25-753-cr

IN THE UNITED STATES COURT OF APPEALS
FOR THE SECOND CIRCUIT

UNITED STATES OF AMERICA,

Appellant,

– v. –

TYLER SCOTT JOHNSTON,

Defendant-Appellee.

On Appeal from the United States District Court
for the Eastern District Of New York

BRIEF OF *AMICI CURIAE* THE LEGAL AID SOCIETY, NATIONAL ASSOCIATION OF CRIMINAL DEFENSE LAWYERS, NEW YORK STATE ASSOCIATION OF CRIMINAL DEFENSE LAWYERS IN SUPPORT OF DEFENDANT-APPELLEE

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CORPORATE DISCLOSURE STATEMENT

Pursuant to Federal Rules of Appellate Procedure 29(a)(4)(A) and 26.1(a), *amici curiae* The Legal Aid Society, the National Association of Criminal Defense Lawyers, and New York State Association of Criminal Defense Lawyers disclose that no *amici* has a parent corporation and that no publicly held corporation owns 10% or more of *amici* stock.

INTEREST OF THE *AMICI CURIAE*[1]

The Legal Aid Society is the nation's oldest and largest private non-profit legal services agency, dedicated since 1876 to providing quality legal representation to low-income New Yorkers. It has served as the primary public defender in New York City since 1965 and, each year, represents thousands of people who are arrested and unable to afford private counsel.

In 2013, Legal Aid's Criminal Defense Practice established a DNA Unit to assist the more than 1,000 Legal Aid staff attorneys representing clients citywide with the increasing use of DNA evidence. The Unit has since expanded and remains among the largest practice groups dedicated to forensic evidence of any public defender office in the country. Its staff attorneys review lab reports and files produced during the testing of forensic DNA evidence across New York City, cross-examine forensic analysts on issues similar to those presented in this case, and assist other lawyers in doing so in cases ranging from misdemeanor charges to homicides. The DNA Unit conducts trainings nationwide on DNA and other forensic science

[1] The parties consent to the filing of this brief. Pursuant to Federal Rule of Appellate Procedure 29(c)(5), *amici curiae* state that no counsel for a party authored this brief in whole or in part, and no such counsel or party made a monetary contribution intended to fund the preparation or submission of this brief. No person other than the *amici curiae*, their members, or their counsel made a monetary contribution intended to fund the preparation or submission of this brief.

issues for both scientific and legal audiences. As such, the DNA Unit is uniquely positioned to address the issues before the Court.

The National Association of Criminal Defense Lawyers (NACDL) is a non-profit, voluntary professional bar association that works on behalf of criminal defense attorneys to ensure justice and due process for those accused of crimes or misconduct. NACDL was founded in 1958. It has a nationwide membership of many thousands of direct members, and up to 40,000 with affiliates. NACDL's members include private criminal defense lawyers, public defenders, military defense counsel, law professors, and judges. NACDL is the only nationwide professional bar association for public defenders and private criminal defense lawyers. NACDL is dedicated to advancing the proper, efficient, and just administration of justice. NACDL files numerous amicus briefs each year in the U.S. Supreme Court and other federal and state courts, in cases that present issues of broad importance to criminal defendants, criminal defense lawyers, and the criminal legal system as a whole.

The New York State Association of Criminal Defense Lawyers (NYSACDL) is a not-for-profit corporation with a subscribed membership of more than 1,500 attorneys, including private practitioners, public defenders, and law professors, and is the largest private criminal bar in the State of New York. It is a recognized state affiliate of the NACDL and, like that organization, works on behalf of the criminal

defense bar to ensure justice and due process for those accused and convicted of crimes.

PRELIMINARY STATEMENT

Modern DNA testing instruments are highly sensitive, detecting minute quantities of DNA that may have been deposited through normal, everyday activities, thereby complicating the interpretation and significance of biological findings. The presence of a person's DNA in a crime-scene sample must be evaluated within the context of a case now more than ever.

Indeed, a robust body of research on DNA transfer, persistence, prevalence, and recovery (TPPR) has made clear that DNA can be deposited through direct or indirect contact from everyday activities (even if involuntarily), and can persist for months or years on common surfaces and lived-in environments. This research has shown that DNA transfer scenarios are highly variable depending on the circumstances.

Forensic scientists have recognized the need for activity-level likelihood ratios (LRs), which evaluate competing explanations for 'how' and 'when' questions pertaining to an observed DNA result. However, the expertise, data, and case-specific information required for such evaluations remain limited in the United States. As a result, there is a substantial risk that DNA evidence – particularly complex mixtures evaluated through LRs addressing only the "who" question – will be misinterpreted or overstated if presented without the necessary case-related scientific context.

The DNA results here are a perfect storm of complex results wrenched out of context, and a prime example of the importance of adhering to methodological limits in forensic science. By setting propositions that do not reasonably reflect the limited universe of potential contributors who may have had contact with the comforter, the analyst generated a flawed LR value, from which the government will ask the jury to speculate that the presence of Tyler Johnston's and Doe's DNA can only be due to assault. However, the government is wrong on the science for the reasons that follow, and the District Court prudently exercised its discretion to exclude this evidence. This Court should affirm.

