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PAPER

GENERAL

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The 2018 California Wildfires: Integration of Rapid DNA to Dramatically Accelerate Victim Identification*

ABSTRACT: In November 2018, Butte County, California, was decimated by the Camp Fire, the deadliest wildfire in state history. Over 150,000 acres were destroyed, and at its peak, the fire consumed eighty acres per minute. The speed and intensity of the oncoming flames killed scores of people, and weeks before the fire was contained, first responders began searching through the rubble of 18,804 residences and commercial buildings. As with most mass disasters, conventional identification modalities (e.g., fingerprints, odontology, hardware) were utilized to identify victims. The intensity and duration of the fire severely degraded most of the remains, and these approaches were useful in only 22 of 84 cases. In the past, the remaining cases would have been subjected to conventional DNA analysis, which may have required months to years. Instead, Rapid DNA technology was utilized (in a rented recreational vehicle outside the Sacramento morgue) in the victim identification effort. Sixty-nine sets of remains were subjected to Rapid DNA Identification and, of these, 62 (89.9%) generated short tandem repeat profiles that were subjected to familial searching; essentially all these profiles were produced within hours of sample receipt. Samples successfully utilized for DNA identification included blood, bone, liver, muscle, soft tissue of unknown origin, and brain. In tandem with processing of 255 family reference samples, 58 victims were identified. This work represents the first use of Rapid DNA Identification in a mass casualty event, and the results support the use of Rapid DNA as an integrated tool with conventional disaster victim identification modalities.

KEYWORDS: Rapid DNA Identification, FlexPlex assay, ANDE, A-Chip, I-Chip, DNA ID, short tandem repeat (STR), disaster victim identification

Following a mass casualty event, identifying victim remains is critical to providing support to family and friends of those lost. Postmortem identification can be challenging due to the nature, location, and accessibility of the disaster site, cause of death, time to recovery of a given set of remains, time to transport to a morgue or laboratory, and time required to initiate and perform the various analyses. Furthermore, some biometrics are of limited utility even soon after time of death and most are ineffective as the body decomposes. Identifying features may be destroyed or altered beyond recognition by explosions, fire, caustic chemicals, projectiles, decay, or combinations of these factors. In contrast, DNA is well suited to survive physical and chemical insults and is of enormous value in identifying the deceased. Although genomic DNA from soft tissue degrades soon after death due to autolysis, microbial contamination, and environmental insult, DNA present within bone (particularly dense cortical bone) and teeth may remain relatively intact for years, decades, or millennia (1,2). Over the past 20 years, DNA typing in forensic laboratories has become a standard tool for disaster victim identification (DVI) following mass casualty events including the World Trade Center and other terrorist attacks (3–5), natural phenomena (6–9), and plane crashes (10,11).

Conventional laboratory DNA processing of DVI samples requires sophisticated equipment, highly skilled technical operators, complex data interpretation, and kinship analysis—all of which require considerable time and effort. Local laboratories may be rendered nonfunctional by the disaster, and distant laboratories may be overwhelmed by the volume of samples to be analyzed. Furthermore, neither makeshift morgues nor the vast majority of permanent morgues have the equipment or trained staff to perform DNA analysis. Accordingly, even a relatively small disaster such as a commercial plane crash can take years for body parts to be identified by conventional processing. When large mass disasters occur, bodies may be unidentified for years or decades—frequently, thousands of bodies are disposed of in mass graves (12,13). The 2004 Indian Ocean earthquake and tsunami is a tragic example—more than a decade later,

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approximately 10% of victims remained unidentified (14). Rapid DNA Identification (15–22), the fully automated process of generating a DNA ID (also referred to as a short tandem repeat [STR] profile or DNA profile) from a forensic sample, typically performed outside the laboratory by nontechnical operators with results available in less than two hours, offers the possibility of accelerating DNA processing of DVI samples.

In November 2018, Butte County, California, was decimated by the Camp Fire, the deadliest and most destructive wildfire in state history. Over 150,000 acres were destroyed, and at its peak, the fire consumed eighty acres per minute (Fig. 1). On the day the Camp Fire started (23,24), emergency systems in the State of California were already severely strained by a mass fatality shooting in Ventura (25) and the Woolsey Fire in southern California (26). The Camp Fire response was coordinated by the Butte County Sheriff's Office and involved collaboration with California's Office of Emergency Services, CAL Fire, the National Guard, California Highway Patrol, and sheriff-coroners' investigators and search-and-rescue teams from throughout the state. In addition, Butte County Sheriff's Office requested the assistance of forensic anthropology teams from California and Nevada.

The Sacramento County Coroner's Office was tasked with determining cause of death and identifying Camp Fire victims. Unidentified Human Remains (UHR) were transported daily from the fire impacted area to the Sacramento County Morgue. UHR intake typically consisted of cataloging personal items found with remains, photographing remains and the items found, fingerprinting if possible, documentation of information on the decedent, full x-rays, and examination by one or more forensic pathologists, anthropologists, and odontologists. Within a day of the initial UHR being received at the morgue, it became clear that conventional identification modalities would not be effective for the severely degraded remains, that DNA analysis would be critical to make identifications, and that Rapid DNA technology should be integrated into the morgue's identification workflow.

