SHORT ARTICLES / INVESTIGACIÓN

Characterization of paternity exclusions in the Forensic Medicine service (Honduras) between 2018 and 2022

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DOI. 10.21931/RB/2023.08.02.35

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Abstract: The data on inclusions and exclusions of paternities carried out by the Forensic Genetics of the Public Ministry in Honduras are presented. All these paternities verifications have been included. The results show that there is an exclusion of about one-third of all official filial paternity tests, which has remained constant in proportion between the years 2018 to 2022; the data used corresponds to the totality registered in the Public Ministry, carried out throughout the territory of Honduras in the indicated years, for a total sample of 321 filial paternity test records. Genetic testing has been developed with sequenced, court-ordered, or civil samples where filial parentage is desired. The exclusion rates are like other countries in the region; it is a pending task to determine the associated cultural element for the high variability in the different countries of the exclusion rates.

Key words: Paternity, forensic medicine service, polymerase chain reaction, filial parenthood exclusion, DNA sequencing.

Introduction

A paternity test, also known as parentage testing or DNA testing, is a process by which it is determined whether a man is the biological father of a child. These tests are performed to confirm or rule out the biological relationship between a presumed parent and a child. The paternity test is based on the analysis of DNA, which is the genetic material present in every cell of the human body. During the process, DNA samples are collected from the alleged father, the child, and, in some cases, the mother. Samples can be obtained through a cheek swab or blood draw[1].

DNA is unique for everyone except for identical twins, who share the same DNA. By comparing DNA samples from the presumed father and child, scientists can identify specific genetic patterns passed down from parent to child. A high probability of biological kinship is established if enough matches are found in the genetic patterns. Paternity tests are used in different situations, such as legal custody disputes, alimony applications, and inheritances, or to satisfy people's curiosity about their biological ancestry. Importantly, these tests are reliable, and their accuracy can exceed 99% in many cases[2-4].

For the specific case within the legal framework of Honduras, only the Forensic Medicine Service of the Public Ministry can perform legally binding paternity tests, either to be presented as evidence in trials or to carry out family recognition and related procedures. This service is available as of the year 2008[5].

A paternity test exclusion occurs when the results of a paternity test indicate that a man cannot be the biological father of a child. In other words, the test shows no genetic relationship between the man tested and the child in question. To do this, the genetic profiles of the presumed father and the child are compared. Exclusion is determined if no matches are found in the genetic markers analyzed. This means the man does not share the necessary genetic material with the child to be considered his biological father[6,7].

When there is an exclusion in a paternity test, it is concluded that the man analyzed is not the child's biological father. This information may be relevant in legal situations, such as custody disputes or paternity claims, where establishing the biological truth is required. It is important to note that modern paternity tests are high accuracy, and the exclusion is reliable. However, it is critical to follow proper protocols and obtain DNA samples from all parties involved correctly and safely to ensure the accuracy of the results[1].

There are no precise global statistics on paternity exclusions, as not all cases of paternity doubt undergo genetic testing. In addition, exclusion rates may vary by region, population, and other factors. There are studies in this regard, but they have been collected using statistical techniques by specific authors. There needs to be official information from the states in this regard. Overall, it is estimated that 10% to 30% of men undergoing paternity testing are excluded as biological fathers worldwide. These approximate figures may vary depending on the studies and the samples analyzed[8].

Determining an infant's paternity is vital in a developing country like Honduras, where economic and social conditions require clarifying the legal and economic responsibilities of the children. In other cases, many children result from rape processes where, for legal purposes beyond child support, knowing paternity is decisive for applying sanctions

Citation: Molina Y, Raudales I, Moya Z, Zablah J I, Loureiro A G, Madrid M, Agudelo-Santos C, Diaz S and Valle-Reconco J.Characterization of paternity exclusions in the Forensic Medicine service (Honduras) between 2018 and 2022. Revis Bionatura 2023;8 (2) 35. http://dx.doi.org/10.21931/RB/2023.08.02.35
Received: 10 February 2023 / **Accepted:** 15 May 2023 / **Published:** 15 June 2023
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to the abusers. In other cases, where the filial paternity tests are exclusive, they can relieve the presumed fathers who are wrongly responsible for the mothers who are not sure about the filial relationship from a legal and economic burden.

An erroneous paternity record can result in denying rights and benefits to both children and mothers. This includes limited access to education, health care, and social welfare programs. Furthermore, the lack of indeterminate paternity can contribute to an increased risk of abandonment and intrafamily violence, thus perpetuating a cycle of poverty and inequality.

