one time approva	l to be taught as a	non-lab secree	Dummer 2013 only
LSC Use Only Proposal No:	UWUCC Use Only Proposal No: / A	-98	
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Curriculum Proposal Cover Sheet - University-Wide Undergraduate Curriculum Committee

Contact Person(s)		Email Address			
Bharathan Narayanaswamy/Seema Bha	rathan	bharathn@iup.edu/bharaths	@iup.edu		
Proposing Department/Unit		Phone			
Biology Check all appropriate lines and complete all information. Use a	separate cover sheet for each course proposal a	724-357-2584/357-2224			
		na or program proposal.	****		
Course Proposals (check all that apply)	0 0 0 0		L of		
	Course Prefix Change	Course Deletion			
Course Revision	Course Number and/or Title Change	Catalog Description (Change		
Current course prefix, number and full title:			'		
	28				
Proposed course prefix, number and full title, if ch		tion to Forensic Biolog	Y		
2. Liberal Studies Course Designations, as ap	propriate				
X This course is also proposed as a Libera	al Studies Course (please mark the app	ropriate categories below)	Egg 5		
•					
Learning Skills X Knowledge Area	Global and Multicultural Awar	eness Writing Intensive (inc	clude W cover sheet)		
Liberal Studies Elective (please mark the	designation(s) that applies - must meet	at least one)			
Global Citizenship	Information Literacy	Oral Communication	-6. 9		
Quantitative Reasoning Scientific Literacy Technological Literacy					
3. Other Designations, as appropriate					
1	her: (e.g. Women's Studies, Pan Africa	n)			
4. Program Proposals					
Catalog Description Change Pr	ogram Revision Program	Title Change	New Track		
New Degree Program Ne	w Minor Program Liberal Stud	ties Requirement Changes	_ Other		
Current program name:			N 101		
Proposed program name, if changing:		i li	-1 5 2000		
5. Approvals	Sign	ature	Date		
Department Curriculum Committee Chair(s)	Jan Howell		15 Feb 2013		
Department Chairperson(s)	alhus		Feb 15 701		
College Curriculum Committee Chair					
College Dean		/			
Director of Liberal Studies (as needed)	J. J. H. Pult		7 Merch 13		
Director of Honors College (as needed)					
Provost (as needed)					
Additional signature (with title) as appropriate					
JWUCC Co-Chairs					

Received

Part II. New Syllabus of Record

I. Catalog Description

BIOL 107 Introduction to Forensic Biology

3 class hours 3 credits (3c-0l-3cr)

Prerequisites: Non-Biology department majors and minors only

A broad overview of basic principles underlying modern applications of biology in forensic science. The course explores the science of forensic biology, traditionally known as *serology*, and the broad scope of laboratory tests used to investigate crimes involving DNA, blood, and other body fluids. The course will focus on the issues related to DNA fingerprinting as they apply to public or legal proceedings in the law enforcement arena. (Does not count toward Biology Electives, Controlled Electives, or Ancillary Sciences for Biology majors and minors).

II. Course Outcomes

Objective 1:

Describe and apply an understanding of the general methodology of modern biology in forensic science.

Expected Student Learning Outcomes 1 and 2:

Informed and Empowered Learners

Rationale:

Assignments will require students to have a level of knowledge DNA fingerprinting and serology that will enable them to understand how these techniques work. Assignments will also require students to evaluate ways to collect evidence, preserve samples of biological evidence, and critically analyze DNA data and to use this examination to explain how DNA fingerprinting tests are interpreted.

Objective 2:

Compare how general forensic evidence has been perceived over the years by identifying the issues related to forensic science and modern methods and strategies in forensic biology.

Expected Student Learning Outcome 2:

Empowered Learners

Rationale:

Assignments will require students to evaluate common forensic evidence collected by crime scene labs from the crime scene (photography, weapon, narcotics, ballistics tissues, and blood). In addition, these assignments will engage students in assessing a knowledge base in science and how that knowledge influenced the perception and management of DNA fingerprinting data with crime scene investigation.

Objective 3:

Describe how Forensic Biology and DNA fingerprinting technology are perceived as an investigative tool with in today's society.

Expected Student Learning Outcomes 3:

Responsible Learners

Rationale:

Assignments will require students to assess their own views and concerns about the impact of DNA technology on threats to liberty and privacy and how DNA typing technology has the potential for uncovering and revealing a great deal of information that most people consider to be intensely private. Other assignments will have the students analyze the impact of DNA fingerprinting technology in the public realm (e.g. legal system; prosecution of crimes; and in civil litigation) and to use this analysis to determine how their personal lives are and will be affected.

Objective 4:

Assess historical data last 25 years that have made contributions to our understanding of DNA fingerprinting in the court room.

Expected Student Learning Outcome 1 and 2:

Informed and Empowered Learners

Rationale:

Assignments will require students to gain an understanding of how quality DNA analysis and methods are fundamental to forensic DNA technology and its use in tracing the origins of criminal evidence. They will then apply these analyses to the evaluation of their own view for high-quality final results which are comparable to the results of other laboratories and to ensure the making of correct and impartial decisions in DNA testing.