ARGUMENT

I. FROM SOURCE LEVEL TO ACTIVITY LEVEL: A PARADIGM SHIFT IN FORENSIC DNA.

A. Complex Mixtures Are Vastly More Difficult to Analyze, Interpret and Communicate to Juries Than "Gold-Standard" DNA Testing.

The purpose of traditional forensic DNA analysis is identification: to determine who deposited biological material on a piece of evidence by comparing the DNA markers detected there to samples taken from potential suspects. Forensic DNA analysis earned its "gold standard" reputation based on laboratories analyzing high-quality DNA samples from a single individual and simple mixtures (unambiguous DNA profiles from two people). Today, however, many DNA samples contain trace amounts of DNA from three, four and even five or more

separate individuals, and those samples – for the reasons discussed *infra* – are far more difficult to interpret.

Recent technological advances in the instruments and materials used in DNA analysis have vastly increased the sensitivity with which labs can detect DNA. Nat'l Inst. of Standards & Tech., *DNA Mixture Interpretation: A NIST Scientific Foundation Review*, NIST IR 8351, at 21-22 (2024), https://nvlpubs.nist.gov/nistpubs/ir/2024/NIST.IR.8351.pdf [hereinafter *NIST Mixture*]. Because greater sensitivity brings with it an increased ability to detect minute quantities of DNA, it has been aptly described as a "double-edged sword":

> We often shed small amounts of DNA when we talk, sneeze and touch things. As a result, many surfaces are likely to contain mixtures of minute amounts of DNA from several people. These mixtures have always been present at crime scenes, but when sensitivity was lower, they wouldn't have been detected or, if they were, labs would not have attempted to interpret them. That is no longer the case.

Nat'l Inst. of Standards & Tech., *DNA Mixtures: A Forensic Science Explainer* (2019), https://www.nist.gov/feature-stories/dna-mixtures-forensic-science-explainer. The resulting DNA mixtures (when biological material from more than one person is detected on a surface) are substantially more difficult to measure and analyze. *See NIST Mixture* 37-43 (discussing factors that contribute to increased uncertainty in measurement and interpretation); A993-98 (explaining the challenges in determining the number of contributors to DNA mixtures).

Because of the difficulty with mixture interpretation, forensic DNA labs have shifted toward probabilistic genotyping systems (PGS), which are software programs that use statistical theory, biological modeling, and complex algorithms to infer the genotypes of contributors to a DNA mixture. In essence, PGS attempt to account for the uncertainties inherent in forensic DNA analysis using biological and statistical models, and quantify the strength of the observed evidence by calculating the relative probability (under the modeled-for assumptions) of competing propositions, which is expressed as an LR. *NIST Mixture* 46-47. While far from infallible, PGS can be used within appropriate limits "to support the interpretation of DNA profile information and to inform opinions on the value of the findings given case-relevant propositions." Nat'l Inst. of Standards & Tech., *Forensic DNA Interpretation and Human Factors: Improving Practice Through a Systems Approach*, NIST IR 8503, at 69-71 (2024), https://nvlpubs.nist.gov/nistpubs/ir/2024/NIST.IR.8503.pdf [hereinafter *NIST Human Factors*].

Due to the sharp rise in the use of PGS among forensic laboratories, most DNA results are now expressed as an LR. An LR "involves a ratio of two conditional probabilities: the probability of the evidence given that one proposition (hypothesis or narrative) is true and the probability of the evidence given an alternative proposition is true." *NIST Mixture* 48. In forensics, the first proposition (H_1) – the

numerator in the LR – is generally derived from the prosecutor's theory of the case, and therefore posits that the defendant or person of interest contributed DNA to the sample in question. The second proposition (H_2) – the denominator in the LR – is meant to align with the defense position and therefore generally assumes another unknown individual in the mixture, instead of the defendant or person of interest. The second proposition is typically assigned by the laboratory and may not represent the actual position of the defense.

The numerical value of an LR depends on a host of factors, including the "evidence available, statistical models applied, propositions selected based on case information, and the scientist making various judgments." *NIST Mixture* 48. But at bottom, the LR is a comparison of the propositions set by the evaluator, and it will accordingly vary as different assumptions and methodologies are applied. *See* P. Gill et al., *DNA Commission of the International Society for Forensic Genetics: Assessing the value of forensic biological evidence—Guidelines highlighting the importance of propositions. Part I: Evaluations of DNA profiling comparisons given (sub-)source propositions*, 36 Forensic Sci. Int'l: Genetics 189, 191 (2018) [hereinafter Gill, *ISFG Part I*] ("Depending on our assumptions, our knowledge and the results we want to assess, different models will be adopted, hence different values for the LR will be obtained.").

Perhaps the most impactful of these factors is the forensic scientist's choice of propositions. As Professor Tim Kalafut acknowledged during the *Daubert* hearing below, "for the likelihood ratio to have meaning, the relevant information in the case needs to be considered" in formulating the propositions. A836-37. This is especially important when the case circumstances suggest that contributors to a DNA mixture may be related, as "[n]on-contributors who are relatives of true contributors can produce high LRs when considering propositions" that assume any unknown contributor is also unrelated. *See NIST Human Factors* 64-66 (recommending, *inter alia*, that labs consider case circumstances closely when evidence derives from a "family home" and condition using reference profiles from known relatives wherever possible).