Significantly, these estimates are based on cases of doubt or dispute about paternity, and tests are performed to clarify it. The exclusion rate is much lower in the general population, with no obvious doubt about paternity. It should be noted that these statistics may vary in different countries or regions, as cultural, social, and legal factors can influence the frequency with which paternity tests are conducted and exclusion rates. This study presents retrospective data on paternities performed between 2018 and 2022 in the Forensic Medicine service.

Among the objectives is to determine if the number of filial paternity tests has had a significant variation in frequency, to know the proportion of exclusive tests compared to the inclusive ones, the correlation between the results, and how these values compare with what is found in studies. Similar to other geographic regions, all in the annual period from which the data used comes.

Materials and methods

The data used in this article comes from the Serology and Genetic Services performed by the Public Ministry in Honduras between January 2018 and December 2022. This sample is composed of 321 filial relationship tests. All paternity tests were considered; none have been ruled out, so they represent the total attention that Forensic Medicine carried out in this regard between 2018 and 2022.

An Applied Biosystems model 3500 genetic sequencer is used for filial paternity tests. This instrument uses capillaries designed to obtain electropherograms, which contain genetic markers. The DNA fragments used in the sequencer are bound to fluorochromes detected by a laser that excites them at different wavelengths, thus achieving the analysis of multiple DNA fragments concurrently.

To perform a paternity test, informed consent must first be signed by the adults involved in the paternity test to carry out the extraction and paternity test. This document is standard throughout the Honduran territory and regulated by the Public Ministry; it has been approved for use since the creation of the Forensic Medicine service, specifically for Forensic Genetics.

Samples of the buccal epithelial cells of those involved in the paternity test are then taken, including the infant using a hyssop for DNA testing. These samples are placed on a classic FTA card. FTA (Flinders Technology Associates) cards are made of cotton-based cellulose paper; These contain chemicals that bind cells, denature proteins, and protect DNA, leaving samples suitable for molecular identification without the risk of contamination[9,10].

To these cards that have been impregnated with the epithelial cells, the Powerplex Fusion 6C amplification reagent is applied to later introduce it with the controls in a thermal cycler. At the end of this cycle, the amplified DNA is introduced to the genetic sequencer to obtain the electropherograms and be able to perform the analysis. Using M-FISys software version 7.87 allows the probabilities of kinship and identification based on genetic analysis[11,12].

Samples are discarded when amplification is incomplete. Usually, this is known in the sequencer due to a bad extraction procedure, a defective FTA card, poor transport, and poor preservation, among others. These conditions rarely occur because the Forensic Medicine service procedures are under rigorous quality control processes and protocols.

The filial paternity test is positive when 99.99% of the genetic markers are shared between the infant and the alleged father. Any lower measurement results in a negative test or exclusion criterion. In some cases, you may be asked to repeat the test. Usually, this decision is made at the judicial level.

Results

Of a sample of 321 filial relationship tests, 151 were done by court order and 170 voluntarily by citizens who wish to clarify paternity issues. All the samples have 98 exclusions (30.5%) and 221 inclusions (69.50%). The results of the exclusions yearly test have an arithmetic mean of 19.60, a median of 21, a standard deviation of 5.1769, and a range of 13. The inclusions yearly test has an arithmetic mean of 44.6, a median of 47, a standard deviation of 11.9708, and a range of 32. According to Shapiro-Wilk, exclusions and inclusions were tested, with the p-values obtained being 0.7330 (W=0.9494) for exclusions and 0.9261 (W=0.9784) for inclusions. These results indicated insufficient evidence to reject the null hypothesis of normality for both data sets. Figure 1 shows the frequencies and distribution of filial relationships in the years of study.

An independent t-test was performed to compare exclusions and inclusions in the provided dataset. The results revealed a significant difference between the two variables ($t = -3.09$, $p < 0.05$). On average, fewer exclusions were observed (mean = 19.6, standard deviation = 5.1769) to that inclusion (mean = 44.6, standard deviation = 11.9708). These findings suggest a statistically significant disparity in the number of exclusions and inclusions recorded in the years analyzed.

Notably, the independent t-test allows us to determine whether there is a statistically significant difference between the two groups. In this case, we assessed whether there was a significant difference between exclusions and inclusions. The results indicated a disparity between these two variables, suggesting that events related to exclusions and inclusions have a different impact depending on the data collected.

