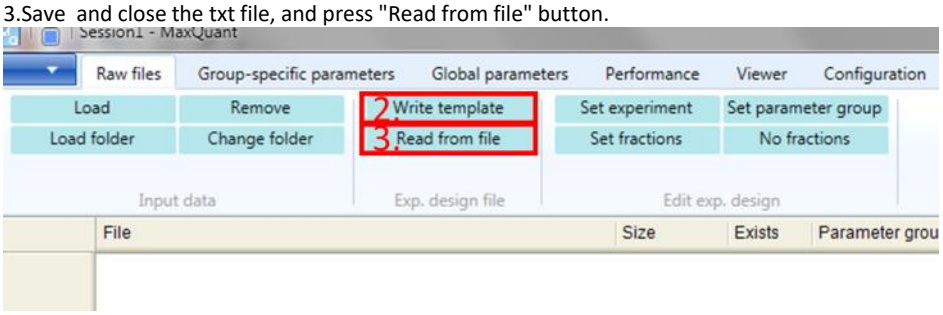
2016年3月31日　　上午 11:33

1.open Maxquant software, and press "Load" button to load your raw files.

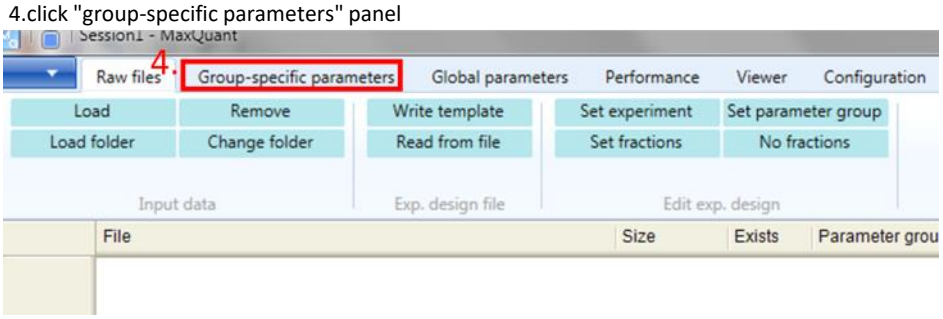


2.press "Write template" button, and opne "designtemplate.txt" file to set sample name and number of fractions.

3.Save and close the txt file, and press "Read from file" button.



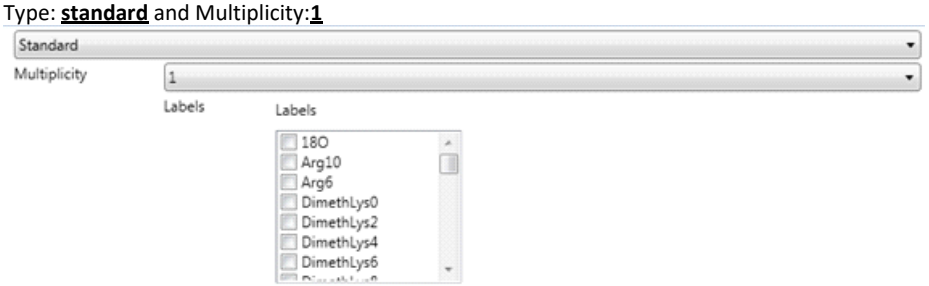
4.click "group-specific parameters" panel



4.select the type of data analysis:

a)identification only and label-free quantification (LFQ)

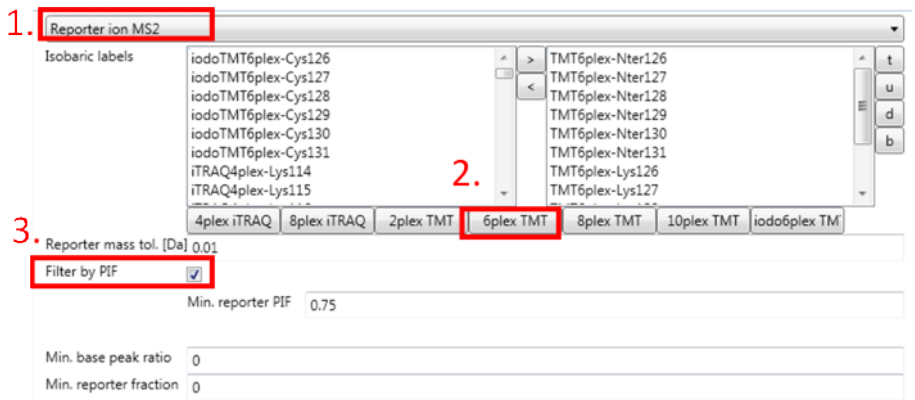
Type: **<u>standard</u>** and Multiplicity:**<u>1</u>**