Materials and Methods

Recovery of Human Remains

The forensic teams were composed of anthropology faculty, students, and alumni from California State University, Chico (CSU, Chico) as well as faculty, students, and experts from UC Santa Cruz, the University of Nevada, Reno, the University of Nevada, Las Vegas, and the Clark County Medical Examiner's Office in Las Vegas. Between November 10–28, 2018, forensic teams aided in the search and recovery of human remains within Concow, Paradise, and Magalia, California. Three additional search days were conducted in December and January of 2019 to search for additional remains of fire victims.

Forensic anthropology teams were provided with missing persons lists and addresses each day to guide the search and recovery operations. In addition, smaller anthropology teams received *ad hoc* "call-outs" by search-and-rescue teams to addresses where suspected remains were found. Each team evaluated whether the materials were osseous or nonosseous and whether osseous remains were human or nonhuman in origin. Search patterns began with a perimeter walk around the structure, followed by a minimally invasive foot survey through the structure, and finally a thorough search with small hand tools. Forensic anthropology teams carefully excavated each scene with hand trowels, sieves, and other small hand tools to ensure a thorough recovery of remains, especially fragmentary teeth and dental appliances (27). Each excavation team fully screened each area with visible bone and at least a meter around the periphery to ensure complete recovery of remains. These methods allowed for the recovery of dental remains and appliances, pacemakers, and orthopedic devices. Remains were transferred to paper evidence bags, which were then placed into coroner's body bags for transport.

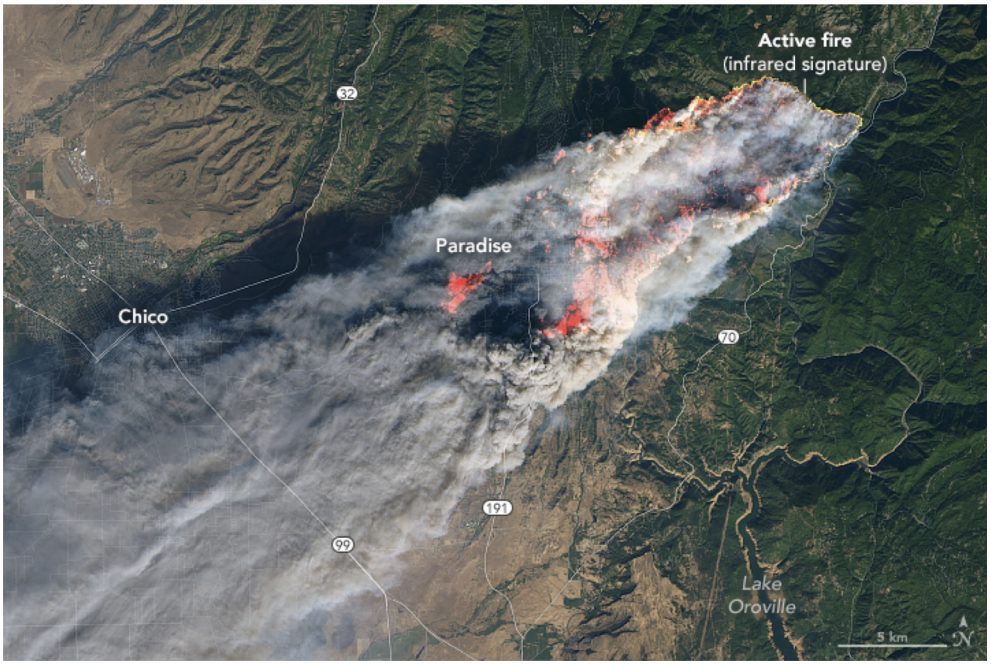

FIG. 1—*Landsat 8 imagery composite of Butte County approximately 4.5 h after the start of the Camp Fire. The image was created using Landsat bands 4-3-2 (visible light), along with shortwave-infrared light to highlight the active fire. [Color figure can be viewed at wileyonlinelibrary.com]*

Anthropologic Methods

Age, sex, and stature were determined, when possible, using standard methods (28,29). However, biological profile information was only obtained in a few cases due to the extent of thermal damage to the remains.

Sample Preparation and Processing for Rapid DNA Identification

Samples processed fell into three categories: those obtained from UHR, antemortem specimens from potential victims, and family members of potential victims. UHR sample selection was an iterative process that gradually improved as the range and types of degradation became apparent. All remains were processed using protocols that minimized sample consumption.

Dried Blood on FTA Cards. Most bloodcards were lightly stained, and 6 × 6 mm cuttings were processed. Each cutting was placed in a 2-mL microcentrifuge tube to which 120 μL of TE-4 was added. The cutting was thoroughly macerated with a pipette tip and then incubated at 50°C for 15 min. An ANDE® collection swab was inserted into the tube, and all fluid was absorbed onto a swab for Rapid DNA Identification.

Dried Blood/Blood Clots. For blood samples that were too dry to be preserved on an FTA card, approximately 10–20 μg fragments of dried blood were macerated with 100 μL sterile water to form a thick slurry. The slurry was then spread over the head of an ANDE swab for Rapid DNA Identification.

Organs. Organs that retained moisture content (e.g., the hepatic interior) were swabbed with a dry ANDE swab until the moisture saturated the cotton head. For desiccated tissues, small slices were taken, moistened with sterile water, macerated, and the resulting slurry was spread over the cotton swab head for Rapid DNA Identification.

