IN REPLY REFER TO:

United States Department of the Interior
FISH AND WILDLIFE SERVICE
Office of Law Enforcement
Clark R. Bavin
National Fish and Wildlife Forensics Laboratory
1490 East Main Street
Ashland, Oregon 97520

June 13, 2018

GENETICS EXAMINATION REPORT

Agency:
USFWS/LE, Billings
2900 4th Avenue North
Suite 301
Billings, MT 59101

Lab Case #: 18-0134
Examiner: Straughan
Agency Case #: 2018602411
Investigator: Conrad
Subjects:

Case Title: WOLF HYBRID

EVIDENCE RECEIVED:

The following evidence was received in the Evidence Unit of the Laboratory on June 01, 2018, and was transferred to the undersigned examiner on June 01, 2018:

LAB-1: "2 ml vial containing dessicant and muscle tissue" [ST#47861;Item#1]
LAB-2: "2 ml vial containing dessicant and muscle tissue" [ST#47861;Item#2]
LAB-3: "50 ml vial containing dessicant and muscle tissue" [ST#47861;Item#3]

EXAMINATIONS REQUESTED:

The submitting officer, Special Agent Shawn Conrad, requested that the species origin of the individual represented by LAB-1, LAB-2 and LAB-3 be determined. Agent Conrad confirmed that all three samples originated from the same individual canid carcass and matching would not be necessary.

EXAMINATIONS CONDUCTED:

Species Identification by Mitochondrial DNA
A segment of the mitochondrial DNA (mtDNA) control region of LAB-1, LAB-2 and LAB-3 was amplified by PCR and subject to DNA sequence analysis. The resulting sequences were compared to those from the National Fish and Wildlife Forensics Laboratory (NFWFL) canid database.

Species Identification by Nuclear DNA
The DNAs from LAB-1, LAB-2 and LAB-3 were characterized by short tandem repeat (STR) analysis at eight autosomal nuclear loci designated as 109, 123, 172, 200, 204, 225, 250 and 377 and at seven loci located on the Y-chromosome designated as 34A, 34B, 41A, 41B, 35.4, 650 79.2I and 650 79.2II.

Examiner's Initials

The resulting STR genotypes and Y-chromosome haplotypes were compared to those from the National Fish and Wildlife Forensics Laboratory (NFWFL) canid database. Assignment tests were used to determine if the STR genotypes of LAB-1, LAB-2 and LAB-3 were most likely to occur in western North America gray wolf (*Canis lupus*) populations, eastern North America gray wolf populations, Mexican wolves (*C. l. baileyi*), domestic dogs (*C. familiaris*), gray wolf hybrids (*C.lupus* x *C. familiaris*), red wolves (*C. rufus*) or coyote (*C. latrans*) populations.

EXAMINATION RESULTS:

Species Identification by Mitochondrial DNA
The mtDNA sequences of LAB-1, LAB-2 and LAB-3 were identical to reference sequences originating from western gray wolves (*Canis lupus*; GenBank Acc# AF005312) at 232/232 base pairs.

Species Identification by Nuclear DNA
The autosomal STR genotypes of LAB-1, LAB-2 and LAB-3 were consistent with originating from gray wolves from the Northern Rocky Mountains and Alaska. The probability that the individual represented by LAB-1, LAB-2 and LAB-3 originated from domestic dog or coyote is less than 0.002 with an exclusion probability threshold of 0.05.

No Y-STR haplotype was obtained from LAB-1, LAB-2 or LAB-3 indicating that this animal was a female.

CONCLUSIONS:

The individual represented by LAB-1, LAB-2 and LAB-3 was a female gray wolf from the Northern Rocky Mountains (*Canis lupus*). It is the opinion of the undersigned examiner that this individual was not of either a gray wolf x coyote, or gray wolf x domestic dog, origin.

DISPOSITION OF EVIDENCE:

The evidence item was transferred to the Evidence Unit pending return to the submitting agency.

Digitally signed by DYAN STRAUGHAN
DN: c=US, o=U.S. Government, ou=Department of the Interior, ou=U.S. Fish and Wildlife Service, cn=DYAN STRAUGHAN, 0.9.2342.19200300.100.1.1=14001000664468
Date: 2018.06.13 14:33:30 -07'00'

Dyan J. Straughan
Forensic Scientist