

A novel approach to forensic molecular biology education and training: it's impact on the criminal justice system

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The managers of crime laboratories face significant hurdles when preparing new hires to become productive members of the laboratory. New hires require six months of training/experience in the crime laboratory before becoming a productive member of the Biology (DNA) section. To address this deficiency in forensic DNA education, a novel forensic education curriculum was developed and tested for three consecutive years in the forensic science program at Fayetteville State University, Fayetteville, NC. The curriculum used a CTS proficiency kit, which is the same kit used to validate the proficiency of forensic scientists in crime laboratories in the US. A cost benefit analysis suggests that training students in a classroom instead of in a crime laboratory provides both direct savings to the laboratory and significant societal savings as more DNA profiles are entered into the database. The societal benefit from the combined reduction in the amount of training in a crime laboratory and increasing the number of DNA database profiles entered into a database suggests a societal saving of \$8.28 million for each of these months of reduced training.

Keywords: DNA; training; cost benefit; education; forensic; crime laboratory

Introduction

Over the past decade there has been significant growth in the demand for forensic laboratory services while the growth in capacity has developed at a much smaller pace¹. In no other investigative area has that gap between services requested and services rendered been more severe than forensic biology analysis, even in the face of dramatic funding increases for backlog reduction. There is a huge need for trained forensic biology analysts to tackle the backlog of Deoxyribose Nucleic Acid (DNA) evidentiary items. Statistical analysis of the 2005 Census of Publicly Funded Forensic Crime Laboratories determined that crime laboratories would need to increase the number of DNA analysts by 73% to achieve a 30 day turnaround time for analyzing DNA evidentiary items^{2,3}. Results from the 2009 Census of Publicly Funded Crime Laboratories suggests only a modest reduction in the backlog following the large influx of funding from the DNA Backlog Reduction Program⁴. 'Without a commitment to find permanent funding solutions for crime laboratories, it is likely that laboratory dependence on federal grants will continue'⁴. This is a challenging demand for the forensic

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crime laboratories due to lack of skilled manpower and an increase in demand for DNA analysis of evidentiary items.

Reports in the literature reveal the staffing shortage in forensic laboratories. It has been suggested that the staffing demands, driven largely by DNA analysis, calls for as many as 10,000 more forensic scientists in the United States alone if backlogs are to be erased⁵. More recent inquiries into the efficiency of forensic science delivery suggest that this figure may be somewhat lower if resources are dedicated to take advantage of economies of scale^{6,7}. Yet, even with efficiency adjustments, the staffing shortage remains severe and novel solutions for greater efficiency should be considered.

The pool of recent forensic science graduates is large as evidenced by 138 forensic science undergraduate, 59 graduate, and six doctoral degree programs in the US⁸. So the problem is not lack of interest among students; students are pursuing careers in forensic biology. Popular television programs such as CSI have made careers in forensic biology attractive even though the Hollywood depiction of a forensic investigation is not realistic.

The problem is that recent forensic science graduates lack the skills necessary to begin working in forensic laboratory without significant additional forensic training at crime laboratories. The educational deficiency in new forensic science graduates is a reflection of the forensic science curriculum currently being taught in colleges and universities⁸. In traditional science programs, such as biology or chemistry, the curriculum has been well established and thoroughly tested. The same cannot be said of forensic science, which is a relatively new scientific discipline.

The educational basis of the undergraduate/graduate forensic science programs varies widely. Some programs are taught within criminal justice departments and others are taught in biology or chemistry departments⁸. It is generally accepted that forensic science programs taught with a strong foundation in natural sciences best prepares students for a career in forensic science. The natural sciences foundation is essential for the forensic science profession. These forensic science programs focus on a typical undergraduate biology or chemistry curriculum concentration with a few specialized courses in biology/chemistry forensics. However, these biology/chemistry forensic courses are not adequately preparing graduates for a career in forensics⁸. We propose a forensic biology curriculum for students majoring in Forensic Biology (123 credit hours) for the Bachelor of Science in Forensic Biology, preparing students for careers as forensic scientists in a crime laboratory. Students enrolled in this program are required to satisfactorily complete eight courses (38 credit hours) in forensic biology with the following topics: evidence collection, documentation, chain of custody, forensic serology, forensic DNA analysis, Y-STR analysis, mitochondrial DNA analysis, microscopy analysis of trace evidence including hair, fiber, soil, paint, ethics, expert testimony, technical and scientific report writing, and other related topics. A complete description of these courses can be found in the supplemental materials, available online at <http://dx.doi.org/10.1080/00450618.2014.925974>.