Although LRs are accepted as a method to convey DNA results, they are notoriously difficult for lay audiences – including juries – to understand. The LR, represented by a single numerical value, expresses the relative probability of the observed results given two specific propositions, *not* the probability that either proposition is itself true. The prosecutor's fallacy occurs because "most legal end-users are not fluent in Bayesian statistical concepts and may misunderstand the likelihood ratio as a statement about the probability of the propositions themselves." Tex. Forensic Sci. Comm'n, *Final Report on Complaint No. 23.67*, at 21 (2023), https://www.txcourts.gov/media/1458950/final-report-complaint-2367-roy-tiffany-

073024_redacted.pdf. A simple example illustrates that the statements are not equivalent: The statement that "the probability that an animal has four legs if it is a cow" does not mean the same thing as the statement "the probability that an animal is a cow if it has four legs." Jo-Anne Bright & Michael Coble, FORENSIC DNA PROFILING: A PRACTICAL GUIDE TO ASSIGNING LIKELIHOOD RATIOS 32 (CRC Press 2020). This case presents a textbook example of the prosecutor's fallacy: As the District Court noted, the government stated in a brief below that the DNA results were a "finding that the defendant's and Doe's DNA were contained in" the bedspread stains at issue, when in fact those statements were the *proposition*, and the LR could not and did not evaluate the truth of the proposition itself. *See* A1058.

B. Known Unknowns: DNA Transfer, Persistence, Prevalence and Recovery (TPPR) Studies Provide a Glimpse into the DNA All Around Us, but Further Study is Required for Use in Casework.

There is a widely-cited aphorism in forensic science, attributed to the French criminologist Edmond Locard, that "every contact leaves a trace." *NIST Mixture* 106. While that principle has long been a tenet in the field, it has never been more important – nor, in a literal sense, more demonstrably true – than it is today. Scientists and researchers have undertaken in-depth experiments to understand the dynamics of DNA in the environment. Of course, for a DNA analyst set to testify in a criminal trial, this presents a daunting challenge. Having collected a DNA sample, analyzed it, interpreted the results and produced an LR, the question often remains:

How and when did the DNA come to be there? In other words, what used to be the endpoint in the testimony of a forensic DNA analyst is now merely the midpoint.

In a landmark 1997 article, researchers showed for the first time that forensic DNA analysis could develop a person's "DNA Fingerprints from Fingerprints." R. van Oorschot, *DNA Fingerprints From Fingerprints*, 387 Nature 767 (1997). The article demonstrated that not only did the simple act of touching a surface or an object leave behind biological material containing a person's DNA profile, but forensic DNA technology was sensitive enough to analyze that material and develop a DNA profile from a sample that was invisible to the naked eye. *See id*. Since that seminal article, research on DNA transfer, persistence, prevalence and recovery (TPPR) has exploded.

As *amici* discuss in greater detail below, this emerging body of scientific research offers numerous insights directly relevant to the analysis in this case. DNA transfers readily to cloth surfaces, like a bedspread, and can remain there for nine months or more. That is especially true in a person's home, where his or her DNA is expected to be prevalent throughout, but especially on items that are used frequently. Moreover, research has shown that caution must be exercised when attempting to infer how DNA was deposited from DNA testing results: The person who contributed the most DNA is not necessarily the last person to come into contact with a surface or the person who did so the most, nor do those characteristics

illuminate when the surface was last contacted. Finally, a high-template DNA mixture does not always indicate the presence of a DNA-rich bodily fluid.

DNA transfer refers broadly to the movement of DNA-containing biological material from one place to another. A person's DNA can be transferred via primary DNA transfer, or 'direct' transfer, by any form of contact: bleeding onto clothing or an adjacent surface after an injury, touching another person or surface with a bare hand or wearing an item of clothing onto which DNA adheres. R. van Oorschot et al., *DNA Transfer in Forensic Science: A Review*, 38 Forensic Sci. Int'l: Genetics 140 (2019) [hereinafter van Oorschot 2019]. Secondary DNA transfer, or 'indirect' transfer, refers to the transfer of DNA from an object or person to another via an intermediary. F. Sessa et al., *Indirect DNA Transfer and Forensic Implications: A Literature Review*, 14 Genes 2153 (2023). In other words, if DNA is deposited via direct transfer onto a surface, and is then transferred onto the fingertip of the next person to touch that surface, a secondary DNA transfer has occurred. A single contact event can act as a vector for both primary and secondary transfer – as when Person A grasps a knife handle and thereby transfers onto it both her own DNA and that of Person B, with whom she recently shook hands – and in both directions, insofar as Person A can initiate primary and second transfer simultaneously and receive such DNA transfer as well. *See* van Oorschot (2019) 141-43.

Several factors are known to impact how much transfer occurs in a given circumstance. The substrate – of both the surface where the DNA resides and the surface it comes in contact with – impacts the amount and likelihood of transfer. More DNA will be transferred onto porous surfaces like wood and fabric, while less DNA will generally be transferred from a porous surface to the contacting surface. The opposite is true for non-porous surfaces like metal or glass, which tend to carry less transfer DNA after being contacted but from which DNA is more likely to transfer. *Id.* at 145.