Muscle. Muscle typically presented as thin striated bands with durable sinewy coatings. Depending on the desiccation level (pliable, somewhat pliable, or leathery), sample maceration with sterile water was added to form a slurry and applied to an ANDE swab for Rapid DNA Identification.

Bone. Bone fragments ranged from moderately burned samples to fully calcinated. Several bone types were processed, including fragments of cranium, vertebra, scapula, femur, and phalange. Bone fragments were crushed using a mortar and pestle until a sandy/powdery texture was achieved, and, based on the condition of the samples, one of four protocols was applied prior to Rapid DNA Identification.

- Protocol 1 (for bone fragments with minimal thermal damage). Approximately 5–10 mg crushed bone was placed in a 2-mL microcentrifuge tube and 120 μL of ANDE Bone Solution was added. The sample was vortexed for 5–10 sec and incubated for 1–2 min at room temperature. The entire solution was then pipetted onto an ANDE swab for Rapid DNA Identification.
- Protocol 2 (for bone fragments with moderate thermal damage). Approximately 50–100 mg crushed bone was placed in a 2-mL microcentrifuge tube and 120 μL of ANDE Bone Solution was added. The sample was vortexed for 5–10 sec and incubated at 56°C for 3 h. The solution (entire or aliquot) was then pipetted onto an ANDE swab for Rapid DNA Identification.
- Protocol 3 (for bone fragments with severe thermal damage and calcination). Approximately 500–700 mg crushed bone was placed in a 2-mL microcentrifuge tube and 1.4 mL of ANDE Bone Solution and 70 μL of 20 mg/mL Proteinase K (Qiagen, Inc.) were added. The tube was then placed into a thermomixer at 56°C and agitated for 90 minutes before incubating the sample overnight. The sample was then subjected to centrifugation at 37,565 × *g* for 1 min to pellet any remaining bone particulates. The supernatant was then concentrated using a 10KD Amicon® Ultra-0.5-mL Centrifugal Filter Unit (MilliporeSigma, Burlington, MA). The concentrated sample was pipetted onto an ANDE swab for Rapid DNA Identification.
- Protocol 4 (for bone fragments that were essentially ashes). Approximately 1–2 g crushed bone was placed in a 15-mL conical tube and 15 mL of ANDE Bone Solution and 200 μL of 20 mg/mL Proteinase K were added. The tube was placed in an incubated rotator (Benchmark Scientific Roto-Therm™) set to 56°C and 12 rpm overnight. The sample was then subjected to centrifugation at 3381 × *g* for 1 min to pellet any remaining bone particulates. The supernatant was then concentrated using a 10KD Amicon® Ultra-15-mL Centrifugal Filter Unit (MilliporeSigma, Burlington, MA). The concentrated sample was pipetted onto an ANDE swab for Rapid DNA Identification.

The bone processing procedures all require less than 5 min of handling, with incubation times ranging from 1 min (Protocol 1) to overnight (Protocols 3 and 4). Accordingly, total bone processing time ranged from 3 min (Protocol 1) to 12 h (Protocols 3 and 4). The processing was conducted outside a conventional DNA laboratory—only small instrumentation (a tabletop centrifuge and portable incubator) was utilized.

Antemortem Tissue Block. Paraffin-embedded tissue was cut into small fragments and placed in a 2-ml microcentrifuge tube. To the tube, 120 μL of ANDE Lysis Solution and 1 mg of Proteinase K were added and incubated overnight at 56°C. The lysate was purified using a guanidinium-based protocol as previously described (30). Purified DNA was pipetted onto an ANDE swab for Rapid DNA Identification.

Buccal samples. Buccal swabs were collected on an ANDE swab for Rapid DNA Identification.

Rapid DNA Identification

The ANDE Rapid DNA Identification system consists of a fully automated instrument, single-use microfluidic chips, and fully integrated expert system analysis software. Two types of chips were utilized: I-Chips for UHR samples and A-Chips for buccal swabs from family reference samples (FRS). Both chips perform automated purification, PCR amplification of 27 STR markers (the FlexPlex assay (17), which includes the 20 loci of the FBI's CODIS system), electrophoretic separation of amplified and fluorescently labeled fragments, and laser-based fluorescence detection. A major difference between the two chip types is that I-Chips perform a concentration step following DNA purification to maximize the quantity of DNA available for amplification (15,16,22).

Up to four swabs are placed into an I-Chip (five for an A-Chip), and the chip is inserted into the ANDE instrument. All required chemical reagents are preloaded into the chip, and, following processing, the DNA ID is analyzed and interpreted automatically by the expert system, generating DNA IDs in less than two hours without human intervention or interpretation. The ANDE instrument is ruggedized to Military Standard 810G for

transport and use outside of a controlled laboratory environment. The system generates run data consisting of (i) an allele table for all samples; (ii) .FSA file and .PNG files for each sample; and (iii) an .XML file for each passing sample. The .XML file consists of a list of called loci and alleles of the sample in a CODIS V3.2 format (31). Run data generated by the instrument are encrypted.