Forensic scientists require specialized forensic science education that is not taught in a typical biology or chemistry course⁸. Forensic science courses are typically developed and taught by natural sciences faculties who have little if any forensic science expertise. Forensic science courses should be presented with the scientific/legal investigative methodology of forensic science and, most importantly, focus on the needs of forensic laboratories. The DNA Advisory Board (DAB) quality assurance standard 5.3.2 states 'minimum of six (6) months of forensic DNA laboratory experience, including the successful analysis of a range of samples typically encountered in forensic case work prior to independent case work analysis using DNA technology'⁹.

However, it is typical for new hires with no prior forensic DNA laboratory experience to receive specialized training that is completed in 6 months to 2 years depending upon the forensic agency and forensic specialty¹⁰. Consider, instead, a forensic science curriculum for serological and DNA forensics that closely follows the specialized training of new hires in the forensic laboratories. Students who successfully complete such a curriculum will be better prepared for working in the forensic laboratory. Forensic laboratory training will be reduced for new hires and these new graduates can become productive forensic scientists sooner than has been the experience to date. In addition, the training of new forensic biology scientists' financial burden on the crime laboratories can be reduced.

Consider the use of the Collaborative Testing Services (CTS) proficiency test kit for the training of forensic students. The CTS kits are used by forensic crime laboratories all over the US to evaluate forensic scientists to ensure that their technical skills are current. Crime laboratories use the results of the CTS kit(s) to maintain accreditation and satisfy federal regulations.

Materials and methods

A novel forensic biology course (four hundred level) was developed and tested at Fayetteville State University, Fayetteville, NC. The course was 16 weeks meeting twice a week for 3 h each period and taught in a hands-on laboratory. Prior to enrolling in this course students completed two consecutive courses (16 weeks) in Forensic Serology and Forensic DNA. These courses were hybrid courses with a lecture and laboratory component to teach both Forensic theory and laboratory skills. The required forensic DNA training equipment and forensic expertise was available at the Forensic program at FSU, and therefore the department of Natural Sciences (currently the department of Biological Sciences) decided to offer the Forensic Internship in-house at FSU. The nearest crime laboratory was too far away for students to commute daily for 16 weeks while enrolled in other courses. In addition, the crime laboratory will not allow students to handle any evidentiary item(s) or to perform any forensic analysis on crime laboratory equipment. The course provided students with a realistic forensic DNA experience that otherwise could only be obtained in a forensic crime lab. The course was taught three consecutive years to undergraduate students majoring in forensic biology.

Students were given the 2007 CTS Proficiency Forensic Biology kit (Collaborative Testing Services, Sterling, VA), the test kits used in this study were kindly donated by CTS. A rape crime scene scenario is included in CTS kit. The proficiency kit (see Figure 1) contains two known blood stains: item 1 (known blood from the victim) and item 2 (known blood from a suspect) on Whatman FTA Gene Cards. In addition, there are two suspected dried body fluid stains: item 3 (stain from victim's skirt) and item 4 (stain from suspect's shirt). A positive control (known blood sample on clean cloth) was included as a quality control measure (see Figure 1) by the instructor. Adding a positive control is common practice in the forensic laboratory. Following the crime laboratory biology section guidelines, the students recorded the measurements of the evidentiary items and stains (see Figure 1). The known blood stains, suspected dried body fluid stains and positive control were cut with scissors into approximately 2 mm pieces for DNA analysis.



Figure 1. Contents of Collaborative Testing Services (CTS) proficiency test kit: item 1 (victim's blood), item 2 (suspect's blood), item 3 (stain from victim's skirt), item 4 (stain from suspect's shirt), and positive control (blood).

DNA extraction

DNA was extracted from the cut samples of items 1, 2, 3, 4, and a positive control. All the samples were tested in triplicates. All the samples and the reagent blank were soaked in 500 μ L of digest buffer (10 mM Tris-HCl, 10 mM EDTA, 50 mM NaCl, 2% SDS and 20 μ g/mL proteinase K) and incubated at 56°C for a minimum of three hours. As a quality control measure, a reagent blank was included. Subsequently, DNA was extracted from each sample using phenol: chloroform: isoamyl alcohol (25:24:1)¹¹. The aqueous phase of the DNA extraction was transferred to an Amicon Ultra-4 Centrifugal Filter Device (Millipore, Billerica, MA) for DNA concentration, and the manufacturer's recommended protocol was followed. The tube contents were washed twice with TE (10 mM Tris, 10 mM EDTA, pH 8) buffer pH 7 with centrifugation at 5000 g for 15 min. The recovered DNA was stored at -20°C.

