Modification Structure & Add Mass Calculation

Please complete and return the following form

User name:_____________________     Sample name: _____________________    Date:_______

In order for us to better understand and expedite your project, please provide the following information:

- The structure, formula and monoisotopic molecular weight of the reagent
- The chemical reaction involved
- The monoisotopic molecular weight that is added to the protein, ("delta mass", "net add weight").

Any additional information concerning the stability of the modified protein, e.g. acid, base, thermal or light lability, should be noted below. Please include the vendor and product number of the reagent, if possible. These steps insure that all lab members have a clear idea of both the structure and the chemistry involved. An example is provided on page 2.

REACTION OF REAGENT WITH PROTEIN (Please draw):

NET ADD MASS CALCULATION (List and calculate all masses as monoisotopic):
(A calculator can be found at http://www.sisweb.com/cgi-bin/mass11.pl. Choose high resolution)

Reagent source:                           _________________________________

Literature ref. (if needed)                  _________________________________

Molecular formula of reagent                  _________________________________

Molecular weight of neutral reagent            _________________________________

MW of FULLY PROTONATED reagent               _________________________________

Minus groups lost from reagent                _________________________________

Plus groups gained by reagent                 _________________________________

Minus groups lost from protein                _________________________________

Net add weight (monoisotopic & neutral)       _________________________________

Please return this completed form to proteomics@fas.harvard.edu or fax 617-495-1374, and print a copy to accompany your sample when submitted.
Here is an example of how this form should be used to provide the information needed to determine a specific modified amino acid, in this example a derivatized cysteine.

**REACTION OF REAGENT WITH PROTEIN**

\[
\begin{align*}
\text{Na}^+ & \quad \text{O} \quad \text{I} \\
\text{O} \quad \text{C} \quad \text{H}_2 \quad \text{O}_2 \quad \text{I} \quad \text{Na} \\
\text{HS} & \quad \text{N} \quad \text{H} \quad \text{R} \\
\text{C} \quad \text{H}_2 \quad \text{O}_2 \quad \text{I} \\
\text{O} \quad \text{C} \quad \text{H}_3 \quad \text{O} \\
\end{align*}
\]

Sodium Iodoacetate

\[
\begin{align*}
\text{O} \quad \text{C} \quad \text{H}_2 \quad \text{O}_2 \quad \text{I} \\
\text{O} \quad \text{C} \quad \text{H}_3 \quad \text{O} \\
\end{align*}
\]

Cysteine containing protein

\[
\begin{align*}
\text{O} \quad \text{C} \quad \text{H}_2 \quad \text{O} \\
\text{R} \quad \text{N} \quad \text{H} \quad \text{N} \quad \text{R}' \\
\text{O} \quad \text{C} \quad \text{H}_3 \quad \text{O} \\
\end{align*}
\]

Alkylated cysteine

The modification is not acid, base or light sensitive.

List the following masses as Monoisotopic (http://www.sisweb.com/cgi-bin/mass11.pl) (Choose High Resolution)

Reagent source: Sigma I-6125

To calculate the add weight:

- Calculate the molecular weight of the **FULLY PROTONATED REAGENT**:
  - sodium iodoacetate (C\(_2\)H\(_2\)O\(_2\)I) = 207.9012
  - sodium ion = -22.9900
  - proton (C\(_2\)H\(_3\)O\(_2\)I) = +1.0100
  - protonated reagent molecular weight = 185.9212

- Subtract the weight of any **reagent groups lost** during the reaction:
  - iodide ion = -126.9000
  - net add weight = 59.0200

- Add the weight of any **reagent groups gained** during the reaction:
  - no change in this case = 0.0000

- Subtract the weight of any **protein groups lost** during the reaction:
  - proton = -1.0100

- The total is the add weight:
  - Add weight = 58.0100