Three Rapid DNA instruments were utilized to process UHR samples. These instruments were operated in a rented recreational vehicle located outside the Sacramento County morgue. An additional instrument was utilized to process FRS. This system was operated in a Family Assistance Center located in a commercial building in Chico, California.

Kinship Analyses

The FAIRS software package imports and decrypts run data and performs all kinship analyses. FAIRS allows nontechnical users to perform the following types of familial searches: (i) search for references attempts to match valid references in the database to the selected victim: (ii) search for victims attempts to match valid victims in the database to the selected reference, and: (iii) comprehensive search that allows any victim or a reference to be searched against all samples in the database. The third approach is useful in a number of circumstances, including, for example, when it is suspected that multiple victims are biologically related.

The following is calculated for each victim–reference pair:

- The number of matched loci: A matching locus is defined as one in which at least one allele is shared between the victim and the reference.
- Combined Relationship Index (CRI): The CRI is the likelihood of the tested relationship for the victim–reference relative to that of two unrelated individuals. This is determined by calculating the likelihood ratio (LR) at each locus and then determining the CRI by multiplying the LR for each locus (32). ANDE FlexPlex27 includes two sets of loci that are genetically linked—loci SE33 and D6S1043; and D12S391 and vWA. The LR from one of the linked loci is included in the CRI. The kinship algorithms utilized by FAIRS meet the guidelines set by the American Association of Blood Banks (33).

When performing kinship analysis, the references with the highest CRI (and above a preselected threshold) are presented to the user. For victim searches, the victims with the highest CRI (above the threshold) are presented. Finally, the probability of relationship is calculated and is based on the CRI and an assigned prior probability. An operational prior probability of 0.2% was used early in the analysis when the number of victims was thought to be approximately 500. The prior probability was gradually increased to 5% as the number of missing diminished.

Fragment Size Distribution

Bone samples from remains that did not generate DNA IDs were processed utilizing Bone Protocol 4. DNAs were purified from the concentrated solutions using a guanidinium-based purification protocol essentially as described (30). Following library preparation, samples were sequenced (2x150 bp reads) on the NovaSeq 6000 instrument (Illumina, Inc., San Diego, CA). The resulting sequence data were aligned to the hs37d5 (GRCh37) human genome reference sequence. This produced a BAM-format file for each sample, which was then processed with CollectInsertSizeMetrics from Picard (34) to infer a fragment size distribution.

Results

Initial Characterization of Human Remains

Condition of remains. The vast majority of Camp Fire victims showed extensive thermal destruction, consisting mainly of burned bones ranging from single bones to partial skeletons most of which were calcined. Occasionally, bodies with the head, torso, and extremities with variable amounts of burned soft tissues and organs were recovered. Fragments of jaws and loose teeth were present with some of the remains. The condition of most individuals was analogous to remains that are commercially cremated (prior to their pulverization into ash), reflecting the immense heat and long duration of thermal exposure (Fig. 2). The majority of fire victims were recovered from residences, with the remaining recovered from vehicles or outside.

Age, sex, and stature. These determinations were complicated by the extensive thermal destruction suffered by the majority of victims. This resulted in the loss of much of the useful information from the skull, pelvis, and long bones for establishing biological characteristics. The fact that first responders frequently revisited certain recovery sites led to multiple separate case numbers for remains from a single victim, which further complicated efforts to reconstruct biological profile information, identifiable markings, and features. Although extensive searching for tattoos and body piercings was performed, the severe degradation made identification of these features impossible.

Commingling of remains. Three cases included two or three sets of commingled human remains. Two cases also included a

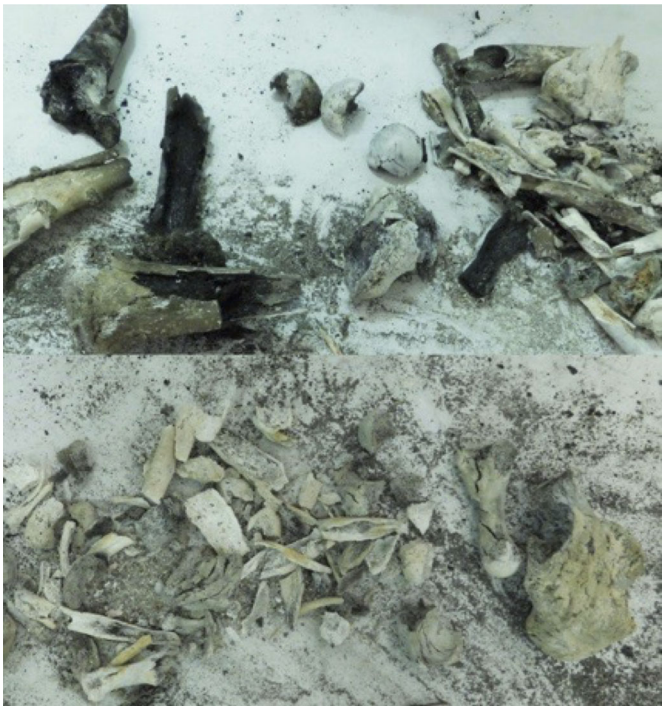

FIG. 2—*Photograph of recovered remains (primarily bone fragments) belonging to a single victim as confirmed by anthropology. [Color figure can be viewed at wileyonlinelibrary.com]*

set of human and one or two sets of animal remains, likely household pets. Commingling of human remains is a common occurrence in mass fatality scenes given that multiple fire victims often live at the same residence. These cases are complex for fire scenes, as human remains have been reduced to small calcined bone fragments, making it difficult to recognize skeletal duplication (i.e., evidence of the remains of more than one individual).