b)6-plex TMT

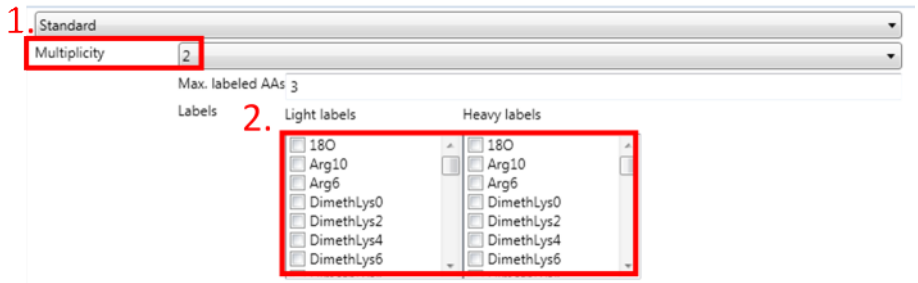
Type: **<u>Reporter ion MS2</u>** and click **<u>6plex TMT</u>**

enable "filter by PIF"

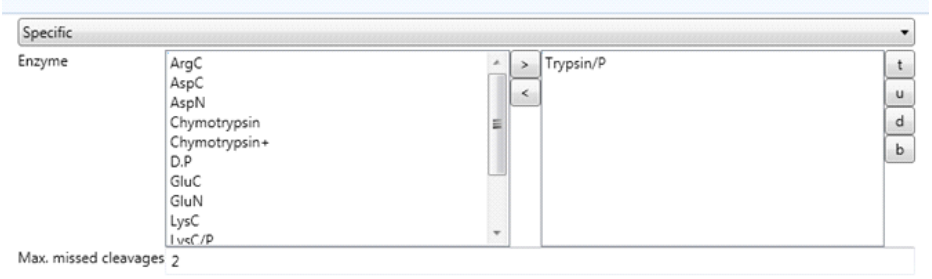


c)2plex SILAC and dimethyl labeling

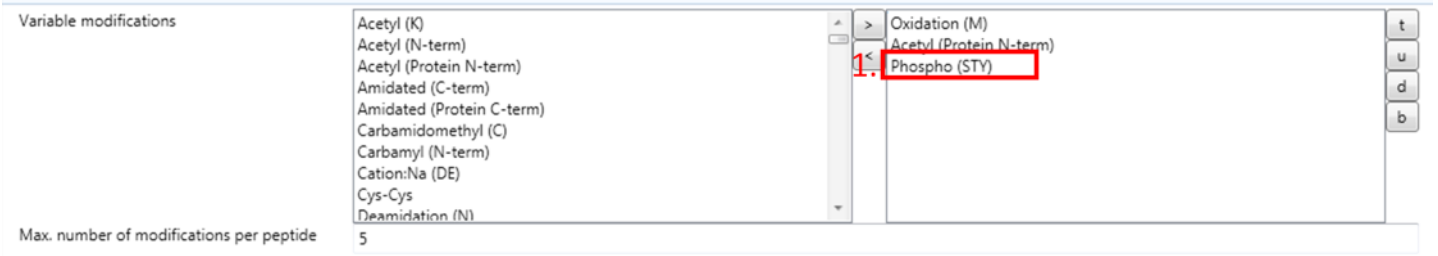
Type: **standard** and Multiplicity:**2**
Select the light and heavy isotope labels you used



