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DNA Database Utility in Resolving Complex Incestuous Paternity Disputes

جدوى قاعدة بيانات الحمض النووي (DNA) في حل نزاعات إثبات النسب المعقدة الناشئة عن

العلاقات المحرمة للأقارب



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Abstract

Incestuous paternity investigations are considered among the most analytically and ethically complex challenges in forensic genetics. Extensive allele sharing occurs among family members and close biological relatives. Mutations in short tandem repeat (STR) markers, together with the concealment or frequent absence of key family members, can interfere with reliable paternity interpretation and increase the risk of false inclusion or exclusion. This study describes a legally significant forensic DNA case, where the use of the national DNA database revealed unexpected paternal candidates, ultimately resolving a highly complex case. Initial autosomal STR profiling using AmpFISTR™ Identifiler™ Plus and GlobalFiler™ multiplex systems indicated non-exclusion of an alleged father (AF1) accused of sibling incest, despite a single-locus mismatch consistent with mutation. Subsequent blind comparison within the DNA Database identified a second related individual who also could not be excluded as the biological father (AF2). Further investigation, including Y-chromosomal STR analysis and expanded testing of multiple maternal relatives, ultimately established a third individual (AF3), a maternal nephew, as the true biological father without mutational inconsistencies. These findings highlight the essential role of mutation-aware interpretation,

المستخلص

تُعد تحقيقات إثبات النسب في حالات العلاقات المحرمة بين الأقارب من بين أكثر التحديات تعقيداً من الناحيتين التحليلية والأخلاقية في علم الوراثة الجنائي؛ حيث يحدث اشتراك واسع في الأليلات بين أفراد الأسرة والأقارب البيولوجيين المقربين. إن وجود طفرات في مؤشرات التكرارات القصيرة المتتالية (STR)، إلى جانب إخفاء أو الغياب المتكرر لأفراد أساسيين من العائلة، يمكن أن يعوق التفسير الموثوق لإثبات النسب ويزيد من خطر الإدراج أو الاستبعاد الخاطئ. تعرض هذه الدراسة حالة حمض نووي جنائي ذات أهمية قانونية كبيرة، حيث أدى استخدام قاعدة البيانات الوطنية للحمض النووي إلى الكشف عن مرشحين أبويين غير متوقعين، مما أسهم في النهاية في حل قضية شديدة التعقيد. وأظهر التحليل الأولي لمؤشرات STR الجسمية باستخدام نظامي المضاعفة المتعددة AmpFISTR™ Identifiler™ Plus و GlobalFiler™ عدم استبعاد الأب المدعى الأول (AF1) المتهم في قضية علاقة محرمة بين الأشقاء، على الرغم من وجود عدم تطابق في موقع جيني واحد (-lo- Single mismatch) يتوافق مع حدوث طفرة. وعقب ذلك، أدى إجراء مقارنة عمياء داخل قاعدة بيانات الحمض النووي إلى تحديد فرد ثانٍ ذي صلة قرابة (AF2) لم يكن بالإمكان استبعاده أيضاً كأب بيولوجي. وأسفرت التحقيقات اللاحقة، بما في ذلك تحليل STR للكروموسوم Y وتوسيع الفحوصات لتشمل عدة أقارب من جهة الأم، عن تحديد فرد ثالث (AF3)، وهو ابن أخت/أخ الأم، بوصفه الأب البيولوجي الحقيقي دون وجود أي تعارضات طفرة.

Keywords: forensic science, incest, paternity testing, STR profiling, DNA database

الكلمات المفتاحية: علوم الأدلة الجنائية، العلاقات المحرمة للأقارب، فحص الأبوة (إثبات النسب)، التنميط الجيني لمؤشرات STR، قاعدة بيانات الحمض النووي



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comprehensive kinship analysis, and forensic DNA databases in accurately resolving complex incest-associated paternity disputes and preventing wrongful judicial outcomes.

تُبرز هذه النتائج الدور الجوهرى للتفسير الواعى بالطفرات، والتحليل القرابى الشامل، وقواعد بيانات الحمض النووى الجنائى فى الحل الدقيق لنزاعات إثبات النسب المعقدة المرتبطة بالعلاقات المحرمة بين ذوى القربى، ومنع صدور أحكام قضائية خاطئة.