Differential DNA extraction

Differential extraction was employed to isolate male DNA (sperm) and female DNA (epithelial cells). As per a rape crime scenario, a sample was cut from item 3, suspected of having sperm and female epithelial cells. The sample was soaked in 500 μ L of 10 mM Tris-HCl, 10 mM EDTA, 50 mM NaCl, 20% Sarkosyl and 20 μ g/mL Proteinase K at 37°C for 30 min. Note, the DNA extraction buffer preferentially extracts DNA from epithelial cells. The DNA extract was centrifuged at 15,000 g for 15 min to separate epithelial cells DNA (female fraction) from spermatozoa. The sperm pellet was washed, re-pelleted and re-suspended in 500 μ L of the DNA extraction buffer with the inclusion of 0.39 M Dithiothreitol (DTT) at 37°C overnight¹². The DTT reduces the thiol bonds in the sperm acrosomes thus lysing the sperm cells. The female epithelial and sperm DNA extracts were treated according to the protocol described above (see DNA extraction). The negative controls for male and female reagent blanks (no cells), and the positive control of a sperm sample were ran parallel to test samples.

DNA quantification by real-time Polymerase Chain Reaction (PCR)

The human genomic DNA was determined using the Quantifiler[®] Human DNA Quantitation Kit (Applied Biosystems, Foster City, CA) run on an ABI 7500 Real-Time

PCR System (Applied Biosystems, Foster City, CA). Quantification standards were prepared by serial dilutions in TE pH 7 of the 200 ng/μL Quantifiler[®] Human DNA standard included in the kit. Eight serial dilutions ranging from 50 ng/μL to 0.023 ng/μL in three fold increments were used. The manufacturer's protocol was 10.5 μL of Primer Mix, 12.5 μL of PCR Reaction Mix and 2 μL of DNA extract for a total reaction volume of 25 μL. In addition, the NTC control (no DNA) was performed. The DNA samples were loaded into a MicroAmp[®] Optical 96-Well Reaction plate and sealed using Optical Adhesive Covers (Applied Biosystems, Foster City, CA). The manufacturer's recommended amplification protocol was followed: 95°C 10 min. 40 cycles of 95°C for 15 s and 60°C for 1 min.

Data collection points were recorded at all stages of the PCR reaction. Quantifiler[®] Human DNA standards were assayed in duplicate and experimental samples were assayed in triplicate. A standard curve was generated for the Quantifiler[®] Human DNA standards. Finally, IPC results from the experiment did not reveal any inhibitors.

Amplification

The Polymerase Chain Reaction (PCR) reactions were prepared according to the manufacturer's recommended protocol using the AmpFISTR[®] Identifier[™] Kit. The DNA template was 1 ng in final volume of 25 μL PCR reaction. PCR reagents were loaded into 96-Well GeneAmp PCR System 9700 (Bio-Rad) for PCR amplification. All amplification reactions were accompanied by negative and positive controls.

Detection of alleles

Following PCR amplification, the ABI PRISM[®] 310 Genetic Analyzer (Applied Biosystems) was employed for electrophoretic separation of amplified products. For ABI 310 sample preparation, 24.5 μL Hi-Di[™] Formamide (Applied Biosystems), 0.5 μL GeneScan[™] 500 LIZ[®] Size Standard (Applied Biosystems), and 1 μL of PCR amplified product or AmpFISTR[®] Identifier[™] Allelic Ladder (Applied Biosystems) were added to each sample. The reaction tubes were heated at 95°C for 3-min denaturation step, immediately snap-cooled on a freezer block for three minutes, and then subjected to capillary electrophoresis. The samples were separated on a 47 cm × 50 μm capillary tube (Applied Biosystems).

Amplified products were electrokinetically injected for 5 s and fractionated on an ABI Prism 310 Genetic Analyzer using POP4 (Applied Biosystems). Data were analyzed using a peak detection threshold of 100 relative fluorescence units (RFU) for all dyes with GeneMapper[®] ID v3.2.1 (Applied Biosystems), which was used for data collection and analysis.

Results

Students were organized into groups of two to witness the transfer of evidence. This is a standard quality control and quality assurance practice used in crime laboratories. This training has been conducted for three consecutive years. The data presented were from only one group of students. Class performance was measured by the use of course learning objectives (see Table 3). All students enrolled in the course satisfactorily demonstrated mastery of the course learning objectives.