III. Course Outline

A. Introduction- What is Forensics?

(3 hours)

- 1. Forensic Sciences: Type of Evidence
- 2. Forensic Anthropology, Forensic Dentistry and Toxicology
- 3. Definition and scope of Forensic DNA
- 4. Functions of the Forensic Scientist

B. Forensic Biology terminology and definitions

(7 hours)

- 1. Blood
- 2. Serum
- 3. Proteins
- 4. Saliva
- 5. Semen
- 6. X and Y chromosomes; Loci
- 7. DNA- source and types
- 8. Nucleotide Repeats

Exam 1 (1 hour)

C. DNA fingerprinting – Evidence Based Science

(6 hours)

- 1. DNA structure
- 2. DNA Evidence: Basics of Identifying, Gathering and Transporting
- 3. Types of Samples Suitable for DNA Testing: Questioned or Unknown Samples
- 4. Samples From Unidentified Bodies: Samples collected from unidentified bodies can include: blood, buccal swabs, hairs, bone, teeth, fingernails, tissues from internal organs (including brain), muscle, and skin.
- 5. Significance of Reference Samples From Known Individuals
- 6. Use of Samples from Relatives for Testing: Because a child inherits half of its DNA from each parent, it is possible to use reference samples collected from close relatives
- 7. Determination of Paternity or Maternity of a Child or Fetus Aborted fetal tissue significance for determining paternity, for example, in sexual assault and/or incest cases where conception occurred
- D. Safety as it relates to Evidence Collection, Sample Preservation, and Examples of contamination (4 hours)
 - 1. Crime Scene Integrity: Protection of the crime scene is essential to the protection of evidence
 - 2. Contamination: The risk of contamination of any crime scene can be reduced by limiting incidental activity
 - 3. Chain of Custody: If DNA evidence was contaminated, it may be necessary to identify persons who have handled that evidence
 - 4. Transportation and Storage: Any probative biological sample that has been stored dry or frozen, regardless of age, may be considered for DNA analysis
 - 5. DNA fingerprinting and criminal justice system

Exam 2 (1 hour)

E. DNA Evidence: Basics of Analyzing

(3 hours)

- 1. Overview of Steps in Analyzing DNA Evidence
- 2. Steps in DNA Sample Processing: Samples obtained from crime scenes or paternity investigations are subjected to defined processes involving biology, technology, and genetics

- 3. Types DNA Evidence Analysis Polymerase Chain Reaction (PCR); Short Tandem Repeats (STR); Y-Chromosome; and Mitochondrial DNA
- F. Evolution of DNA testing: Restriction Fragment Length Polymorphism (RFLP); and PCR (7 hours)
 - 1. DNA Typing Short Tandem Repeat (STR) Analysis of Short tandem repeat (STR) technology is a forensic analysis that evaluates specific regions (loci) that are found on nuclear DNA.
 - 2. Significance of 13 specific STR loci and The Federal Bureau of Investigation (FBI)
 - 3. DNA Typing Y-Chromosome Analysis: Several genetic markers have been identified on the Y chromosome that can be used in forensic applications. Y-chromosome markers target only the male fraction of a biological sample.
 - 4. DNA Typing Mitochondrial Analysis Mitochondrial DNA (mtDNA) analysis: significance of mtDNA testing to the investigation of an unsolved case.

Exam 3 (1 hour)

G. DNA Initiative: Advancing Criminal Justice through DNA Technology

(4 hours)

- 1. DNA Initiative Goals: Significance of DNA technology to ensure accuracy and fairness in the criminal justice system. DNA can be used to identify criminals with incredible accuracy when biological evidence exists, and DNA can be used to clear suspects and exonerate persons mistakenly accused or convicted of crimes.
- 2. The Initiative calls for increased funding, training, and assistance Federal, State, and local forensic labs; to police; to medical professionals; to victim service providers; and to prosecutors, defense lawyers, and judges.
- 3. DNA Legislative Milestones: Significance of "Justice for All Act of 2004," enforceable rights for victims of crimes; enhances DNA collection and analysis efforts; provides for post-conviction DNA testing; DNA Backlog Elimination Act (2000)-- To make grants to States for carrying out DNA analyses for use in the Combined DNA Index System of the Federal Bureau of Investigation; Crime Information Technology Act (1996),-- CITA allowed for grants for programs relating to the identification and analysis of DNA.
- 4. DNA Initiative Partners: Office on Violence against Women, U.S. Department of Justice; Federal Bureau of Investigation, U.S. Department of Justice
- H. Post-conviction Testing and Wrongful Convictions

(5 hours)

1. Overview of Wrongful Convictions: The strength of our criminal justice system depends on its accuracy — its ability to convict the guilty and to clear the innocent

Post-conviction DNA Testing Post conviction DNA testing is a major factor contributing to the increased discovery of wrongful convictions.
 Research on Actual Innocence and DNA Exoneration: The increase in exonerations over the last 20 years has accentuated the need for research on how, why and how often wrongful convictions occur.

