Hair sample analysis process

Each sample was segmented by 3cm (N= 33) if they had enough length, otherwise, their original length was kept, between 2cm-2.9cm (N = 11). Segmented samples were then washed twice in 5ml of high-performance liquid chromatography (HPLC) isopropanol with constant rotation. Hair was dried under forced air for 3-4 hours in a fume hood to dry. Dried samples were weighted between 5 and 15 mg depending on their amounts. Weighed samples were ground in a ball mill for 8 minutes at 30hz. The powdered hair was then extracted in 1.5ml of methanol over a 24-hour incubation period using continuous inversion at room temperature. After the incubation period, samples were centrifuged at 5000rpm for 5 minutes before 1ml of supernatant was transferred to a new clean vial and dried under a nitrogen evaporator in a water bath (50 °C). The extract was then reconstituted with 150µL of kit-provided assay diluent and assayed using commercially available enzyme-immunoassay kits (Salimetrics, PA). The Inter-assay C.V. (between plates) was 1.70 and Intra-assay C.V. (between samples) was 5.54. The cortisol levels for each respondent were examined for irregularities.