Personal effects. The remains were transported with various items that the recovery teams thought might help with identification. Personal effects for 29 of the victims recovered were transported to the morgue to assist with identification. Only two such effects, in both cases military dog tags, supported identification.

Conventional Identification Modalities

Radiographic features and hardware. Occasionally, antemortem injuries that are documented in medical records can assist with the identification of human remains. All Camp Fire remains were x-rayed to look for identifying features such as previous injuries or surgical hardware that might assist with identification. The state of the majority of the remains made it impossible to distinguish previous fractures or injuries. Twelve sets of remains had some type of co-recovered hardware, including shoulder implants and knee hardware, and two sets of remains had pacemakers. Most of the hardware and the pacemakers were so badly burned that they were not helpful with identification.

In two cases, surgical implants (artificial knee and shoulder) were identified, and in both of these cases, after extensive cleaning, serial numbers were identified that were compared to medical records to support the identification process. The local hospital survived the fire, but the medical record room was severely damaged. Furthermore, the lack of witnesses to provide information on medical history led to a massive cold call effort conducted by Butte County Sheriff's Office, California Department of Justice (Cal DOJ), and the Sacramento County Coroner's Office to track down medical records for these two individuals. Positive identifications were made for both cases.

Forensic Odontology. Forensic Odontology can be valuable in mass fatality incidents when evidence is degraded by fire or fragmentation (35). Indeed, in other recent northern California wildfires, forensic odontology accounted for more than 50% of victim identifications. With only 8 of 18 local dental offices surviving the fire, antemortem and postmortem dental comparisons were severely hampered. Although the lack of records limited the possibility of dental identifications, some dental offices utilized digital dental radiography and maintained records on backup drives or cloud storage, allowing images to be accessed for comparisons.

Victims examined for possible dental identification varied from severe superficial burning to near cremation. Remains were extremely brittle and fragile. Dental evidence varied from complete intact dentitions to root fragments. Evidence that survived relatively well included metallic dental restorations such as metal frameworks for removable partial dentures, metal posts within teeth, silver amalgam fillings in teeth, metal crowns, metal substructures for porcelain fused to metal crowns, entire porcelain fused to metal crowns, and root canal filling material (gutta-percha).

Tooth enamel is the hardest substance in the body (36), yet in nearly all the Camp Fire cases, no dental enamel was recovered. The temperatures of the fires and the length of time the fires burned greatly degraded the evidence, and individual roots of teeth were the only dental evidence available. Making dental identifications was not possible in the absence of unusual root morphology that had been captured in antemortem dental records. Instead, the dental identifications were based on teeth that had a unique dental restoration or an associated metal restoration. In total, dental evidence was present in 35 of 60 sets of remains examined by the dental team. Taken together, the extensive sample degradation and absence of dental records led to only 15 identifications being made by forensic odontology.

Fingerprint and Thumbprint Analysis**.** Due to the degradation of most of the remains, only five victims were identified by fingerprints. Four victims were positively identified biometrically through the Livescan fingerprint database, maintained statewide by Cal DOJ. A fifth identification was made by thumb print casting (37). The resulting print was submitted to the Cal DOJ Latent Prints Unit for comparison to a print from a tentative identification made by the Butte County Sheriff's Department.

Rationale for Rapid DNA Identification

Taken together, the series of conventional evaluations described above is typical following a mass casualty event, but their success rates were low due to the severely degraded conditions of the Camp Fire remains. With only 22 of 84 victims identified by conventional modalities, DNA analysis represented the last opportunity to identify the remaining victims. However, though DNA analysis of STRs is an extremely powerful and reliable tool for human identification, its application presents substantial challenges.

Although STR analysis has been established for more than three decades, current approaches demand numerous manual procedures and decision/interpretation points by expert analysts and a sophisticated laboratory infrastructure with specialized instruments. Even with semi-automated sample batching and processing, a substantial sample backlog has developed, generally requiring months or years to obtain DNA results (38). In particular, it has been estimated that over 400,000 sexual assault evidence kits are backlogged in the United States alone (39). Based on the complexity and time required for DNA analysis, DNA-based identification of human remains has typically been the biometric of last resort following a mass disaster.