DNA quantification

The results from DNA quantification using 7500 Real-Time PCR in conjunction with the Quantifiler Human DNA Quantitation Kit are shown in Table 1. The amount of DNA recovered from all the samples was greater than 1 ng and sufficient to proceed with the next step of DNA amplification.

Amplification

Students amplified the DNA and fractionated the PCR products following the protocol described in the materials and methods section. The GeneMapper[®] ID v3.2.1 (Applied Biosystems) was used for data collection and analysis. Allele frequencies of the 15 short tandem repeats (STRs) plus the amelogenin marker for gender identification are generated (see Figure 2). Complete DNA profiles were obtained for the reference (victim and suspect) samples, item 4 (suspect's shirt), and from the positive control (see Table 2). However, the item 3 (victim's skirt) female fraction (fr) shows X>Y at amelogenin marker, indicate the presence of male. The item 3 (victim's skirt) male fraction shows a mixed profile of at least two individuals (see Table 2).

Table 1. Table shows the volume and amount of DNA extracted from the reference samples, evidentiary items, a reagent control, Blank-female fraction (fr), Blank-male fr, male positive controls, and positive controls.

Sample	Human DNA		Total DNA Concentration (ng)	Final Vol (μL)	Final DNA Concentration (ng/μL)
	Volume (μL)	Concentration by RT-PCR (ng/μL)			
Item 1A – known victim	40	0.376	15.04	37	13.912
Item 1B – known victim	80	0.607	48.56	77	46.739
Item 1C – known victim	50	0.286	14.3	47	13.442
Item 2A – known suspect	50	0.644	32.2	47	30.268
Item 2B – known suspect	55	0.276	15.18	52	14.352
Item 2C – known suspect	40	0.338	13.52	37	12.506
Item 3A – unknown victim's skirt – female fr	50	0.499	24.95	47	23.453
Item 3A – unknown victim's skirt – male fr	40	0.230	9.2	37	8.510
Item 3B – unknown victim's skirt – female fr	50	0.737	36.85	47	34.639
Item 3B – unknown victim's skirt – male fr	35	0.270	9.45	32	8.640
Item 3C – unknown victim's skirt – female fr	55	0.274	15.07	52	14.248
Item 3C – unknown victim's skirt – male fr	45	0.300	13.5	42	12.60
Item 4A – unknown suspect's shirt	55	0.130	7.15	52	6.76
Item 4B – unknown suspect's shirt	55	0.050	2.75	52	2.6
Item 4C – unknown suspect's shirt	50	0.111	5.55	47	5.217
Reagent Blank	100	0	0	97	0
Blank – female fr	40	0	0	37	0
Blank – male fr	35	0	0	32	0
Male positive control 1	40	0.180	7.2	37	6.660
Male positive control 2	35	0.320	11.2	32	10.240
Male positive control 3	45	0.210	9.45	42	8.820
Positive control 1	50	5.290	264.5	47	248.63
Positive control 2	45	2.590	116.55	42	108.78
Positive control 3	55	0.389	21.395	52	20.228

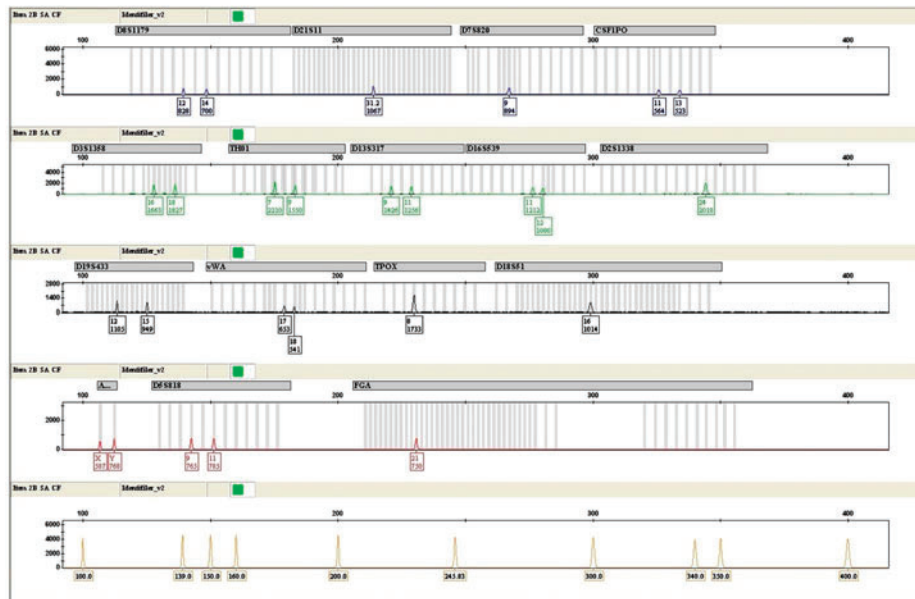


Figure 2. A representative electropherogram showing 15 STR markers plus an amelogenin marker and LIZ size standard (100 to 400) peaks showing the quality of student's work.