Final exam (Exam 4) during final exam week

(2 hours)

IV. Evaluation Methods

- 1. 60% Four examinations (15% for each exam) three during the semester and a fourth during exam week. Exams will be short answer essays.
- 2. 20% Four case studies (5% for each case study) Students will be analyzing four case studies that use DNA to solve crimes These case studies will have questions that must be answered and turned in by the student. Each case study will be worth 5% of the final grade.
- 3. 15% Students will develop one case study for the class. This case study will be based on articles and ideas related to DNA evidence that is linked to DNA offender profiles through DNA databases. Student resources may include newspapers, newsmagazines, and popular science and medical magazines (e.g. Discover Magazine, Science and Medicine or Journal of Forensic Research). It will follow the format of the case studies given by the professor and will be worth 15% of their final grade. This will be a class assignment (no presentations in class).
- 4. 5% Critique of the non-textbook reading. Students will submit a critique with a maximum of five printed pages.

V. Grading Scale

Grading scale: A 90-100%; B 80-89%; C 70-79%; D 60-69%; F 59% and below

VI. Undergraduate Course Attendance Policy

The IUP attendance policy will be followed.

VII. Required Textbook

Gunn, A. 2009. Essential Forensic Biology, 2nd ed., Wiley-Blackwell, West Sussex U.K.

Supplemental Non-text book readings

Bowen, T.R. 2009. Ethics and the Practice of Forensic Science, CRC press

Gerber, S.M. 2011. Chemistry and Crime: From Sherlock Holmes to Today's Courtroom (American Chemical Society Publication) ISBN: 0841207852. Publisher: Oxford Univ. Press

Koff, C. 2005. Bone Woman: A Forensic Anthropologist's Search for Truth in the Mass Graves of Rwanda, Bosnia, Croatia, and Kosovo - 04 edition ISBN13: 978-0812968859 Publisher: Random House, Inc.

<u>Ramsland</u>, K. 2007. The Human Predator: A Historical Chronicle of Serial Murder and Forensic Investigation. Published by Berkley Trade

Suggested Readings

Alessandrini, F., M. Cecati, M. Pesaresi, C. Turchi, F. Carle, and A. Tagliabracci, 2003. "Fingerprints as evidence for a genetic profile morphological study on fingerprints and analysis of exogenous and individual factors affecting DNA typing," *J. Forensic Science* 48(3): 1–7

Bertino A.B., and Bertino, P.N. 2012. Forensic Science: Fundamentals and Investigations ISBN 13: 978-0-538-73155-3 South-Western Publishers

Butler, J.M., 2005. Forensic DNA Typing, Second Edition: Biology, Technology, and Genetics of STR Markers ISBN-13: 978-0121479527

Houck, M.A., Siegel, J.A., 2010 Fundamentals of Forensic Science 978012374989-5 Academic Press

Levy, R.J., 2011. The Michael Jackson Autopsy: Insights Provided by a Forensic Anesthesiologist. J Forensic Res 2:138. doi:10.4172/2157-7145.1000138

President's DNA Initiative, Lessons Learned From 9/11, 2006. DNA Identification in Mass Fatality Incidents, NCJ 214781 (Washington, D.C.: U.S. Department of Justice, National Institute of Justice, Available at http://massfatality.dna.gov/.

Roman, J., K. Walsh, P. Lachman, and J.Yahner, "Post-Conviction DNA Testing and Wrongful Conviction" 2012, Final report to the National Institute of Justice, contract number 2008F-08165, NCJ 238816.

Verma, K., Joshi, B., 2012. Different Animal Species Hairs as Biological Tool for the Forensic Assessment of Individual Identification Characteristics from Animals of Zoological Park, Pragti Maidan, New Delhi, India. J Forensic Res 3:160. doi:10.4172/2157-7145.1000160

VIII. Bibliography

Barnett, P.D. 2001. Ethics in Forensic Science: Professional Standards for the Practice of Criminalistics, CRC Press, Boca Raton

Carracedo, A. 2005. Forensic DNA Typing Protocols (Methods in Molecular Biology, V. 297.) Humana Press

Casarett, T., & Doull's K. 2003. Essentials of Toxicology edited by Curtis D., and Watkins, J.B. McGraw-Hill

Colin, E. 2002. Question of Evidence: The Casebook of Great Forensic Controversies, from Napoleon to O. J. John Wiley & Sons Inc.

Coyle, H.M. 2004. Forensic Botany: Principles and Applications to Criminal Casework CRC Press, Boca Raton

DiMaio, V.J.M. 2001. Forensic Pathology, Second Edition, CRC Press, Boca Raton

Inman, K. 2000. *Principles and Practice of Criminalistics: The Profession of Forensic Science*, CRC Press, Boca Raton

James, S.H. and Nordby, J.J. 2003. Forensic Science: An Introduction to Scientific and Investigative Techniques, CRC Press, Boca Raton

Kubic, T. 2005. Forensic Science Laboratory Manual and Workbook, Revised Edition, CRC Press, Boca Raton

LeBeau, M.A. 2004. Quality assurance guidelines for laboratories performing forensic analysis of chemical terrorism: Scientific Working Group on Forensic Analysis of Chemical Terrorism, Thomson Gale.

