TSS, TXS, SDS Serial Extraction

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This protocol is used to serially extract protein from mouse brain tissue in increasingly stringent buffers.

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1 Weigh brain sample and add 3X volume of TSS buffer + 1X protease inhibitor. 2m
TSS Buffer: 140mM NaCl + 5mM Tris-HCl in H_2O

2 Homogenize in a dounce homogenizer then transfer to a clean tube. 2m

3 Centrifuge at 21130g for 30 minutes at 4^{o}C then save the supernatant as 'TSS fraction'. 30m

Resuspend the pellet in the same volume of TSS buffer then centrifuge at 21130g for 30 35m

4 minutes at 4^{O}C and discard the supernatant.

5 Resuspend the pellet in the same volume of TXS buffer then incubate on ice for 10 minutes. 15m
TXS Buffer: TSS buffer + 0.5% Triton X-100

6 Centrifuge at 21130g for 30 minutes at 4^{O}C then save the supernatant as 'TXS fraction'. 30m

7 Resuspend the pellet in the same volume of TSS buffer then centrifuge at 21130g for 30 minutes at 4^{O}C and discard the supernatant. 35m

8 Resuspend the pellet in the same volume of SDS buffer then incubate at room temperature for 10 minutes. 15m
SDS Buffer: TSS buffer + 1% sodium dodecyl sulfate (SDS)

9 Centrifuge at 21130g for 30 minutes at room temperature then save the supernatant as 'SDS fraction'. 30m

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