Both the type of biological material and the manner of contact also play a role. Biological materials that are wet or in liquid form, such as blood or saliva, transfer more readily than dried deposits, but this difference diminishes as drying occurs. *See id.* at 146. When contact takes place, increased pressure and friction tend to result in increased transfer of biological material. *Id.* at 147.

Innate differences from person to person also appear to matter. Every person sheds DNA naturally into the environment, and studies have suggested that a person's individual "shedder" status – his or her propensity to leave DNA behind when touching something – is a significant factor impacting DNA transfer. L. Jansson et al., *Assessing the Consistency of Shedder Status Under Various Experimental Conditions,* 69 Forensic Sci. Int'l: Genetics 103002 (2024). Research continues into shedding and uncertainty remains about the factors that impact how

much DNA a person will shed, such as skin conditions, age and personal habits. M. Goray et al., *Emerging Use of Air eDNA and Its Application to Forensic Investigations – A Review*, 45 Electrophoresis 916 (2024); C. Fantinato et al., *Non-self DNA on the Neck: a 24 Hours Time-Course Study*, 57 Forensic Sci. Int'l: Genetics 102661 (2022) (noting that children tend to transfer for more DNA than adults). In the end, "[t]he amount of DNA transferred and retained on an item is highly variable between individuals and even within the same individual between replicates." J. Butler, *Recent Advances in Forensic Biology and Forensic DNA Typing: INTERPOL Review 2019-2022*, 6 Forensic Sci. Int'l: Synergy 100311 (2023).

However, DNA transfer is not limited to surfaces that a person comes into physical contact with. Coughing and sneezing is known to transfer DNA over a large area. G. Meakin et al., *DNA Transfer: Review and Implications for Casework*, 7 Forensic Sci. Int'l: Genetics 434, 435 (2013). Studies have also demonstrated that merely speaking can spread DNA to the area in front of the speaker in as little as 30 seconds. N. Port et al., *How Long Does it Take a Static Speaking Individual to Contaminate the Immediate Environment?*, 2 Forens. Sci., Med. & Pathology 157 (2006).

Research has shown, however, that very little can be inferred about the mode of transfer from the properties of the DNA profile obtained. To be more specific, it

is not possible to infer that the major donor to a DNA mixture – that is, the person who contributed DNA in the greatest quantity – touched it the *most*, or touched it most *recently* or even touched it at all. *See* R. van Oorschot et al., *Persistence of DNA Deposited by the Original User on Objects After Subsequent Use by a Second Person*, 8 Forensic Sci. Int'l: Genetics 219 (2014) (major contributor is not always last person to come in contact with item); K. Atkinson et al, *Transfer and Persistence of DNA on Items Routinely Encountered in Forensic Casework Following Habitual and Short-Duration One-Time Use*, 60 Forensic Sci. Int'l: Genetics 102737 (2022); D. Taylor et al., *Observations of DNA Transfer Within an Operational Forensic Biology Laboratory*, 23 Forensic Sci. Int'l: Genetics 33, 43 (2016) ("[T]he individual that last touches an item is not always going to deposit the predominance of DNA on that item."). Similarly, although bodily fluids like semen and saliva are DNA-rich, researchers have cautioned against inferring that a DNA result showing a high quantity of DNA came from a bodily fluid over another source, such as skin cell deposits. *See* G. Meakin et al., *Evaluating Forensic DNA Evidence: Connecting the Dots*, 3 *WIREs Forensic Sci.* e1404 (2021) ("[I]t is not as simple as concluding that the DNA came from a body fluid when the amount of DNA is 'high' and casework scientists should exercise caution when considering this approach to evaluations given source level propositions."); Butler, *Recent Advances*, *supra*, at 13-14 (same).

DNA persistence is the extent to which DNA remains on a surface over time. Biological samples degrade over time, and environmental factors such as temperature, sunlight, humidity and exposure to the outdoors negatively impact how long DNA will remain detectable on a surface. However, DNA samples – including some samples thought to have been deposited by touch – have been recovered from surfaces years, or even decades, after deposition. Van Oorschot (2019) 151. Substrate is also a significant factor, and DNA can persist on porous surfaces like cloth for an extended period of time. T. Kaesler, et al., *Persistence of Touch DNA on Commonly Encountered Substrates in Different Storage Conditions*, 348 Forensic Sci. Int'l 111728 (2023) (finding full DNA profiles on cotton fabric at nine months after deposition in majority of samples tested). Finally, certain biological materials are known to persist, including semen, which can persist on fabric for years. *See* A498-99.

The more a person interacts with an area or surface, the more his or her DNA appears to persist there. Humans constantly shed their biological material into the environment, such that the DNA of the occupant of a home or office can be detected in common household dust for years after they were last there. C. Fantinato et al., *The Invisible Witness: Air and Dust as DNA Evidence of Human Occupancy in Indoor Premises*, 13 Sci. Reports 19059 (2023) (reporting ability to detect an office occupant after two years from dust sample on top of mirror).