Lincoln, P.J., Thomson, J.A.1998. DNA Profiling Protocols: Methods in Molecular Biology ed., by Humana Press, New Jersey, USA

Mozayani, A, Noziglia, C. 2005. *The Forensic Laboratory Handbook: Procedures and Practice (Forensic Science and Medicine*), Humana Press.

Ngaire E.G. 2008. The Forensic Casebook: The Science of Crime Scene Investigation Prentice hall

Ogle, R.R., and Fox, M.J. 1998. Atlas of Human Hair: Microscopic Characteristics CRC Press

Richard, Li. 2008. Forensic Biology: Identification and DNA Analysis of Biological Evidence ISBN-13: 978-1420043433. CRC Press, Boca Raton

Roberts, G.W. 2012 Forensic Crime Scenes Health and Safety CRC Press

Rudin, N. 2001. An Introduction to Forensic DNA Analysis, Second Edition, CRC Press

Saferstein, R.E. 1982, 1988, & 1994. Forensic Science Handbook Vols. I, II & III, Prentice-Hall, Englewood, NJ

Stuart, J. 2005. Forensic Science: An Introduction to Scientific and Investigative Techniques, 2nd ed., Humana Press, New Jersey, USA

Stuart, J.2005. Principles of Bloodstain Pattern Analysis: Theory and Practice2nd ed., Humana Press, New Jersey, USA

Answers to Liberal Studies Questions

- 1) Not applicable, only one instructor will teach this course.
- 2) Major aspect including women will be a section about specific investigative and forensic processes related to sex crimes from the work of John Savino, Brent Turvey, and J. Baeza and the recent advances that has led to a new, more efficient approach to processing DNA from rape evidence. The majority of contributions by women in the field of Forensic science have come in the past decade. In the last portion of the class we will discuss the contributions of two of the major contributors in the field of forensic research Katherine Ramsfield and Clea Koff. Three of the four non-textbook readings available are related to crimes case studies that will incorporate women and minorities as part of the study.
- 3) Students will be required to read one of the following books a supplementary book in addition to the required text for the course. In addition to the textbook ", a number of non-textbook readings like *The Human Predator: A Historical Chronicle of Serial Murder and Forensic Investigation by K.* Ramsland ; Chemistry and Crime: From Sherlock Holmes to Today's Courtroom by Samuel M. Gerber will be incorporated into the course.
- 4) This is an introductory course. It differs from our non-majors beginning courses (General Biology I) by focusing on one theme – forensic biology rather than the entire realm of biology. In addition, the General Biology I course does not cover any DNA Initiative involving Criminal Justice and DNA Technology.

Answers to Course Analysis Questionnaire

Section A: Details of the Course

- A1 How does this course fit into the programs of the department? For what students is the course designed? (majors, students in other majors, liberal studies). Explain why this content cannot be incorporated into an existing course.

 This course is intended to satisfy the Liberal Studies Natural Science non-Lab Science requirement. This course will give the student as up-to-date introduction of a particular field of forensic biology that is intended to be relevant to their everyday lives. The content of this course reflects growing recognition of the importance of biological evidence in forensic science. This course will include several topics previously not taught in any of the courses offered by the biology department.
- A2 Does this course require changes in the content of existing courses or requirements for a program? If catalog descriptions of other courses or department programs must be changed as a result of the adoption of this course, please submit as separate proposals all other changes in courses and/or program requirements. This will not change the content of any other existing course in the program.
- A3 Has this course ever been offered at IUP on a trial basis (e.g. as a special topic) If so, explain the details of the offering (semester/year and number of students). This is a newly designed course that is being created to fulfill the new requirements of the Liberal Studies component at IUP.
- A4 Is this course to be a dual-level course? If so, please note that the graduate approval occurs after the undergraduate.

 No.
- A5 If this course may be taken for variable credit, what criteria will be used to relate the credits to the learning experience of each student? Who will make this determination and by what procedures?

 N/A
- A6 Do other higher education institutions currently offer this course? If so, please list examples (institution, course title).

University of Central Florida: Forensic Biochemistry

University of Portsmouth: Forensic Biology and Biochemistry

Suffolk University: Forensic Biochemistry

University of Kent: Forensic Biology

Southern Illinois University: Forensic Biochemistry
New Mexico State University: Forensic Biochemistry

A7 Is the content, or are the skills, of the proposed course recommended or required by a professional society, accrediting authority, law or other external agency? If so, please provide documentation.

No.

Section B: Interdisciplinary Implications

- B1 Will this course be taught by instructors from more than one department or team taught within the department? If so, explain the teaching plan, its rationale, and how the team will adhere to the syllabus of record.