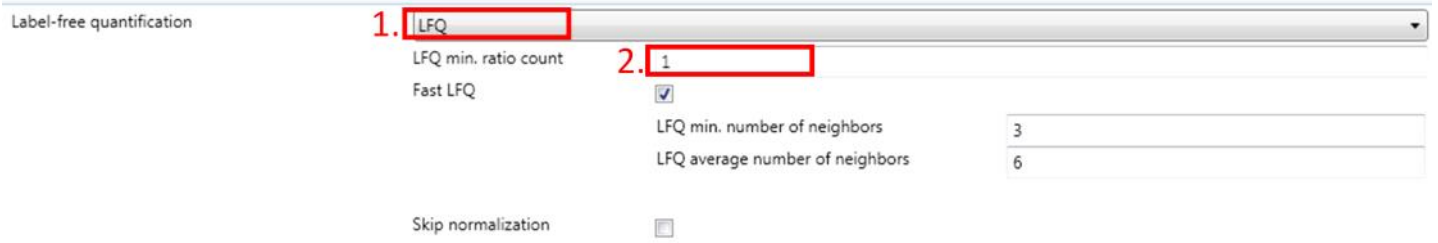
5.click "Digestion" tab, and select the digestion enzyme you used(usually Trypsin/P or Trypsin)



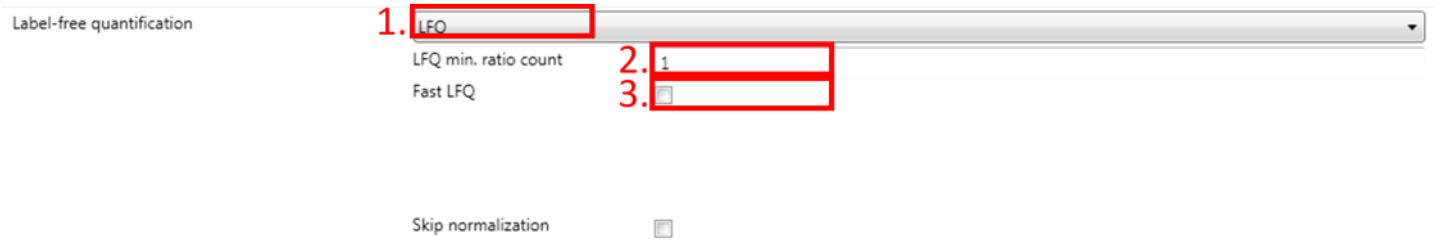
6.click "modifications" tab, and select the variable modifications you would like to search to the right panel
e.g. add phospho(STY) for phosphoproteomic data



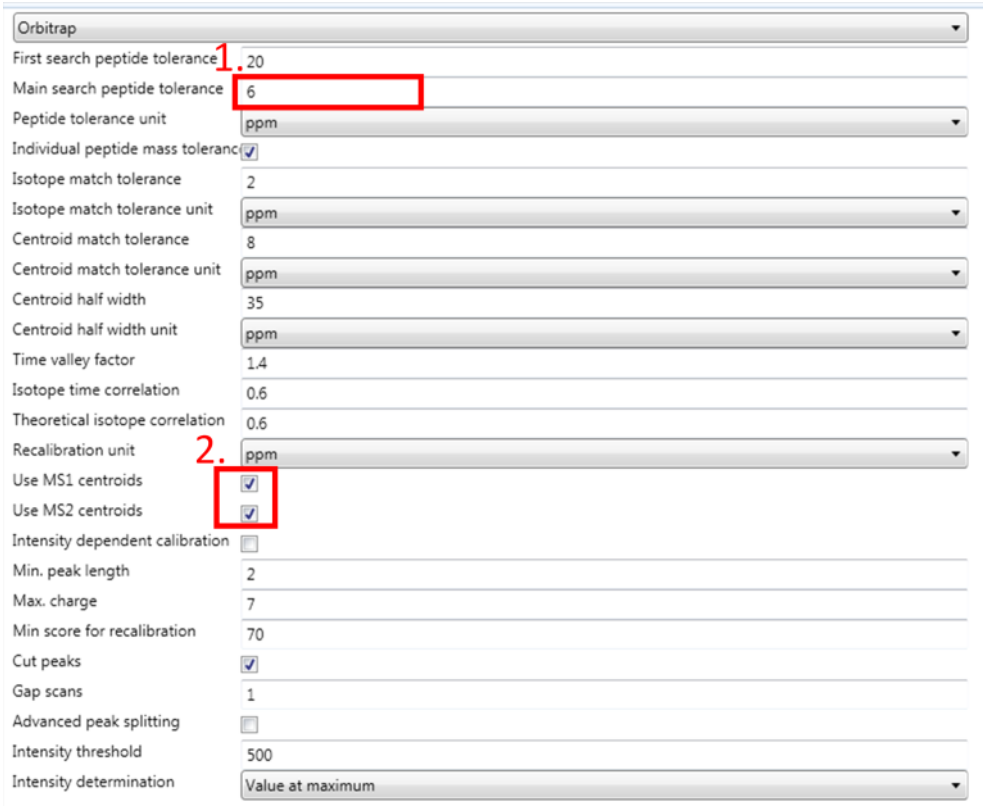
7.If you would like to perform LFQ and the number of samples more than 10, click "label-free quantification" tab, and (1) enable LFQ, and (2) LFQ ratio count: **1**