DNA prevalence flows from transfer and persistence: Because DNA is known to transfer readily in the environment and under the right conditions can persist indefinitely, it is "reasonable to assume that most surfaces and items, unless new or cleaned, will have some DNA on them that has been acquired from previous use." Van Oorschot (2019) 148. Conceptually, this can take on two forms. Background DNA is present but not tied to a known source or activity, while prevalent DNA (often one's own DNA) is from a known source and a known activity in a way that is presumed or expected. Gill et al., *DNA Commission of the International Society for Forensic Genetics: Assessing the value of forensic biological evidence—Guidelines highlighting the importance of propositions. Part II: Evaluation of biological traces considering activity level propositions*, 44 Forensic Sci. Int'l: Genetics 102186, at 18 (2020) [hereinafter Gill, *ISFG Part II*].

The dynamics of prevalence and persistence together mean that surfaces in a home or workplace will harbor DNA from the occupants, clothing will retain the DNA of the wearer, and so on. *See* B. Szkuta, et. al., *The presence of background DNA on Common Entry Points to Homes*, 7 Forensic Sci. Int'l: Genetics Supplement Series 784, 785 (2019) (concluding individuals living together are each likely to be sources of background DNA in the home); J. Burrill et al., *A Review of Trace 'Touch DNA' Deposits: Variability Factors and an Exploration of Cellular Composition*, 39 Forensic Sci. Int'l: Genetics 8, 11 (2019) (detecting non-self DNA on many

everyday objects and environments, including on clothing and in forensic laboratories).

Finally, **DNA recovery** is dependent on accurately identifying an area where probative DNA has been deposited, efficiently sampling the targeted area and the overall effectiveness of the DNA testing process to produce a result. While studies have shown that the amount of DNA recovered varies according to the sampling methodology and the overall DNA analysis process, but this area has received relatively little attention overall. Van Oorschot (2019) 156.

Despite all the knowledge garnered from transfer studies, testimony about transfer issues in court has remained inconsistent and unscientific. Although research on different variables affecting DNA TPPR has mushroomed over the last two decades, the body of research overall is not harmonized (the studies lack standard research design principles, have different limitations and assumptions, employ different test kits and methodologies, etc.), and is therefore of limited use when applied to real cases. There remains a need to educate forensic analysts on how to apply existing DNA-TPPR data and published research to casework samples to evaluate biological findings. *See NIST Human Factors* 172-81.

C. The Scientific Community's Response To "How?" And "When?": Evaluations Given Activity-Level Propositions.

"It is a principle of forensic science that results only have meaning in context." *NIST Mixture* 145. Forensic scientists have taken steps to implement a new

framework to evaluate not only the physical properties of a DNA sample but its relevance within an investigation.

The shift in focus toward context and relevance led directly to the development of the Case Assessment and Interpretation (CAI) method, also known as evaluative reporting, to "ensur[e] that case context is considered when evaluating evidence." *NIST Mixture* 146. Within the CAI framework is an analytical tool, known as the hierarchy of propositions, that is intended "to organize questions about the evidence from general to specific, so the forensics findings can be interpreted at the correct level of meaning and in context." R. Cook et al., *A Hierarchy of Propositions: Deciding Which Level to Address in Casework*, 38 Sci. & Just. 231 (1998). The hierarchy is often represented as a pyramid:

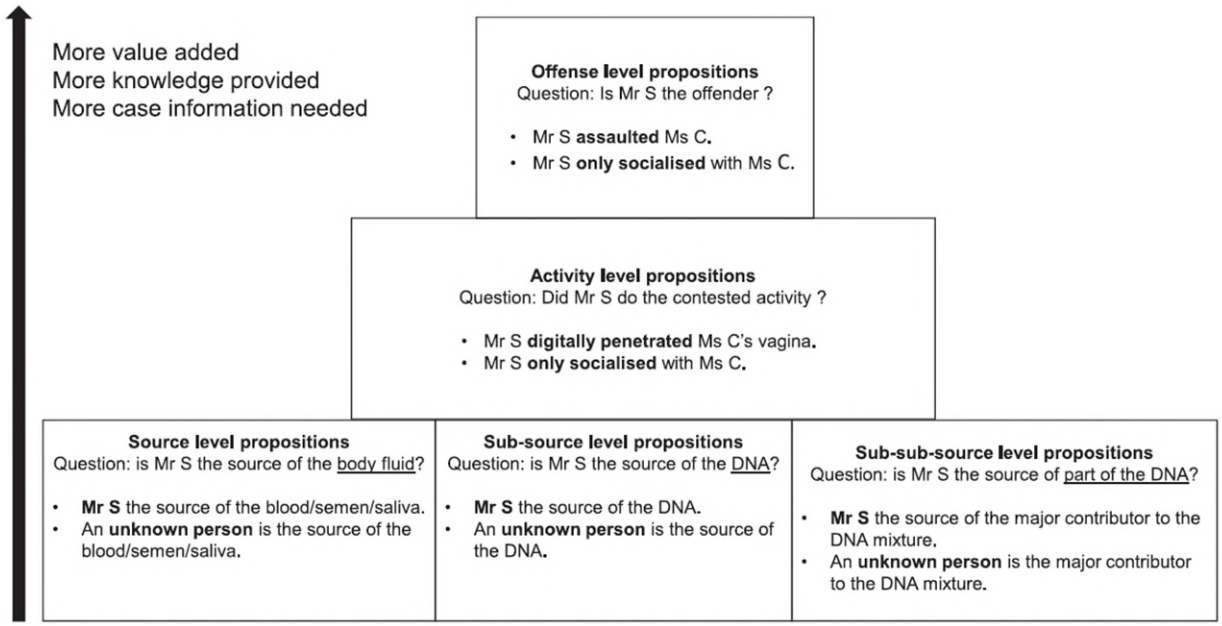

Fig. 1 Examples of pairs of mutually exclusive propositions at different level in the hierarchy of propositions. One should remember that case information would also be given, so that propositions remain snappy.