 Not applicable
- B2 What is the relationship between the content of this course and the content of courses offered by other departments? Summarize your discussions (with other departments) concerning the proposed changes and indicate how any conflicts have been resolved. Please attach relevant memoranda from these departments that clarify their attitudes toward the proposed change(s). There are no conflicts with other departments at IUP.
- B3 Will this course be cross-listed with other departments? If so, please summarize the department representatives' discussions concerning the course and indicate how consistency will be maintained across departments.

 No.

Section C: Implementation

- C1 Are faculty resources adequate? If you are not requesting or have not been authorized to hire additional faculty, demonstrate how this course will fit into the schedule(s) of current faculty. What will be taught less frequently or in fewer sections to make this possible? Please specify how preparation and equated workload will be assigned for this course.

 The current faculty resources are sufficient.
- C2 What other resources will be needed to teach this course and how adequate are the current resources? If not adequate, what plans exist for achieving adequacy? Reply in terms of the following:
 - *Space- the current rooms used for lectures that can accommodate 132 students are adequate.
 - *Equipment- Not applicable
 - *Laboratory Supplies and other Consumable Goods- Not applicable
 - *Library Materials- there will be no necessity of using library materials in this course other than existing electronic databases.
 - *Travel Funds- there will be no travel funds required for this course.
- C3 Are any of the resources for this course funded by a grant? If so, what provisions have been made to continue support for this course once the grant has expired? (Attach letters of support from Dean, Provost, etc.)

 None.
- C4 How frequently do you expect this course to be offered? Is this course particularly designed for or restricted to certain seasonal semesters?

 This course will be offered every other semester. Because of the nature of the topics covered in this course, there are no seasonal requirements.
- C5 How many sections of this course do you anticipate offering in any single semester? While it is difficult to determine ahead of time, it is expected to have one lecture section.

- C6 How many students do you plan to accommodate in a section of this course? What is the justification for this planned number of students?

 In the lecture, we plan to accommodate up to 132 students. This course is a non-lab course and therefore is not limited by laboratory classrooms- capacity.
- C7 Does any professional society recommend enrollment limits or parameters for a course of this nature? If they do, please quote from the appropriate documents. No.
- C8 If this course is a distance education course, see the Implementation of Distance Education Agreement and the Undergraduate Distance Education Review Form in Appendix D and respond to the questions listed.

 It is not a distance education course.

Part III. Letters of Support or Acknowledgment
Department of Criminology
Department of Chemistry

Sample assignment

Assignment 4 (5% of grade)

Critique of the non-textbook reading. Students will submit a critique with a maximum of five printed pages.

- 1. Read the following article.
- 2. Guided Questions for non-text book reading
 - i) What is Forensic entomology? What information can a forensic entomologist provide at the death scene?
 - ii) What are the major challenges of identification of the maggots to the species?
 - iii) What were the major methods of Human DNA analysis that the authors employed to non-human organisms?
 - iv) Based on this article how would you relate the application of Entomological evidence to help determine the circumstances of abuse and rape?

Evaluation: Grading Rubric (non-text book reading)

Organization and Format

-5 Case analysis is neat and organized, word processed, and contains all required components as outlined on the handout including artifacts to support each instructional strategy.
- 4 Case analysis is organized and word processed, and all items are included but are not well developed.
- 3 Case analysis has some organized sections but not all are included or well developed.
-1-2 Case analysis lacks organization and/or required components, not word processed, or is missing artifacts to support instructional strategies.

Word Choice in Writing

- 5 Writing reflects carefully chosen words and uses a precise, professional vocabulary.
- 4 Writing includes a minimal amount of interesting or professional language.
- 3 Writing includes only ordinary or common terms.
- 1-2 Writing lacks precise and appropriate language; does not communicate the purpose of the case analyses.

Case Analyses Content Summary (Reflections and Recommendations)

......5 Summaries are concise and reflective with specific future instructional recommendations based upon assessment data.

-4 Summaries are concise and reflective but lack specific future instructional recommendations.
-3 Summaries are brief and provide limited conclusions; do not demonstrate complete understanding.
-1-2 Summaries are incomplete; do not demonstrate understanding; do not include future recommendations for instruction.

Other exercises including interpretation of practical data from case studies and problem solving will provide informal formative feedback to students and support the preparation of the summative assessed coursework assignment.

Identification of a Death-scene Maggot using Standardized Molecular Methods: *Sarcophagabullata* Parker 1916 (Sarcophagidae) Out-numbers Blowflies (Calliphoridae) on an Urban Cadaver in Southeastern Texas

Rekha Raghavendra¹, Christopher P. Randle² and Sibyl Rae Bucheli²

¹School of Medicine Case Western Reserve University, Cleveland OH, USA ²Department of Biological Sciences Sam Houston State University, Huntsville, TX, USA

Abstract

In forensic entomology,fly data including maggot age are frequently used to help estimate the time since death Accurate identification of the maggot to species is critical for time since death estimations. However, within a family, maggots are notoriously difficult to identify to species. In this study, we employ phylogenetic datafrom the mtDNAgenes COI and COII to identify an unknown maggot to species (member of the family Sarcophagidae) harvested from a cadaver in June 2009 in Harrison County, Texas. The most closely related species to our unknown maggot was SarcophagabullataParker 1916, a somewhat common carrion-feeding species in southeastern United States that is now gaininggreater recognition as a forensically significant species.