The combination of prolonged sample delivery and processing times, sample backlogs, and new applications has inspired a desire for field-forward systems that would accelerate and simplify the generation of DNA IDs. Rapid DNA Identification is based on the automation and integration of four laboratory processes: purification of genomic DNA from a sample; rapid multiplexed amplification of a set of 27 STR loci (amelogenin, three Y-STR loci, and 23 autosomal loci); separation of the resulting STR amplicons by polyacrylamide gel electrophoresis; and data processing and locus and allele assignation using an onboard expert system (17,40). These processes are performed in a single instrument that accepts a single-use cartridge containing all required reagents. To permit operation in the field, outside of controlled laboratory environments, the instruments are ruggedized to protect against shock and vibration during transport and operation and the cartridges are stable for six months at room temperature (15). Advances in Rapid DNA (41), including the FBI's National DNA Index System approval for automated processing of buccal swabs in 2018 (40) and the demonstration of the effectiveness of Rapid DNA Identification for the generation

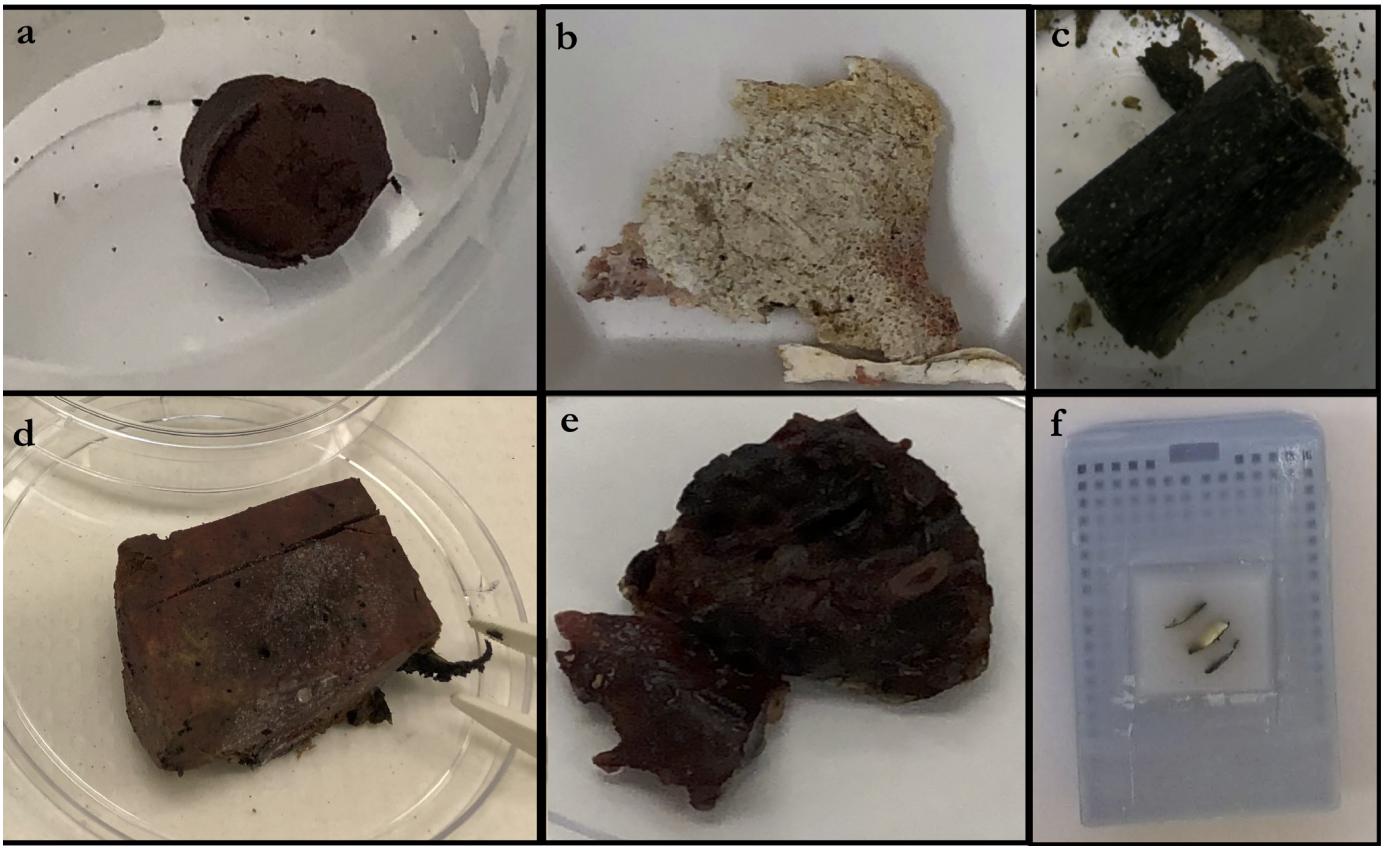

FIG. 3—*Representative photographs of sample types selected for Rapid DNA Identification. All samples generated full FlexPlex DNA IDs (24 of 24 STR loci for females and 27 of 27 loci for males). Cardiac blood clot (a), bone thought to be a fragment of a scapula (b), bone thought to be a fragment of a femur (c), liver section removed from the bulk organ (d), muscle tissue of unknown origin (e), and antemortem tissue block (f). [Color figure can be viewed at wileyonlinelibrary.com]*

of DNA IDs from bone, teeth, and several other tissue types from human remains (16,21), strongly suggested that the technology might be applicable to mass casualty events. It was reasoned that DNA IDs from UHR and from FRS could be generated quickly outside the forensic laboratory and analyzed using automated kinship software.