T. Hicks et al., *DNA Interpretation and Evaluative Reporting*, in ENCYCLOPEDIA OF FORENSIC SCIENCES 91-102 (3d ed. 2023). Sub-source level propositions inquire whether a particular person is the source of the DNA. Source level propositions consider the source of DNA at the cellular level, most often pertaining to a bodily fluid: does the DNA derive from a particular person's blood, semen or saliva? Finally, activity level propositions refer to the nature of the activities that led to the deposit of biological material. *See id*.

One of the primary dangers that the hierarchy of propositions is designed to mitigate is the risk that decision-makers, especially juries, will conflate the questions addressed by (sub-)source-level propositions and those addressed by an activity-level propositions. Pioneering forensic geneticist Peter Gill has highlighted the specific problem, which he refers to as the "association fallacy," where "a probability is transposed from one level of the framework of propositions to a higher level." Peter Gill, MISLEADING DNA EVIDENCE: REASONS FOR MISCARRIAGES OF JUSTICE 162 (2014). In essence, Gill explains, this error occurs when it is "assumed that there is a dependency between two observations or events," such as when evidence of the presence of a person's DNA profile is wrongly construed as evidence of the presence of a specific bodily fluid like semen or saliva. *See id.* An even greater concern is that, "without sufficient guidance from the scientist, the court may simply

carry-over the value of evidence of the DNA profile regarding its source to the 'activity' that led to the DNA transfer." Gill, *ISFG Part II* at 2.

Evaluations given activity-level propositions are among the most discussed topics in the current forensic science literature. As with sub-source evaluations, the evaluator must set mutually-exclusive propositions for the activities that led to the DNA deposit, but many more factors must be taken into account, including "the user history of items, the nature of the questioned activities and prior interactions between persons-of-interest and with their environments, [and] also details on collecting items at the scene, packaging and transport, and analysis methods." Duncan Taylor & Bas Kokshoorn, FORENSIC DNA TRACE EVIDENCE INTERPRETATION: ACTIVITY LEVEL PROPOSITIONS AND EVALUATIONS 95 (CRC Press 2023) [hereinafter Taylor & Kokshoorn].

Once the propositions are set, the next step is to gather relevant information and make certain assumptions to inform the evaluation. For instance, if it is unknown whether an assailant wore gloves, an assumption is needed, since glove use impacts the probability of DNA transferring to the complainant or another surface. *Id.* at 110. Undisputed contextual information must also be considered, such as (in the example above) the cohabitation of the complainant and the assailant, as they would be expected to harbor traces of one another's DNA. *Id.* at 111. If contextual facts

bearing on TPPR are in dispute, then the alternative versions of those contextual facts must be built into the propositions. *Id*.

The final step, in light of the propositions in the case and the framing lent by relevant assumptions and contextual facts, is to identify potential pathways for DNA transfer, translate those pathways into an LR and assign probabilities to each node in the resulting equation. *Id*. at 124-26. There are multiple methods to assign probabilities to discrete steps in the evaluation, from a bespoke experiment designed to approximate the case context and produce data that is closely aligned to it, to assignment of a reasonable value based on the expert's "experience or knowledge." *Id*. at 169.

Despite the academic interest in activity-level evaluations, there is a clear lack of data, expertise and training to support its implementation in the United States. *NIST Mixture* 148 ("There are many references in the literature to the suitability of this approach but little in the way of prescriptive assistance."). The knowledge and expertise to perform such evaluations represents a skillset that is "distinct from 'standard' DNA profiling and interpretation," and at present "there are not adequate educational opportunities to inform these types of issues within the United States." *NIST Human Factors* 173-74. Moreover, in a forthcoming article, a Senior Policy Advisor to the FBI laboratory raises questions about the admissibility of evaluations given activity-level propositions in American courts. Ted R. Hunt, *Activity Level*

Testimony in U.S. Courts: A Legal Problem, 49 U. KAN. L. REV., Volume 74 (forthcoming 2025), https://ssrn.com/abstract=5233773 (challenges include limited or lack of data on TPPR probabilities, burden of proof issues, and difficulty with obtaining case context information for the alternative hypothesis in the U.S. legal system).

Despite these shortcomings, it is difficult to overstate the importance of the widespread acknowledgement in forensic science of the empirical superiority of a true activity-level analysis. Indeed, as the International Society for Forensic Genetics advised, while "[i]t might be tempting for scientists to report a high DNA statistic and then explain their results in court . . ., this can be misleading, [and] we urge scientists not to give explanations in court." Gill, *ISFG Part II* at 11.