A Pearson correlation analysis was performed between exclusions and inclusions, to obtain a statistical measure that evaluates the linear relationship between them. The result obtained was a coefficient of 0.993, indicating a strong positive correlation between exclusions and inclusions. This means that as exclusions increase each year, inclusions also tend to proportionately and grow proportionately in the same year. The positive correlation suggests that these two values are closely related and together vary in the same direction.

Notably, the Pearson correlation only measures the linear relationship between inclusions and exclusions without

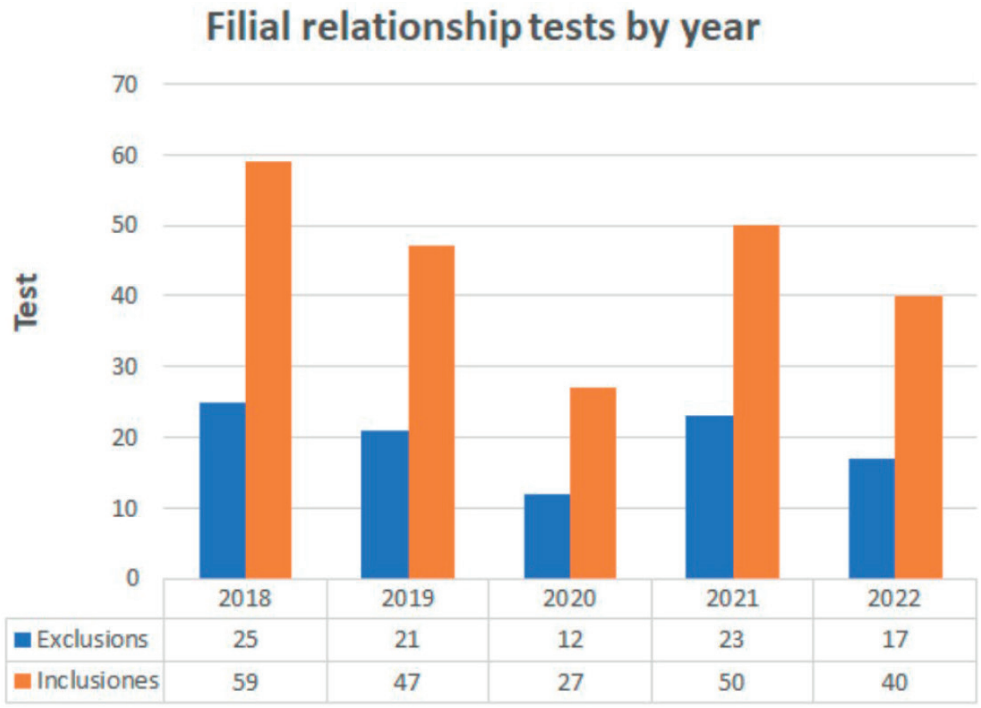

Figure 1. Frequencies of inclusions and exclusions of paternity affiliates between 2018 and 2022.

implying causation. In this case, we cannot say that exclusions directly cause inclusions or vice versa, but we can conclude that there is a strong association between them.

Discussion

The total number of tests of parental and filial relationships has remained similar between the years of study, being lower in 2020 due to the health crisis and quarantine. The ratio is one exclusion for every three paternity tests. If we compare this value with that published by Anderson *et al.*[8], it is an average value when compared with paternity exclusions from other geographic regions. Honduras has the same relative value of exclusions as measured in the United States (New York). Figure 2 shows a worldwide map with exclusion by country.

If we compare the 30.50% of exclusions obtained when compared with Cuba, the study of Collazo *et al.*[13] shows an overall value of 33% of paternity exclusions. This is a similar value in the Caribbean context, of which Honduras is a part. Some factors can cause exclusions to vary significantly in different geographical contexts. Let's compare the Latin American region against less than 1% of exclusions in Switzerland; according to Sasse et al., the region finds different realities in terms of economic, educational, human development, etc.

But Swiss behavior different differs from that reported in its neighboring countries; we find that Italy has 45% exclusions[15] and France has 38%[16], both higher than the Latin American values he indicates. that interestingly, in countries with a shared history, this difference is less marked, as in the case of Brazil with 22%[17] and Portugal with 27%[18].

Zavarin et al. published an exclusion percentage of 23.5% of court-ordered paternities in the Russian Federation, but Molyaka et al., report a value of 14.3%; both percentages are lower than what we measure in Honduras. If we compare G20 member countries[19], we find that Russia and the United Kingdom (16.6%) have the lowest exclusion percentages, which are half of what is reported in the United States on average. U.S. measurements are by city and classified by race[20].