Rapid DNA Identification of Victims

A total of 69 sets of remains were subjected to Rapid DNA Identification using the ANDE system. Of these, 62 (89.9%) generated DNA IDs that were utilized for familial searching, and essentially all these DNA IDs were produced within hours of sample receipt. Representative photographs of sample types selected for Rapid DNA Identification are shown in Fig. 3. One measure of the effectiveness of Rapid DNA Identification is the relative number of samples yielding full DNA IDs (i.e., those containing all 20 of the STR loci selected by the FBI for human identification—also known as the CODIS core loci [(42)]) and partial DNA IDs (those containing less than 20 of the CODIS loci). Table 1 shows that 95.2% of the DNA IDs contained 20 or 19 CODIS loci, a remarkably high percentage, particularly considering the degraded state of the remains. Samples identified using blood, liver, or tissues typically were obtained from remains consisting of small portions of the torso (Fig. S1), while samples using bone were obtained from small bags of bones isolated from surrounding building debris (Fig. S2). Note that the partial DNA IDs contained alleles in an additional 2 or 3 FlexPlex loci (17). The FlexPlex assay contains the CODIS core loci and several additional informative STR loci utilized in the United States and worldwide but not present within the CODIS 20 loci (including D6S1043, SE 33, and Penta E). The FlexPlex loci were utilized in all kinship calculations and were particularly useful in the one UHR sample that generated a CODIS 12 partial DNA ID (due to a relatively uncommon SE 33 allele present in a parent–child pair).

TABLE 1—*Sample types utilized to generate DNA IDs for familial searching. For each sample type, "Total DNA IDs" indicates the number of remains that generated DNA IDs that were used in familial searching.*

Sample type	Total DNA IDs	CODIS 20	CODIS 19	CODIS 18	CODIS 17–12
Blood/clot	37	30	6	1	0
Bone	10	8	1	1	0
Liver	7	4	1	0	2
Muscle	5	3	1	0	1
Tissue[†]	2	2	0	0	0
Brain	1	1	0	0	0

"CODIS 20" indicates those with full CODIS DNA IDs, "CODIS 19" and "CODIS 18" indicate the subset missing one or two CODIS loci, respectively, and CODIS 17-12 indicate the subset missing three to eight CODIS loci.

[†]Two samples consisted of soft tissue of undetermined origin.

The immediate availability of DNA IDs from UHR led to an aggressive law enforcement effort to collect FRS. The Sheriff of