II. THE DISTRICT COURT CORRECTLY CONCLUDED THAT THE STAIN EVIDENCE ON THE COMFORTER WAS NOT RELEVANT, AND EVEN IF IT WAS, THAT THE POTENTIAL FOR UNFAIR PREJUDICE FAR EXCEEDED ANY PROBATIVE VALUE.

There is a stark disconnect between the government's position on the probative value of the evidence in this case and the statements of its own experts. The District Court's decision to act as the gatekeeper of this information, with such a high risk of misleading the jury, was correct.

Amici curiae rely on the District Court's detailed description of the forensic testing, but summarize certain aspects here. Soon after receiving the comforter that

was DNA tested in this case, a United States Army Criminal Investigation Laboratory ("USACIL") case manager noted that the presence of related individuals in the household would significantly "complicate" the analysis and requested reference samples from all of the children who lived there (including the two biological children of Tyler and Monica who slept in the bed each night). Those samples were never provided. A1002. USACIL Forensic Biologist Sara Green conducted an initial examination of the bedspread and found 45 stains that fluoresced under ultraviolet light, indicating the likely presence of biological material. Green testified that six of the 45 stains tested positive for both acid phosphatase and Prostate Specific Antigen (PSA), which are presumptive tests for semen. A1004.

Green's Report documents her analysis on those six DNA mixtures from the bedspread. Five were deemed three-person mixtures, and one was deemed a four-person mixture. Using STRmix, a probabilistic genotyping software program, Green calculated LRs comparing two propositions: that Tyler Johnston, Monica Johnston and Jane Doe contributed DNA to the mixture, versus that Monica Johnson and two unknown, unrelated individuals contributed DNA to the mixture (in the case of the four-person mixture, adding an additional unknown in each proposition), and the resulting LR for each sample was "at least one quintillion times more likely" if the former were true than if the latter were true. A441, 445-46. Green was unable to test for related contributors in her analysis because the lab never received reference

samples for the other Johnston children, and thus could not condition on them; and also could not test for related contributors using the specialized feature in STRmix that is designed to do so, because the lab had not tested that feature in its own casework and therefore was not validated to use it. A1008-09. Additionally, both Green and a second forensic DNA expert for the government, Professor Tim Kalafut, agreed that the DNA results do not indicate how, when or in what order the DNA got on the bedspread. A496 (Green); A842-43 (Kalafut); *see* A1054-55.

As an initial matter, the LR in this case is meaningless because the propositions that it evaluates are meaningless. The lab used alternate propositions – that unknown and unrelated persons contributed DNA to the stains on the Johnston's bedspread – that bear no relation to the facts of this case. Indeed, neither the lab itself nor Mr. Johnston contends that the alternate proposition should be premised on the bedspread stain coming from an unknown, unrelated individual. A1009 (Green testified that she was "limited by the references [she] received"); *see* Def.'s Br. at 44. While the use of an unknown and unrelated person in the alternative proposition may be appropriate in certain cases, the comparison population "depends on the case information" and "should consider close relatives" where, as here, the context requires it. Gill, *ISFG Part I* at 198. Nor does the magnitude of the LR carry any true meaning, as "[d]isregarding a plausible close relative . . . as an alternative contributor may overestimate the LR" as to the person of interest. G. Dorum et al., *Likelihood*

Ratios for Complex Mixtures with Relatives, 4 Forensic Sci. Int'l: Genetics Supplement Series e61 (2013).

Even setting aside these fundamental flaws in the LR, a true understanding of the relevance of the DNA results in this case would require an evaluation of a series of contextual questions. First, the bedspread should be assumed to harbor prevalent DNA *from all six members of the Johnston family*. Four members of the household (Mr. Johnson, Monica Johnston and the two younger children) slept in the bed *every night*, while Jane Doe and her half-brother T.J. spent time on it periodically. A989-90. At a minimum, as the District Court noted, A1004, a substrate control swabbing of the bedspread – that is, swabs from various unstained areas of the bedspread – would have provided valuable information on the donor and quantity of DNA that could reasonably be expected to be present on any area of the bedspread as background DNA. *See* A0495-96. Additionally, the shedder status of each member of the family is an important consideration, as the DNA deposited by a good shedder can "swamp" that of a poor or intermediate shedder when the DNA is recovered. *See* van Oorschot (2019) 147.

The government's deeply problematic insistence that Doe's DNA was "mixed with Johnston's semen" likewise takes the DNA results entirely out of context by implying that the two elements were deposited together. As an initial matter, the record does not indicate when the bedspread was last washed, and semen is known

to persist for months or years after being deposited on fabric. The bedspread, in other words, could retain traces of the activities that occurred on it – sex between Monica and Tyler, a bed-wetting incident, tablet use, general play and hundreds of sneezes, coughs and runny noses – for months or even years beforehand. Moreover, even assuming *arguendo* that Doe, Tyler and Monica each contributed DNA, there is absolutely nothing in the record that can speak to when or in what order any of those individual contributions occurred. And the order of deposition is critically important: if the government contends that the DNA deposits of Doe and Mr. Johnston derive from a sexual assault, what explains the apparent presence of Monica Johnston's DNA "mixed with" theirs in all six samples? The government's proposed inference simply ignores Monica's DNA in favor of its own cherry-picked narrative.