1. Introduction

Paternity testing in the context of incestuous relationships represents one of the most analytically and ethically challenging scenarios in forensic genetics. Close biological relatedness among potential contributors leads to extensive allele sharing across autosomal short tandem repeat (STR) loci, thereby increasing the probability of apparent non-exclusion and complicating statistical interpretation. In addition, single-locus inconsistencies caused by germline mutations—well documented in STR systems—may further obscure correct kinship inference and create a risk of false inclusion or exclusion if not interpreted within accepted mutation models and population-genetic frameworks [1, 4].

In jurisdictions where incest constitutes a serious criminal offense accompanied by profound social and legal consequences, forensic DNA evidence often serves as the primary determinant in judicial decision-making. However, conclusions derived from limited reference samples, incomplete family structures, or absence of maternal comparison may result in erroneous attribution of biological parentage, potentially leading to wrongful conviction or failure to identify the true perpetrator. International forensic genetics guidance therefore emphasizes comprehensive kinship evaluation, cautious interpretation of STR mismatches, and the integration of additional lineage markers or database resources when standard trio analysis is unavailable [2, 3].

National forensic DNA databases provide a powerful but underreported tool for resolving

complex kinship disputes, particularly in cases involving concealed familial relationships or multiple related suspects. When combined with expanded STR profiling, Y-chromosomal lineage analysis, and targeted testing of extended relatives, database-assisted investigation can reveal hidden genetic connections and enable accurate reconstruction of biological relationships.

The present study describes a legally significant forensic case from the Kurdistan Region of Iraq in which initial autosomal STR testing suggested non-exclusion of an alleged father in an incest accusation, despite a single-locus mismatch consistent with mutation. Subsequent blind searching of the regional DNA database, followed by Y-chromosomal analysis and extended familial testing, ultimately identified a different related individual as the true biological father. This case illustrates the critical importance of mutation-aware interpretation, comprehensive kinship analysis, and forensic DNA database integration in preventing misattributed paternity and ensuring just judicial outcomes in incest-associated investigations.

2. Materials and Methods

2.1. Case Information and Biological Sample Collection

This investigation was conducted following formal judicial authorization issued on 5 April 2023 to the forensic genetics laboratory of the Criminal Evidence Directorate, Ministry of Interior, Kurdistan Regional Government, to perform a paternity test. Buccal swab samples were initially obtained from a female child and an adult male suspect (designated



AF1) accused of sibling incest [5]. At the time of the initial examination, the alleged mother was reported missing and was therefore unavailable for reference sampling. All biological materials were collected, labeled, transported, and stored in accordance with established forensic chain-of-custody and evidence-handling procedures.

2.2. DNA Extraction, Quantification, and Autosomal STR Amplification

Genomic DNA was obtained from buccal swabs using a validated silica-based DNA extraction method. The DNA's concentration and purity were measured with real-time PCR system (Applied Biosystems 7500).

Autosomal STR Amplification

Autosomal short tandem repeat (STR) profiling was performed using AmpFISTR Identifiler Plus multiplex kit (Thermo Fisher Scientific) targeting 15 autosomal STR loci, along with the Amelogenin marker for sex determination.

- Initial STR analysis showed that the alleged father (AF) could not be excluded at 15 loci.
- One discrepancy was observed at the TH01 locus: the AF had alleles 7 and 9, whereas the child had alleles 6 and 6.

Follow-up Analysis

A second analysis using a GlobalFiler kit (Thermo Fisher Scientific) having 21 autosomal STR loci confirmed that the AF shared at least one allele with the child at all loci except TH01 [6].

- This supported the conclusion that the AF could not be excluded as the biological father.
- The single TH01 inconsistency was interpreted as a probable mutation in the child, and a paternity index was calculated, yielding a likelihood of 99.99%.

Legal Outcome

Based on the DNA evidence and review of legal arguments, the court concluded:

- The man (AF) was identified as the biological father and sentenced to prison.
- The child was placed with adoptive parents.