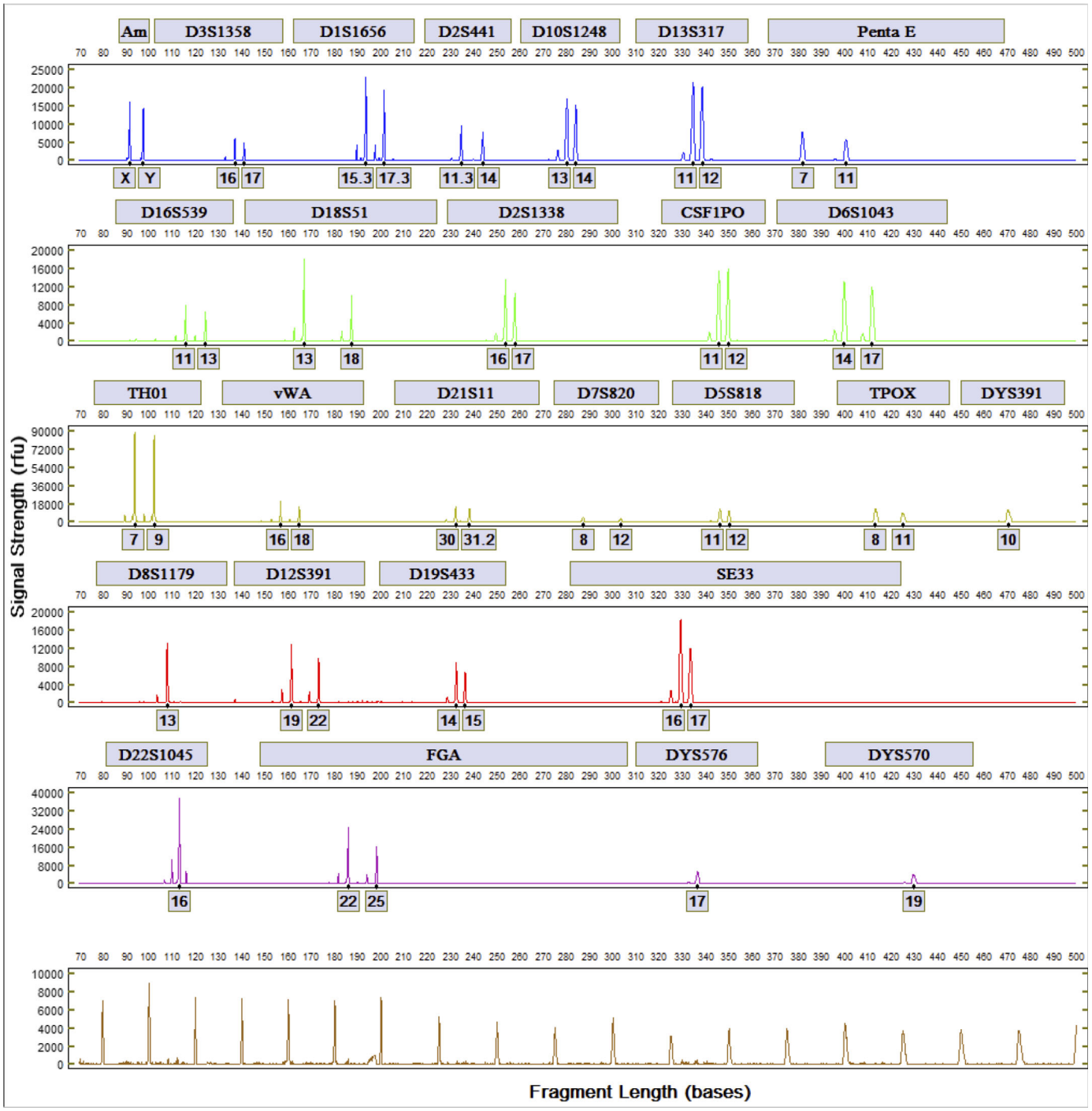

FIG. 4—*Representative full DNA ID of a male victim generated from Rapid DNA Identification of the bone fragment of Fig. 3c. [Color figure can be viewed at wileyonlinelibrary.com]*

Butte County used daily press briefings and organized a program to enable donors to provide buccal swabs across the nation. DNA IDs from a total of 255 FRS were placed into the 2018 Camp Fire DNA database. Of these, 219 were processed in the ANDE system and 36 were generated by conventional laboratory methodologies. Two antemortem tissue block samples were utilized to generate DNA IDs for searching.

Remains from 58 individuals were identified by searching against the FRS database. A full DNA ID was generated from processing the bone fragment (as shown in Fig. 3c) and following Bone Protocol 2 described in Materials and Methods (Fig. 4). Initial identifications were made on the fourth day of the fire (the day Rapid DNA systems were deployed), and most identifications were made within two weeks (by which time the vast majority of remains and FRS were received by the morgue), well before the fire was brought under control.

Table 2 shows the familial relationships that allowed the identifications to be made. Of note, two identifications were made by searching of victim DNA IDs against those generated from antemortem tissue blocks. All but two of the 58 individuals were identified based on likelihood ratios. Likelihood ratios were used to express the probability of relatedness, which in almost all

TABLE 2—*Types of relationships identified by familial searching of rapid DNA IDs from UHR and FRS.*

Relationship	Number of cases
Parent–child	36
Sibling–sibling[†]	15
Antemortem (self)[‡]	2
Half-sibling	1
Grandparent–grandchild	1
Monozygotic twin	1
Avuncular	1
Multiple[§]	1

Of the 62 sets of remains for which DNA IDs were generated, 58 identifications were made based on FRS (56 cases) and antemortem sample (2 cases) searching. Identifications in the remaining four cases could not be made based on DNA IDs as appropriate FRS samples were unavailable. These samples have been subjected to DNA sequencing and are being evaluated using forensic genealogic analyses.

[†]One sibling pair was monozygotic, generating a Combined Relationship Index of greater than 64 quadrillion.

[‡]Based on searching of DNA IDs generated from tissue blocks against the UHR DNA ID database.

[§]Based on an initial possible match to a half-sibling followed by the addition of an avuncular relationship in the kinship calculations.

cases were greater than 99.9%. Eight of these identifications were confirmed by identifications made using other modalities (seven by odontology and one by fingerprints).

Samples from seven sets of remains did not generate DNA IDs. Bones from these cases were subjected to manual DNA processing, and picogram quantities of DNA were generated for all samples. The average fragment size was typically approximately 100 base pairs (data not shown) as analyzed by processing the sequencing results using *CollectInsertSizeMetrics* (34). This explains the samples' inability to generate DNA IDs; most STR alleles are between 100 and 500 base pairs.

Discussion

Rapid DNA Identification was successfully integrated into real-time victim identification during the 2018 Camp Fire. Due to the extensive degradation of the remains, DNA analysis was typically the only effective modality. In approximately 90% of cases, Rapid DNA Identification generated DNA IDs and was the primary identification modality of the Camp Fire, with 58 victims identified (as compared to 22 by conventional modalities). As the remains in the Camp Fire were at the extremes of degradation, it is expected that in other disasters (e.g., earthquakes, tsunamis, terrorist attacks), remains will be less degraded and Rapid DNA Identification will be readily applicable to these mass casualty events.

Unlike previous disasters for which DNA identification may have required months to years (43), the ability to generate DNA results quickly had a major impact on process flow in the morgue. When DNA IDs were generated on particularly degraded remains, odontologists and pathologists were able to focus their efforts on other cases with potentially better outcomes. In many disasters, governments do not permit samples to be shipped to foreign laboratories, and Rapid DNA equipment can be employed locally, near the disaster site, by nontechnical operators. Most importantly, the technology enables rapid identification, critical to providing support to family members. In previous disasters, family members have often waited years for identifications; the pain of losing a family member need no longer be amplified by the pain of waiting for a definitive identification. The first successful deployment of Rapid DNA Identification at a mass disaster augurs well for continued expansions of the use of the technology in mass casualty events and, more broadly, in law enforcement (41) and human trafficking applications (44).

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Fig S1. Remains from a male victim (a) and corresponding DNA ID generated (b). Although severely degraded, remains of a torso with associated flesh routinely allowed generation of Rapid DNA IDs.

Fig S2. Remains of a female victim (a) and corresponding DNA ID generated (b). In the field, construction materials such as drywall and wood can mimic the appearance of human bone.