IDENTIFICATION OF 18 YEARS OLD BONES THROUGH DNA RELATIONSHIP TESTING USING FREEZER MILL WITH LIQUID NITROGEN FOR DNA EXTRACTION

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The Cuyahoga County Medical Examiner's Office received skeletonized unidentified human remains and was charged with the responsibility of identifying the remains. FBI agents found the remains in the isolated sewer manhole under an overpass, just south of Interstate 90 where several highway routes and ramps intersect. Bones were dark in color, soiled and appeared to be very old. Because of the condition of the remains DNA identification was the only option.

Authorities believed that the remains are those of Christina Adkins, a17-year-old who disappeared on January 10, 1995. In 1995, when Adkins disappeared, authorities focused mostly on her boyfriend, Jose Rivera. At the time, Elias Acevedo was questioned in connection with her disappearance but didn't appear to be a feasible suspect. But now, 18 years after Adkins' disappearance, his DNA profile had turned up during testing of a sexual assault evidence kit originally collected in 1993. Along with new information about several other sexual offenses, authorities believed that Acevedo was responsible for Adkins' disappearance. Acevedo eventually admitted to kidnapping and strangling Adkins and led the FBI agents to her remains.

Traditional extraction of the bone fragments and tooth didn't yield any useable DNA.

A portion of the femur bone and some teeth were washed and scraped clean with 10% bleach solution and 70% alcohol. Small pieces of femur bone were used for DNA extraction. Pieces were pulverized using "SPEX SamplePrep" Freezer Mill using liquid nitrogen.

One tooth was also pulverized using "SPEX SamplePrep" Freezer Mill.

Organic extraction was performed and sample processing was carried out using Amicon® concentrators.

Extracts were eluted in a volume of approximately 30ul.

Samples were purified using Centricsep from Prinston Separations.

Quantitation was performed using an AB 7500 using QuantDuo® kit.

Samples were amplified using Promega PowerPlex® 16HS and Fusion kits with 1ng DNA template.

Samples were run for analysis on AB 3500 and 310 where 1µl PCR product was loaded.

Increased injection time can be performed, as needed, and in this case some samples benefitted from an increased injection of 20 seconds.

Partial DNA profiles were developed from both the teeth samples and the bone samples.

10 markers were used from the profile developed from the tooth (amplified with Promega PowerPlex® 16HS) and 15 markers were used from the profile developed from the bone (amplified with Promega PowerPlex® Fusion).

Maternity statistical calculations were performed and a maternity index of 1740 was obtained from 9 Promega PowerPlex® 16HS markers and a maternity index of 2,110,000 was obtained from 15 Promega PowerPlex® Fusion markers.