Discussion

The forensic molecular biology educational model described satisfies two key concerns of forensic laboratory directors/managers. First, new forensic hires are not sufficiently prepared to begin work as a forensic analyst. The Council of Forensic Science Educators stated that, 'Students completing these lesser programs expect to find employment in crime labs but are surprised to learn that lab management is not impressed by the curriculum'⁸. Second, forensic laboratories 'have to spend precious time and resources in the training of new scientists'⁸. The time and resources devoted to training a new hire would be better used reducing the DNA backlog. This is especially true considering the current economic climate.

The effective training of forensic science requires a curriculum and laboratory equipment that closely match the experiences in a crime laboratory. The laboratory equipment used for the education of the forensic science students was comparable to the equipment used by crime labs all over the world, such as the genetic analyzer, Real-Time PCR, thermocycler. In the curriculum, students first study forensic serological methods (blood identification and human blood confirmation) in a 16 week course (manuscript in preparation). This is followed by a DNA forensic course (16 weeks) that teaches students how to extract DNA, differential DNA extraction, quantification of DNA using Real-Time PCR, PCR amplification of DNA, and analysis of DNA profiles with a genetic analyzer. Upon completion of the forensic serological and forensic DNA courses, students were enrolled in a third course that tested their knowledge of serological and DNA tests using the CTS proficiency test kits.

The CTS Proficiency Forensic Biology kit was chosen because it provides students with a realistic forensic investigation. The evidentiary items in the proficiency kit

Table 3. Rubric for measuring students' performance.

	Students achieving course learning objectives
Demonstrate how to examine the evidence, its documentation, and maintain the chain of custody	100%
Apply evidence-based principles to the practice of forensic science	100%
Adherence to safety regulations, the maintenance of lab notebooks and the preparation of written reports	100%
Develop mastery in carrying out work in the laboratories. This includes skills in specific lab techniques, the proper use of equipment	100%
Demonstrate the technical skills such as data analysis, critical thinking, and scientific writings	100%
Demonstrate a sound understanding of the legal, professional and ethical framework for forensic profession	100%
Demonstrate understanding of expert testimony and its importance	Presentation in front of audience including faculty, forensic professionals and a prosecutor

include a stain with biological materials from two different contributors, a common occurrence in forensic case work. The proficiency test evaluates the quality control and quality assurance of the forensic analyst in a crime laboratory. The CTS proficiency kit is widely used in crime laboratories. The DAB requires that every biologist (DNA/serology) in the forensic crime laboratory and every DNA forensic analyst satisfactorily complete this kit after every 180 days to maintain his/her accreditation^{9,13}. In addition, the student training included quality control/assurance such as reagent blank, positive and negative controls, documentation, chain of custody, and witnessing. The students learn skills that can be applied not only to forensics but paternity testing and the DNA database as well.

Cost-benefit analysis

Roman¹⁴ argues that reform in the criminal justice system must be evaluated in terms of the marginal costs and marginal benefits of any proposed change. Those costs and benefits include the direct accounting measures as well as the opportunity costs and measures of the indirect societal impacts. Several recent studies have given rise to metrics that permit such measurement. Beginning with a project involving four European forensic laboratories¹⁵, consistent metrics have been suggested to permit the measurement of the complete costs and benefits of the forensic laboratory. More recent benchmarks are available via Project FORESIGHT¹⁶ using many of the metrics proposed by laboratories in North America¹⁷.

As noted above, the marginal costs are relatively minor in that the DNA training is included as part of their educational program of study at no additional charge to the student. That program of study includes enrollment in a third forensic biology course that tested knowledge of serological and DNA tests using the CTS proficiency test kits. As part of their program of study, the additional cost was zero from the course itself.

And since all associated equipment was already present in the lab, there were no new capital expenditures necessary to support the proficiency program. The test kits represent the additional costs of the program and represent about \$180 per kit; however, the test kits used in this study were kindly donated by CTS.