Keywords: Forensic entomology; case study; Sarcophagidae; Sarcophagabullata Parker 1916

Introduction

Decomposition of a large mammalian carcass is greatly accelerated through the action of insects belonging to the order Diptera (flies) [1]. In southeastern Texas, initial colonizers include members ofthe families Calliphoridae (blow flies), Sarcophagidae (flesh flies), and Muscidae (house flies), with blow flies and flesh flies often arriving and laying eggs or giving birth tomaggots (rather than laying eggs) within minutes of death (unpublished records from cadavers at the Southeastern Texas Applied Forensic Science Facility at the Center for Biological Field Studies at Sam Houston State University). Maggots acquirebiomass as a function of physiological time rather than calendar time and therefore develop at a predictable rate. Since flies arrive and lay eggs or maggots immediately, they are considered useful tools for estimation of the time that has elapsed since death, or the postmortem interval (PMI), by estimating the time since maggot colonization [1-7]. By recreating the conditions of the death scene in the laboratory and working backwards through time to determine the age of the oldest maggot, the forensic entomologist can correlate the age of the maggot to the PMI [1-7].

Identification of maggots to species remains a challenging aspect to forensic science even though maggots are frequently collected evidence during a death scene investigation. Identification keys are not currently available for all life stages are not currently available andmaggots are difficult to identify particularly at early life stages because morphological features among maggots are similar, rendering them virtually undistinguishable beyond the family level [8-12]. Molecular data can aid in the identification of larvae where morphology is limited in utility [8-12]. In this study, we employ an established phylogenetic protocol by Wells et al. [10] using the mitochondrial DNA genes of COI and COIIto identify an unknown maggot of the family Sarcophagidae harvested from a cadaver in Harrison County, Texas.

Materials and Methods

Specimens: The unknown maggot was recovered from a body discovered in June 2009 in Harris County, TX, and was the largest observed maggot and most abundant larval type; in fact, no other

species were collected despite law enforcement agents reporting the remains to be in a state of fresh/bloated decomposition. The unknown maggots were identified as members of the family Sarcophagidae using standard morphological features of the spiracular complex but could not be further identified (Peterson 1960). Common species of Sarcophagidae which frequent cadavers in this area include Sarcophaga (Neobellieria)bullata Parker, 1916 and Sarcophaga (Bercaea) africa (Wiedemann 1824: 49) (=cruentata Meigen 1826;=haemorrhoidalis auct.) [14-16]. Proper species identification is critical to generate proper growth curves for age estimation; Wells et al. [10] demonstrate that these two species grow at rates disparate enough to create as much as a 24 hour discrepancy.

DNA extraction: Genomic DNA was extracted from the unknown maggotstarting with tissue homogenization using a Disruptor Genie TM and followed by a standardChelex DNA extraction method [17].

Amplificationand Sequencing: PCR protocols were modified from Wells et al. [10] using their published primers for COI and COII in various combinations (Table 1) and carried out in 50 µl volumes including 1X PCR buffer (Promega, Madison WI), 0.4 µM forward and reverse primers, 0.2mMdNTPs, 2.5UGoTaq polymerase (Promega, Madison WI), with 3 µl of template DNA. PCR reaction conditions were as follows: 94°C for 2 min(initial denaturation), continued with 35 cycles of 94°C for 1 min (denaturation), 50°C for 1 min (primer annealing), 72°C for 2 min (extension), and 72°C for 10 min (final extension). PCR products were visualized on 1% agaroseand purified

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using a QIAquick PCR Puriheation Kit (QIAGEN INC., Valencia, CA). The COI and COII regions were sequenced on a Bechman-Coulter 8000 CEQ Genetic Analyzer using the GenomeLab DTCS Quick Start Kit method.Contig assembly was performed using Geneious [18].

Selection of Sequences for comparison: NCBI nucleotide MEGABLAST was used to identify 100 closest Genbanksequence matches to the unidentified maggot sequence. These were downloaded, and aligned with the unknown sequence using the MUSCLE [19] algorithm in Geneious [18]. Of these, 81 sequences were chosen that had sufficient overlap with each other and the unknown sequence to allow unambiguous alignment. Multiple accessions for species were retained when available, including two sequences each from Sarcophaga africa and S. bullata. Additionally, a sequence obtained from Eucalliphora latifrons (Calliphoridae) was selected as an outgroup. The reduced matrix was realigned using the MUSCLE [19] algorithmfor phylogenetic analysis.