2.3. Forensic DNA Database Comparison

Several months after the initial report, an independent STR profile obtained from a male suspect in an unrelated criminal investigation (designated AF2) was entered into the regional forensic DNA database (Small Pond). Routine database searching identified a potential biological relationship between AF2 and the child's profile. Comparative interpretation followed recognized forensic database matching and kinship evaluation standards [2,3]. AF2 was sharing at least one allele with the child in all 21 autosomal STR loci from the commercially available kit (Globalfiler, ThermoFisher Scientific) except at the D18S51 locus, where AF2 showed alleles 15 and 15, while the child showed alleles 16 and 17. Allowing one mutation, the AF2 could not be excluded as a biological father.

2.4. Y-Chromosomal STR and Kinship Analysis

Y-chromosomal STR profiling (Y-Filer Plus, Thermo Fisher Scientific) was performed to assess patrilineal relatedness between AF1 and AF2, following international forensic standards. Database comparison revealed that AF1 and AF2 are brothers, explaining shared alleles observed in prior testing and confirming their familial relationship.

2.5. Maternal Identification and Extended Familial Testing

Subsequent investigative efforts identified the biological mother outside the Kurdistan Region. With legal authorization, reference blood and buccal samples were collected and compared genetically to the child to confirm maternity.



Table 1- Summary of genetic analysis outcomes for key individuals in the incestuous paternity investigation.

Individual	Relationship to child	Autosomal STR outcome	Mismatch locus	Interpretation
AF1	Maternal uncle	Not excluded	TH01	Mutation considered
AF2	Maternal uncle	Not excluded	D18S51	Mutation considered
AF3	Maternal nephew	Fully concordant	None	Confirmed biological father
Mother	Biological mother	Fully concordant	None	Maternity confirmed

To clarify the conflicting paternal findings, DNA samples were also obtained from available adult male relatives on the maternal side, including brothers, brothers-in-law, nephews, and first cousins. Autosomal STR profiling was conducted using the SeqStudio system (Thermo Fisher Scientific). Profiles were developed for a younger brother, one brother-in-law, five nephews, and eight cousins.

Comparative analysis revealed that the child's conclusive biological profile, with no mutational discrepancies, matched one of the mother's nephews, a fifteen-year-old male (AF3). Following these findings, the mother was arrested, confessed to the offense, and was charged with sexually assaulting her nephew. The maternal brothers, AF1 and AF2, remained in custody for rape, while the minor, AF3, was placed in a youth shelter for protection.

3. Results

Initial autosomal STR testing indicated that the first alleged father could not be excluded as the biological parent, despite the presence of a mismatch at the TH01 locus, consistent with a potential mutation. Subsequent database comparison identified a second individual (AF2) who similarly could not be excluded. Y-STR analysis demonstrated close patrilineal relatedness between the two alleged fathers. Following recovery of the maternal reference sample, comprehensive kinship analysis of multiple male relatives conclusively identified a third individual (maternal nephew) as the true biological father without evidence of mutational

inconsistencies. A summary of the genetic outcomes for all tested individuals is presented in (Table 1).

4. Discussion

This case highlights the significant risk of false inclusion in incest investigations when maternal samples are unavailable and related individuals are tested in isolation [3]. Single-locus mismatches should be interpreted cautiously, as mutations are well documented in STR loci [4]. The use of blind DNA database searching proved instrumental in revealing hidden familial relationships and guiding further investigation. Ethical safeguards, child protection considerations, and controlled disclosure of incidental findings are essential components of responsible forensic practice in such sensitive cases.

5. Conclusion

The integration of DNA databases with extended STR and kinship analysis provides a powerful framework for resolving highly complex incestuous paternity disputes. This case underscores the necessity of trio testing whenever possible and demonstrates how comprehensive genetic investigation can prevent wrongful convictions and protect vulnerable individuals.

Ethical Approval

This investigation was conducted under full ethical and legal approval from the Kurdistan Regional Government Ministry of Interior. All DNA



sampling, profiling, and database comparisons were performed with explicit judicial authorization for forensic casework purposes.

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Conflict of Interest

The authors declare no conflicts of interest.

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