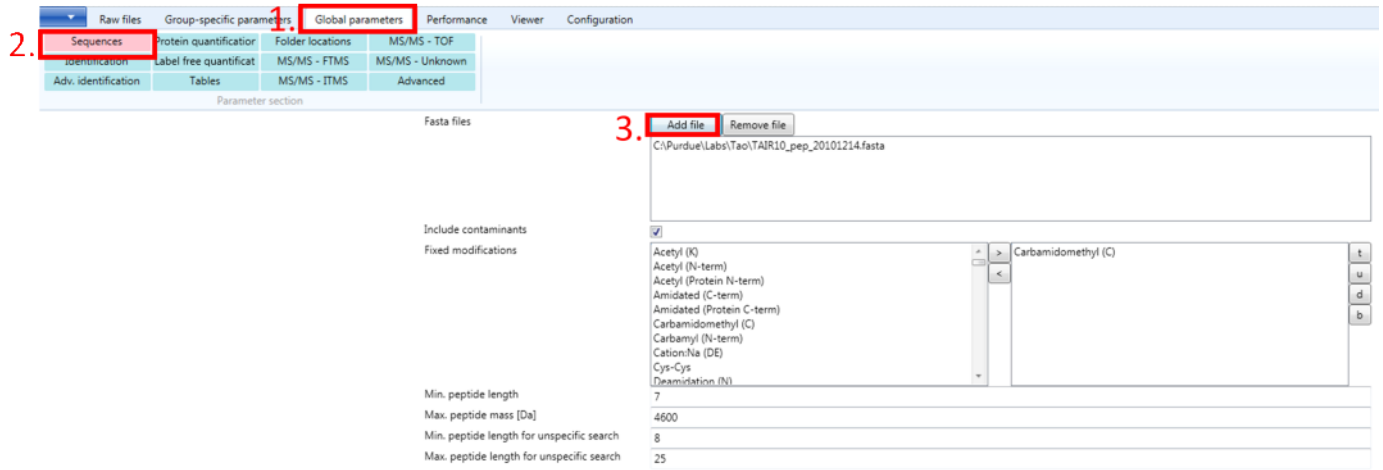
If the number of your samples less than 10, disable "fast LFQ".



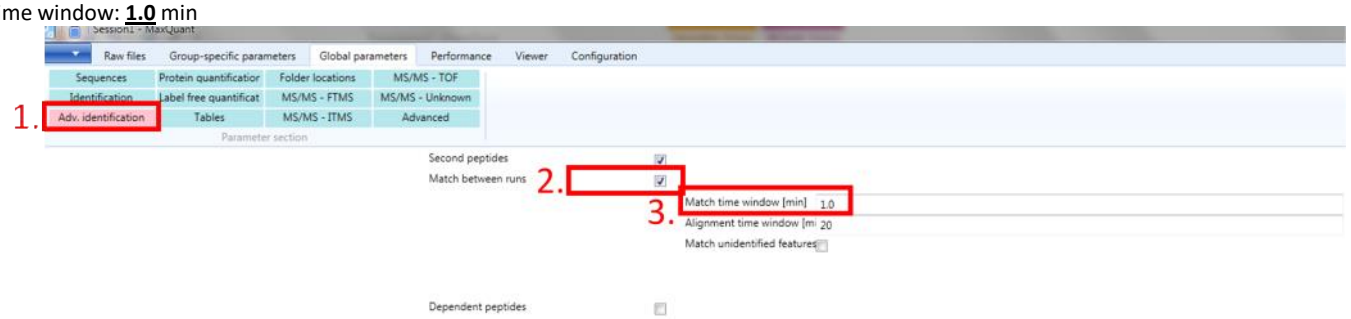
8.click "Instrument" tab, (1) main search tolerance: 6 ppm (2) if you use CID fragmentation, then enable "use MS1 and MS2 centroids"