Phylogenetic Analysis: Maximum parsimony searches were conducted using WinClada [20] as a shell program. The parsimony ratchet [21] was implemented with 200 iterations (10% of the matrix sampled; one tree held per iteration). The tree generated via the ratchet search was the starting tree for a more thorough analysis conducted in NONA ver. 2.0 [22] using the commands "rs 0; hold 1000; mult' 50." Parsimony jackknife percentages (23) were calculated in NONA ver. 2.0 [22] with 1000 replications (200 search steps; one starting tree per replication; rs 0). For maximum likelihood (ML) analysis, the most appropriate stationary model of evolution was inferred using the Akaike Information Criterion [24] in jMODELTEST [25,26]. ML searches were performed using GARLI 1.0 (27) using the default configuration. One thousand non-parametric bootstrap replicates were analyzed with two search replicates each to obtain clade support. Phyutility [28] was used to generate the majority rule consensus of 1,000 bootstrap trees.

Results

NCBI nucleotide MEGABLAST returned a COI sequence from Sarcophaga bullataas the best sequence match with 97% sequence identity, while S. africa obtained 92% identity and the outgroup,

Eucalliphora latifrons obtained an identity of 86%. Sequence alignment resulted in a matrix of 2,305 characters. Parsimony searches resulted in a single most parsimonious trees (L=3921;CI=0.30; RI=0.65). AIC identified GTR+I as the best fitting model of evolution. The maximum likelihood tree obtained anln likelihood score = -21,146.499. MP and ML trees were largely congruent, differing only in the resolution of clades that were poorly supported and inconsequential in the identification of the known sequence. While overall clade support was low, the two most probable species matches, Sarcophaga africa and S. bullata were separated by several strongly supported nodes (jackknife and bootstrap >80%; Figure 1 (included as supplementary data)). Both parsimony and likelihood identified the unknown sequence as sister to the two sequences of Sarcophaga bullata (jackknife =100%; bootstrap = 67%). Sarcophaga polistensis (jackknife =85%; bootstrap = 89%) is sister to this clade, and S. cooleyi is sister to the clade including S. polistensis-S. bullata (jackknife =95%; bootstrap = 97%). Sarcophaga polistensis occurs in Texas, but is not known to feed on carrion [14]. Sarcophaga cooleyi is not known to occur in Texas. Therefore, evidence best supports the hypothesis that the unknown maggot is S. bullata.

Discussion

Many modern forensic techniques that employ DNA profiling to make associations between individuals and individuals, individuals and locations, and/or individuals and events (such RFLP analysis, PCR analysis, STR analysis, AmpFLP, DNA family relationship analysis, Y-chromosome analysis, mitochondrial analysis) [29] are sound due to the process of evolution acting on marker loci. Marker similarityis interpreted as evidence for shared ancestry [30]. Overall, the process leads to situations where more closely related organisms share in common more regions of their DNA. In most situations, DNA profiling analyses are based in principles of phylogenetics (the study of evolutionary relatedness among groups of organisms) and population genetics (the study of the effects of evolutionary processes on allele frequenciesin populations) [30,31]. In a growing number of situations, it has been useful to extend methods commonly employed in human DNA analyses to non-human organisms (for a discussion see 29). For species identification of unknown organisms, modern methods of

Location on the mtDNA				
Primer Sequence	Paired combination of primers used in this study			
1	TY-J-1460	TACAATTTATCGCCTAAACTTCAGCC	2, 4	
2	C1-N-1687	CAATTTCCAAATCCTCCAATTAT	1	
3	C1-J-1751	GGATCACCTGATATAGCATTCCC	6. 8	
4	C1-N-1840	AGGAGGATAAACAGTTCAC/TCC	1	
5	C1-J-2183	CAACATTTATTTTGATTTTTTGG	11	
6	C1-N-2191	CCCGGTAAAATTAAAATATAAACTTC	3	
7	C1-J-2319	TAGCTATTGGAC/ITATTAGG	10, 13	
8	C1-N-2293	AGTAAACCAATTGCTAGTATAGC	3	
9	C1-J-2495	CAGCTACTTTATGAGCTTTAGG	13, 14	
10	C1-N-2514	AACTCCAGTTAATCCTCCTAC	7	
11	C1-N-2659	GCTAATCCAGTGAATAATGG	5	
12	C1-J-2792	ATACCTCGACGTTATTCAGA	16	
13	C1-N-2800	CATTTCAAGT/CTGTGTAAGCATC	7. 9	
14	TL2-N-3014	TCCAATGCACTAATCTGCCATATTA	9	
15	C2-J-3138	AGAGCCTCTCCTTTAATAGAACA	18	
18	C2-N-3389	TCATAAGTTCA[R]TATCATTG	12	
17	C2-J-3408	CAATGATAT/CTGAAGT/ATATGA	18	
18	TK-N-3775	GAGACCATTACTTGCTTTCAGTCATCT	15, 17	

Table 1: PCR primers' used in this study." Primers were taken from Wells et al. [10]. N-forward primer, J-reverse primer.

phylogenic analyses are the preferred method. In a now famous paper, Scadutoet al. [32] demonstrate the source of transmission of HIV strains by standard and rigorous phylogentic analysis (using maximum likelihood and Bayesian estimators). Such methods are frequently employed in insect identification (for a forensic focus on Calliphoridae and Sarcophagidae only see: [8-12,33-36]).