The marginal benefits are notable. These benefits include the additional net output of the laboratory from the more quickly prepared scientist who is able to replace training time with time at the bench. It also includes the return to productivity from the senior scientist who more quickly returns to full-time analysis instead of belabored training of the new hire. Most significant are the societal gains from the processing of more cases in a given period of time and the associated benefits from speedy justice.

Consider first the direct benefits to the laboratory. The benefits will vary greatly from laboratory to laboratory because of regional market forces and the laboratory's caseload with respect to economies of scale¹⁸. Project FORESIGHT metrics provide an indication of the typical laboratory productivity. Using the performance for 2011–2012, the median laboratory analyzed 2395 samples per FTE (FTE is a Full Time Equivalent employee, see the definitions in Project FORESIGHT for more detail¹⁶) in DNA Database and 320 samples per FTE in DNA Casework¹⁹. That translates into monthly production of approximately 200 samples in DNA Database and 27 samples in DNA Casework. (For DNA Database, the summary statistics from Project FORESIGHT 2011–2012 include a mean of 2471 samples, median of 2395 samples and a standard deviation of 1304 samples. For DNA Casework the mean productivity is 327 samples, median of 320 samples, and standard deviation of 143 samples.) Dale & Becker²⁰ argue that the productivity decline for the senior forensic scientist is 50% for a period that lasts at least one year. As noted above, new hires with no prior forensic DNA laboratory experience receive specialized training over a period of 6 months to 2 years. If the graduate earned DNA certification prior to hire, then the educational program can be expected to yield benefits to the laboratory for every month gained from a reduction in the training process. Over that period it translates into a laboratory saving of half the senior scientist's salary for that period and all of the newly hired scientist's salary for those months.

Consider the magnitude of those monthly savings. Worldwide salaries vary widely for every profession, including forensic sciences. Consider data for scientists in the United States. For entry level biologists, salaries range from \$36,000 at the 10% level to \$54,800 at the 90% level with a median of \$44,100²¹. In addition to salaries, the costs of associated employer-provided benefits must also be considered. Using the Project FORESIGHT 2011–2012 median benefit expenditure (42.2%), then the laboratory can be expected to gain over \$5225 per month for each month's reduction in the training period for the new hire. For a senior scientist, the national salary range goes from \$78,400 at the 10% level to \$117,600 at the 90% level with a median salary of \$101,300²². With a recovery of the senior scientist's 50% productivity loss during five months of training, that translates into the recovery of salary and benefits of over \$6000 per month from this program for the median laboratory.

The combined net monthly direct benefit of \$11,225 to the laboratory is attractive, but the largest gains from the program are the societal gains from the additional processing of DNA samples. A cost-benefit analysis of the contribution of the DNA database to the justice system indicates that there is a severe underutilization of DNA analysis. Doleac²³ offers keen insight into the overwhelming magnitude of these benefits as she measures the dramatic gap between the huge benefits from additions to the DNA database versus the relatively low cost of a new entry to the database. An

additional DNA profile entered into the DNA database has an expected result of 0.57 fewer serious offenses at a savings of \$27,600 per profile. (Doleac²³ lists the following offenses until this categorization of serious offense: murder, rape, assault, robbery, burglary, larceny, and vehicle theft.) Again using median productivity of 200 samples per FTE per month, then each additional month's productivity from the new hire and the recovery of the 50% productivity from the senior scientist translates into a combined 300 samples per month for a social gain of \$8.28 million from the reduction in training time. Of course, at the start of the career the new hire's productivity may well be less than the median scientist; likewise, the senior scientist is likely exceeding the median productivity, but even at half the projected productivity gain, the social benefits are still dramatic.

Conclusions

This educational model will reduce the burden upon the crime laboratory to train a new hire. The technical leader at the crime laboratory will give a competency test to the new hire to evaluate his/her technical skills and qualifications for forensic DNA analysis. Upon the successful completion of the competency test, a proficiency test will be administered to qualify the new hire for forensic case work and/or DNA database analysis. It is anticipated that the crime laboratory managers can reduce the six-month training period for new hires possibly to one to two months. A reduction in the training period of a new DNA analyst can save tax payers approximately \$56,139 per new hire. Moreover, the crime laboratory can enter more DNA profiles into the DNA database with projected savings of \$27,600 per profile. The benefit to the public from a reduction in crime laboratory training and increased DNA profiles uploaded into the Combined DNA Index System database would have a projected saving of \$41.4 million.

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