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Isolation of genomic DNA from post mortem tissue

Genomic DNA was extracted from the post mortem tissue samples listed in Table 2.1 for mutation analysis and X chromosome inactivation studies, using a modified version of Sambrook (Sambrook and Russell, 2001).

DNA was isolated from the frontal and occipital cortices in each case, and parietal cortex, cerebellum, substantia nigra, thalamus, temporal cortex, and hippocampus subject to availability. The outer cortical layers were obtained using a scalpel and forceps whilst the specimen remained frozen on dry ice.

Method for DNA extraction

To extract the DNA:

1. Suspend 50 mg of tissue in 2 mL of SE buffer (75 mM NaCl, 25 mM EDTA pH 8.0).
2. Pass the suspension through a 19 gauge (G) syringe.
3. Add Proteinase K to a final concentration of 200 ug.
4. Add 1% SDS. Mix. Incubate overnight at 37C.
5. Add 2 M NaCl. Vortex for 20 seconds. Centrifuge at 1000 x g/ 20 minutes/ 25 C.
6. Transfer supernatant to a fresh tube and add 2 volumes of 100% (v/v) cold ethanol.

Related concepts

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Post mortem samples

Post Mortem Samples

Name	Gender	Age	PMI	Phenotype	Available regions	Source
RTT 1	F					
RTT 2	F					
RTT 3	F					
RTT 4	F					
RTT 5	F					
RTT 6	F					
RTT 7	F					
RTT 8	F					
RTT 9	F					
RTT 10	F					
CTL 1	F					
CTL 2	F					

Name	Gender	Age	PMI	Phenotype	Available regions	Source
CTL 3	F					
CTL 4	F					
CTL 5	F					
CTL 6	F					
CTL 7	F					
CTL 8	F					
CTL 9	F					