The intent of this study was identification to species of the largest larval flies harvested from a cadaver by using established phylogenetic protocols. These protocols have only been worked out for controlled situations and have not been used "in the field." Larvae were identified initially by standard morphological methods as members of the family Sarcophagidae. Both parsimony and likelihood trees generated from COI and COII mtDNA data matrices of GenBank data-based sequences and the largest unknown specimen strongly allied the unknown sequence as sister to Sarcophaga bullata. (Figure 1 (included as supplementary data)) shows all species included in the analysis and their GenBank accession numbers. In the analysis, Sarcophagidae forms a monophyletic group. Analysis of reference sequences downloaded from GenBank database shows little variation between species with different accession numbers. This suggests that the protocol developed by Wells et al. [10] for the use of reference sequences available in the GenBank database is a sensible tool to reveal identity ofan unknown specimen. Phylogenetic analysis using these reference sequences was able to determine the species of the flesh fly collected from a cadaver and hence and may be used to provide supporting information to aid in the estimation of the time sinceinsect colonization.

The occurrence of Sarcophaga bullata as the largest and most abundant species of larval fly recovered from the corpse is note-worthy. Despite the remains being reported by law enforcement as fresh/bloated, this species outnumbered members of the family Calliphoridae (no larvaeof Calliphoridae were recovered). While many published accounts of necrophagous species biodiversity of a corpse note the presence of S. bullata, no published accounts rely primarily on data provided by this species as the largest and most abundant member of the community for applied aspects of the science. Anecdotal accounts of the utility of this species in forensic applications exist; an entry made on the open-access on-line Encyclopedia Wikipedia discusses their forensic importance. Their abundance in this situation may be explained by the location of the corpse and time of death in terms of season. In June, southeastern Texas (Houston and surrounding cities) experiences average daytime high temperatures above 90°F/32°C, nighttime lows around 70°F/21°C, and relative humidity levels that fluctuate widely between 50% at noon and 90% at midnight (average minimum and maximum when not raining) [37]. This generally results in dehydration of tissues of the corpse at an accelerated rate (personal observations made of human decomposition at STAPS at CBPS at SHSU, Bucheli and Lindgren) when compared to published descriptions of cadavers at other forensic anthropology stations through out the United States (2; 3; 5; 6; 38; 39). Furthermore, extensive areas of Montgomery County are urbanized. Unpublished photos of crime scenes from variousHouston, TX, urban and rural locations reveal corpses with fewto no observed species of Calliphoridae and much greater numbers of Sarcophagidae (personal observations, Bucheli). Reasons for the absence of Calliphoridae may include the lack of a constant supply of large, fresh mammalian corpses due to urbanization in certain areas to sustain populations of significant size. The authors recognize this discussion as largely speculative but do so to draw attention to the fact that very little is known regarding the utility of Sarcophaga bullata in forensic situations.

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Additional formative assessment – detail of process and rationale In class exercises including interpretation of practical data from case studies and problem solving will provide informal formative feedback to students and support the preparation of the summative assessed coursework assignment. Revision tests related to the topics assessed in the practical exam will be available via Blackboard (eLP) and will provide immediate formative feedback, with suggestions for additional reading. c. Indication of how students will get feedback and how this will support their learning Individual written feedback will be provided for the summatively assessed coursework and practical exam. Formative feedback will be delivered informally in workshops and during class exercises, and will also be built into the online revision tests, as detailed at point b. above.

CASE STUDY ASSIGNMENT INSTRUCTIONS

- All case study assignments must be <u>TYPED</u> with your name, the course number, and my name in the right hand corner.
- You do not need to re-type the question on your assignment, but should number your answers to correspond to the correct question.

ASSESS Assignments

- You will need to read <u>the Case study provided</u> and provide answers to the ASSESS questions at the end of each case.
- You will have to apply the course material for those topics prior to completing these assignments.
- Your answers will be based on your prior knowledge so personal opinions are acceptable but should be written in a professional manner.
- Assignments must be typed with your name, the course number, and my name in the right hand corner.
- You do not need to re-write the question, but should number your answers.
- Each ASSESS assignment will be worth 4 points:
 - o 2 points for the Elementary School case
 - o 2 points for the High School case
- The grading criteria will be based on:
 - the thoughtfulness or completeness of the answer
 - o professional writing (grammar, spelling, etc.).

REFLECT & EVALUATE Assignment (See a sample assignment below for Module 7)

- You will choose <u>one</u> of the two case studies you read for the ASSESS assignment
 - o Elementary School or
 - High School.
- You will complete the assignment after reading the course material on those topics.
- Answers should be based on integrating information from the course readings (modules) including the use of key concepts and theories.
- Personal opinions are not appropriate in these assignments.
- The length of each answer will vary with some being only a couple of sentences and others requiring more details.
- Each REFLECT & EVALUATE assignment will be worth 20 points (approximately 4 points per question).
- The grading criteria for each question will be based on:
 - 2 points for integration with the module (use key concepts; no personal opinions),
 - o 1 point for the thoughtfulness and completeness of each answer
- o 1 point for professional writing style (grammar, spelling, etc.)