In the country, many people may be unaware of the availability or process of paternity testing. This can lead to underutilization of tests or a lack of access. As a result, many exclusions may not be detected or confirmed. Additionally, the only valid evidence for legal purposes is that provided by the Forensic Medicine service.

There are also stigmatized social environments associated with paternity testing. People may fear social judgment, family breakdown, or legal repercussions when facing a situation of exclusion. This could lead to a reluctance to perform the tests, resulting in many unconfirmed exclusions. Paternity testing turns out to be expensive, requiring significant financial resources. The cost can be prohibitive for some individuals and families, making it difficult to access testing. This can contribute to many exclusions, as many people need help to afford to take the necessary tests to confirm paternity.

Notably, there are social reasons for the presence of exclusions, which can vary by culture, context, and social norms. It is essential to seek ways to address these issues from a multidimensional perspective that considers legal, cultural, economic, and educational factors to promote a greater understanding of and access to paternity tests and their proper interpretation but is focused on raising awareness of responsibility for reproductive decisions, gender, and fighting stigma and abuse of women and men alike. More studies are necessary to more substantial our results.

Conclusions

Although the numbers vary yearly, no clear trend exists in the relationship between exclusions and inclusions. While there was a decrease in both values overall, the changes do not follow a specific pattern and may be due to various circumstances. Therefore, there cannot be concluded that there is a direct relationship or obvious influence between exclusions and inclusions based on these data alone.

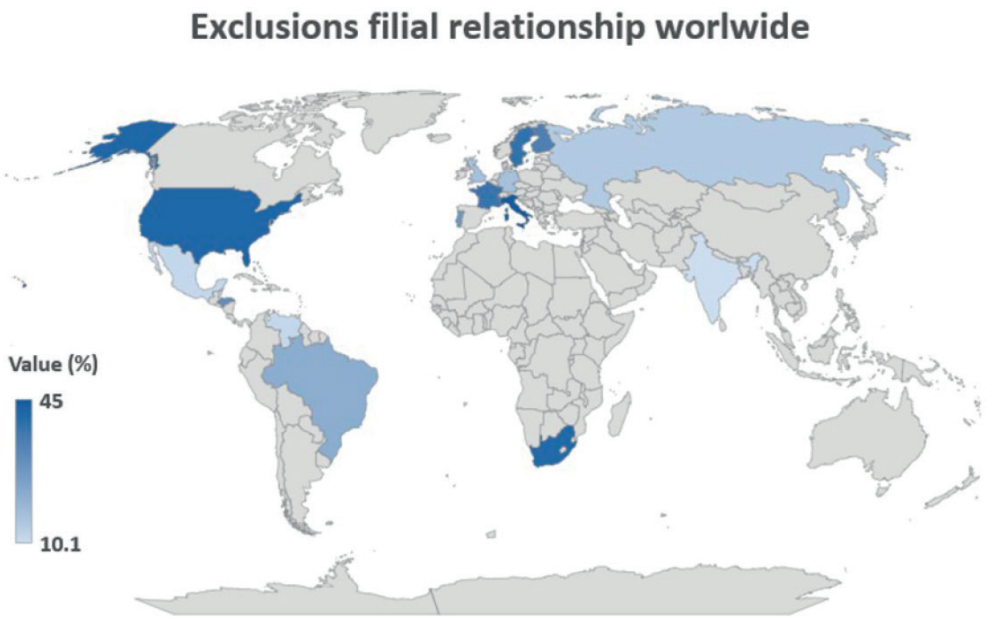

Figure 2. Map of percentages of filial paternity exclusion around the world. It is adapted from Anderson[8].

Since Honduras has complicated family dynamics, such as single-parent families, divorces, separations, or undeclared paternity or maternity, there may be a greater likelihood that paternity tests will result in exclusions. This is because the identification of the biological father may be uncertain, or there may be multiple possibilities.

Supplementary Materials

They have not been provided.

Author Contributions

Conceptualization, Yolly Molina and Ismael Raudales.; Methodology, Salvador Diaz and Marcio Madrid; validation, Zintia Moya; formal analysis, Jorge Valle-Reconco; investigation, Yolly Molina.; data curation, Antonio Garcia Loureiro; writing—original draft preparation, Isaac Zablah; writing—review and editing, Carlos Agudelo-Santos; supervision, Salvador Diaz All authors have read and agreed to the published version of the manuscript.

Acknowledgments

We thank the Public Ministry - Serology and Forensic Genetics for its support to get the data used in the publication of this manuscript.

Conflicts of Interest

The authors declare no conflict of interest.

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