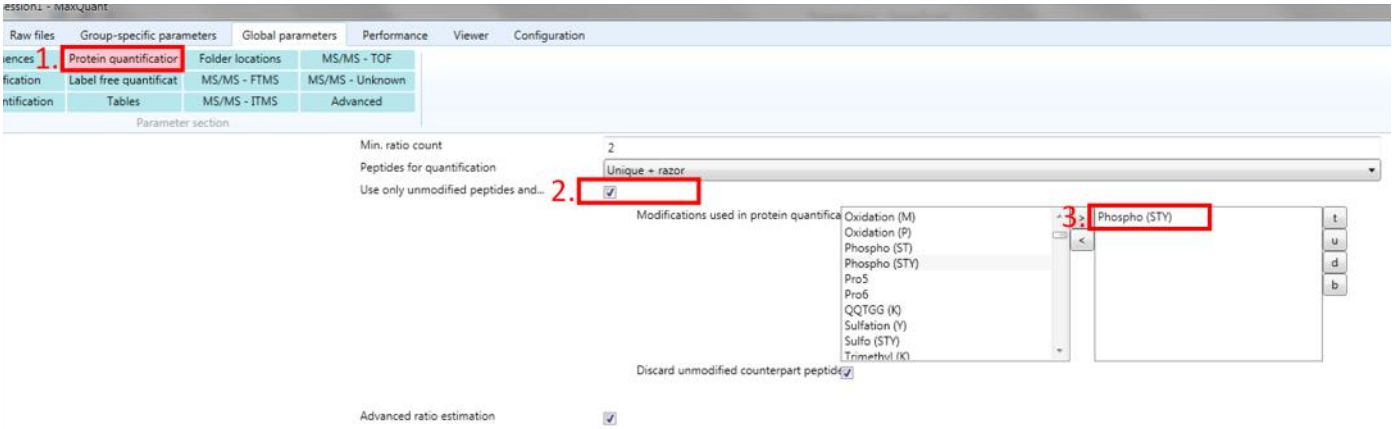
9.click "Global parameters" panel, add the database fasta file



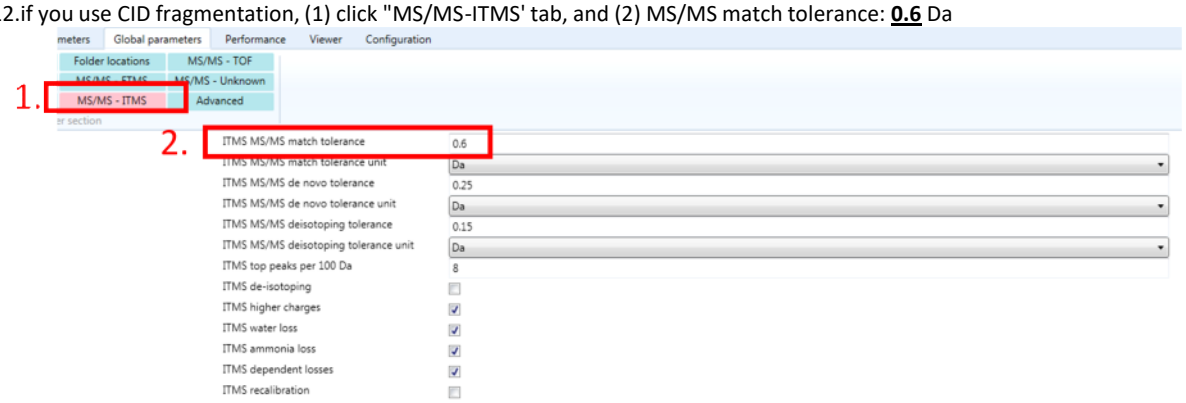
10.If you perform LFQ, (1) click "adv. Identification", (2) enable 'match between runs", and (3) match time window: **1.0** min



11.if you would like to quantify the amount of protein based on the specific modifications, (1) click "protein quantifiacation" tab, and (2)enable use "unmodified and modified peptides" (3) select the modification you want



12.if you use CID fragmentation, (1) click "MS/MS-ITMS' tab, and (2) MS/MS match tolerance: **<u>0.6</u>** Da



13.select the number of threads you want to use (usually 4-8 threads), and click "start" tab to perform Maxquant analysis