To be clear, these are not questions that science can currently answer. As the District Court noted, each of the experts in the case acknowledged these limits and testified accordingly. A1054. Even the government acknowledged that its experts cannot "speak to how this DNA was deposited." A0886. But the government argues that "[e]vidence of Doe's DNA mixed with Johnston's semen on Johnston's bed where Doe alleged he raped her is highly probative of whether Johnston sexually abused Doe." Gov. Appeal Br. at 63; *see also* A1052 ("If a child's DNA is mixed with a defendant's semen, I think it's a fair basis to say it is more likely than not – there's a reason they mixed. And the government submits that reason is he was

sexually assaulting her in that location."). The government is, in other words, inviting the jury to commit the association fallacy, *see supra*, by conflating the sub-source LR for an activity-level conclusion.

The government's position is untenable because context matters. Scientists have recognized for decades that "a probability statement has little meaning unless it includes at least some indication of the information, knowledge and assumptions on which it is based." I.W. Evett, *Avoiding the Transposed Conditional*, 35 Sci. & Just. 127, 128 (1995). Setting aside the fact that its expert has declined to even provide a probabilistic assessment as to how the DNA on the bedspread came to be there, the government treats this foundational requirement as a mere speedbump. Because the government's proposed inference is devoid of scientific support, the District Court's ruling that the evidence is not relevant under Rule 401 should be affirmed.

However, assuming *arguendo* that the LRs – which did not adequately account for a closed universe of biological relatives as potential donors of DNA on a household item in a family home – were to carry some modicum of relevance, it is clear that any "probative value would be substantially outweighed by the risk that it would both mislead and confuse the jury." A1056. Research shows that laypeople often struggle to understand the LR and accurately interpret its meaning in their decision-making. *See e.g.*, W.C. Thompson et al., *Lay Understanding of Forensic*

Statistics: Evaluation of Random Match Probabilities, Likelihood Ratios, and Verbal Equivalents, 39 LAW & HUM. BEHAV. 332 (2015) (finding two-third of participants treated the LR as probability of guilt rather than a measure of evidentiary strength and observing these fallacious interpretations were strongly associated with verdicts); W.C. Thompson et al., *Does Explaining the Meaning of Likelihood Ratios Improve Lay Understanding?*, 65 Sci. & Just. 101352 (2025) (providing explanations of the meaning of the LR did not improve comprehension).

Here, the government's experts will introduce LR values that are not only inflated by their reliance on incorrect propositions, but that also do not address the relevant inquiry in this case: how, when, and under what circumstances the DNA from any individual got onto the comforter. Indeed, Green admitted that "how [the DNA] got onto the comforter is not anything that I could tell." A496. Expert assistance is required to help the jurors understand the meaning of the biological findings in light of the alleged activities, yet the government's experts concede they cannot provide such guidance at the activity level concerning the comforter. Because the jury cannot perform these evaluations on their own, they will necessarily speculate about the significance of the observed profiles relative to the activities alleged. Such speculation renders the evidence more misleading than probative.

Prominent forensic scientists agree with the District Court's position. The prudent course of action is not to report the sub-source level propositions when an

evaluation given activity-level propositions cannot be provided to assist the trier of fact. When an expert cannot provide an activity-level assessment, the expert should ordinarily also decline to provide an evaluation at the sub-source level because the latter "will bear the risk of being inappropriately carried over to a higher propositional level by recipients of expert information without properly acknowledging factors (such as transfer and persistence) that require expert knowledge." D. Taylor et al., *Evaluation of Forensic Genetics Findings Given Activity Level Propositions: A Review*, 36 Forensic Sci. Int'l: Genetics 34, 41 (2018).

Without expert guidance on how to evaluate these biological findings, the jury would be left to speculation and improper inferences that go beyond what the testimony can establish about the presence of semen and Doe. Courts have recognized the danger to defendants of mischaracterized and/or misunderstood expert testimony on DNA evidence. *See, e.g.*, *People v. Wright*, 25 N.Y.3d 769, 783-84 (2015). Presenting an unreliable and artificially inflated sub-source LR value without activity-level analysis in the context of this case would undermine Mr. Johnston's constitutional right to a fair trial, as it invites the jury to commit the same "association fallacy" that has led to wrongful convictions and misleading scientific testimony in other cases[2]: that the presence of semen and Doe's DNA must mean a

[2] *See, e.g.*, P. Gill, *DNA Evidence and Miscarriages of Justice*, 294 Forensic Sci. Int'l e1 (2019); Taylor, *Activity-Level Review*, *supra*.

specific criminal activity occurred. Given the clear prejudice that would result from introduction of this DNA evidence, the District Court properly concluded that the potential for prejudice outweighed its probative value and excluded the DNA evidence.

CONCLUSION

For these reasons, *amici curiae* NACDL, NYSACDL and The Legal Aid Society urge this Court to affirm the District Court's order.

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Respectfully Submitted,

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CERTIFICATE OF COMPLIANCE

I certify that this brief complies with Federal Rule of Appellate Procedure 32(a)(7)(B) because it contains 6,885 words, excluding the portions of the brief exempted by Rule 32(f). I further certify that this brief complies with the typeface and type-style requirements of Rule 32(a)(5)–(6) because it is printed in a proportionally spaced 14-point font, Times New Roman.

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