ZipTip Cleanup of Peptide Samples

Boisvert lab, december 2020

This protocol is adapted for the use of a 96-well (Sarstedt cat: 82.1581.001) plates with the 10 ul Ziptips. If the 100 ul Ziptips are used, then simply multiply by 10 the number of ul used in the different steps. Always use MS-grade water for all of your solutions. Never release the thumb at the end of each step of the protocol, keep your pipet at the ‘’dispense” position before going to the next step (in order to avoid air sucking in the tip). At the end of the protocol, speedvac your tubes containing the eluted peptides, resuspend in 30-100 ul of 1% formic acid, quantify using nanodrop at 205 nm and transfer the resuspended peptides in MS glass vial.

A = 100% acetonitrile (wetting solution, 70 ul (to compensate for evaporation)), aspirate/dispense (in G) 10 ul, repeat twice.

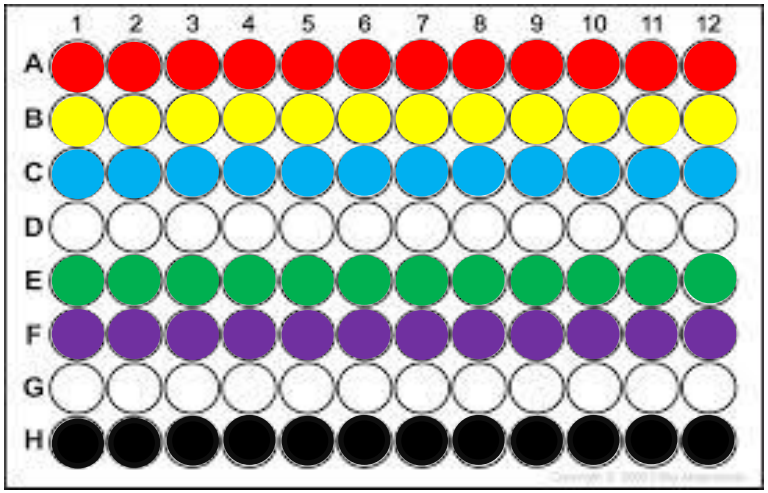
B = 0.1% TFA (trifluoroacetic acid) (equilibrating solution, 40 ul), aspirate/dispense (in G) 10 ul, repeat twice.

C = sample (in 0.1% TFA (trifluoroacetic acid), 30 ul or take it directly from your sample tube).

D = up/down for sample (10 cycles of up/down for 10 ul of sample), then dispense the 10 ul in H, repeat twice.

E= 0.1% TFA (trifluoroacetic acid), (washing solution, 40 ul), aspirate/dispense (G) 10 ul, repeat twice.

F = elution buffer (1% FA (formic acid)/50% acetonitrile, 30 ul, **only one sample at a time and just before starting the protocol of that sample** (evaporation problem)), 10 cycles of up/down for each 10 ul in a **new lobind tube, dispense in the same new lobind tube.**

G = trash for liquid from A, B, E

H = trash for liquid from D