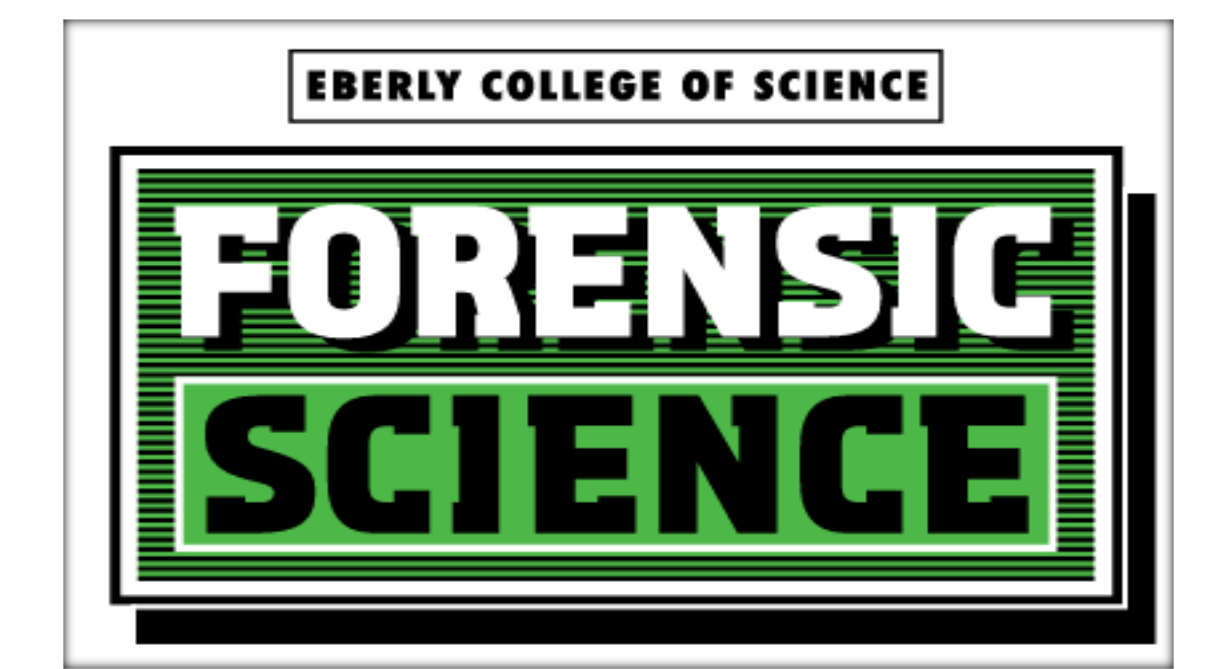# Ferrets Or Us: How Similar Are We?

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ABSTRACT

The objective of the study was to investigate the cross reactivity of blood and saliva samples from various domestic ferrets on immunoassay cards available for the detection of human blood and saliva samples. Further studies using human specific primers in the AmpF/STR® Identifiler™ and AmpF/STR® Yfiler™ kits from Applied Biosystems were initiated to explore homology in the STR sequences between humans and ferrets.

INTRODUCTION

It has been reported that some of the immunochromatographic devices currently available in the forensic community for the detection of human blood can also react with blood from domestic ferrets. Therefore, most of the forensic crime laboratories do not use these assays as confirmatory tests for the detection of human blood. The three immunoassay devices chosen for this study were the following: Hexagon OBTI obtained from BLUESTAR® Forensics, RSID™ Blood, RSID™ Saliva obtained from Independent Forensics and ABAcard® Hematrace® obtained from Abacus Diagnostics. All three assays are based on the fundamental concepts of immunology and chromatography. The three devices are noted for being highly sensitive and specific for the detection of human blood. However, the manufacturers of the ABAcard® Hematrace® and Hexagon OBTI note that positive results may also be obtained with ferret blood. The RSID™ Blood card is not known to react with ferret blood.

The RSID™ Saliva card is used for the detection of human saliva. The SALIgAE® Tube Test, obtained from Abacus Diagnostics, is used to determine the possible presence of trace levels of saliva from unknown samples. Amylase Radial Diffusion Test, a presumptive test for the detection of saliva, is one of the methods currently used to identify the enzyme amylase, in a suspected saliva stain. These saliva tests were conducted using saliva samples from another ten male and female ferrets.

The current study was undertaken to determine if blood and saliva samples from domestic ferrets react with any of the tests currently used to detect the presence of blood and saliva samples from humans.

MATERIALS AND METHODS

Blood samples from 6 male and 4 female ferrets were received on FTA papers. In addition, saliva samples were received from another 6 male and 4 female ferrets on buccal swabs. All of the ferret samples were obtained from Lampire Biological Laboratories.

All analyses on ferret and human blood and saliva samples were performed according to the manufacturers' instructions or standard operating protocols. The following tests were completed: ABAcard® Hematrace®, Hexagon OBTI, RSID™ Blood, RSID™ Saliva, SALIgAE®, and Amylase Radial Diffusion. Samples were extracted by the Qiagen EZ1 DNA/ BIOROBOT®, FTA Paper, and Organic extraction methods. Relevant DNA blood extracts were quantified by the Quantifiler™ Human DNA Quantification Kit. All extracted samples were amplified by both the AmpF/STR® Identifiler™ and AmpF/STR® Yfiler™ kits, followed by capillary electrophoresis on the 3130xl Genetic Analyzer. The data was analyzed using SoftGenetics GeneMarker® HID Version 1.7.

RESULTS AND DISCUSSION

The research indicated that the devices which utilize human hemoglobin for the detection of human blood also cross-reacted with ferret blood samples (e.g. ABAcard® Hematrace® and Hexagon OBTI). Hexagon OBTI cards demonstrated stronger positive reactions than the ABAcard® Hematrace® devices. The assay, which is designed to detect human glycophorin A, appeared to be specific for human blood and showed no reactivity with ferret samples (e.g. RSID™ Blood). All ferret saliva samples tested by RSID™ Saliva, SALIgAE® Tube Test and Amylase Radial Diffusion showed no results. In addition, quantified DNA extracts from ferret blood showed no detectable results by the Quantifiler™ Human DNA Quantification Kit.

Table 1: Immunoassays for Blood and Saliva

Ferret Samples	Gender/Color	ABACard®	RSID™ Blood	Hexagon OBTI	RSID™ Saliva
#1	Female/Siamese	Weak +	-	Strong +	-
#2	Female/Albino	Strong +	-	Strong +	-
#3	Female/Sable	Strong +	-	Strong +	-
#10	Female/Chocolate	Strong +	-	Strong +	-
#4	Male/Chocolate	+	-	Strong +	-
#5	Male/Albino	+	-	Strong +	-
#6	Male/Sable	+	-	Strong +	-
#7	Male/Siamese	+	-	Strong +	-
#8	Male/Cinnamon	+	-	Strong +	-
#9	Male/Violet	+	-	Strong +	-
#11	Liquid ferret blood sample	+	-	Strong +	Not Tested
Positive Control	Human blood and saliva samples	Strong +	Strong +	Strong +	Strong +

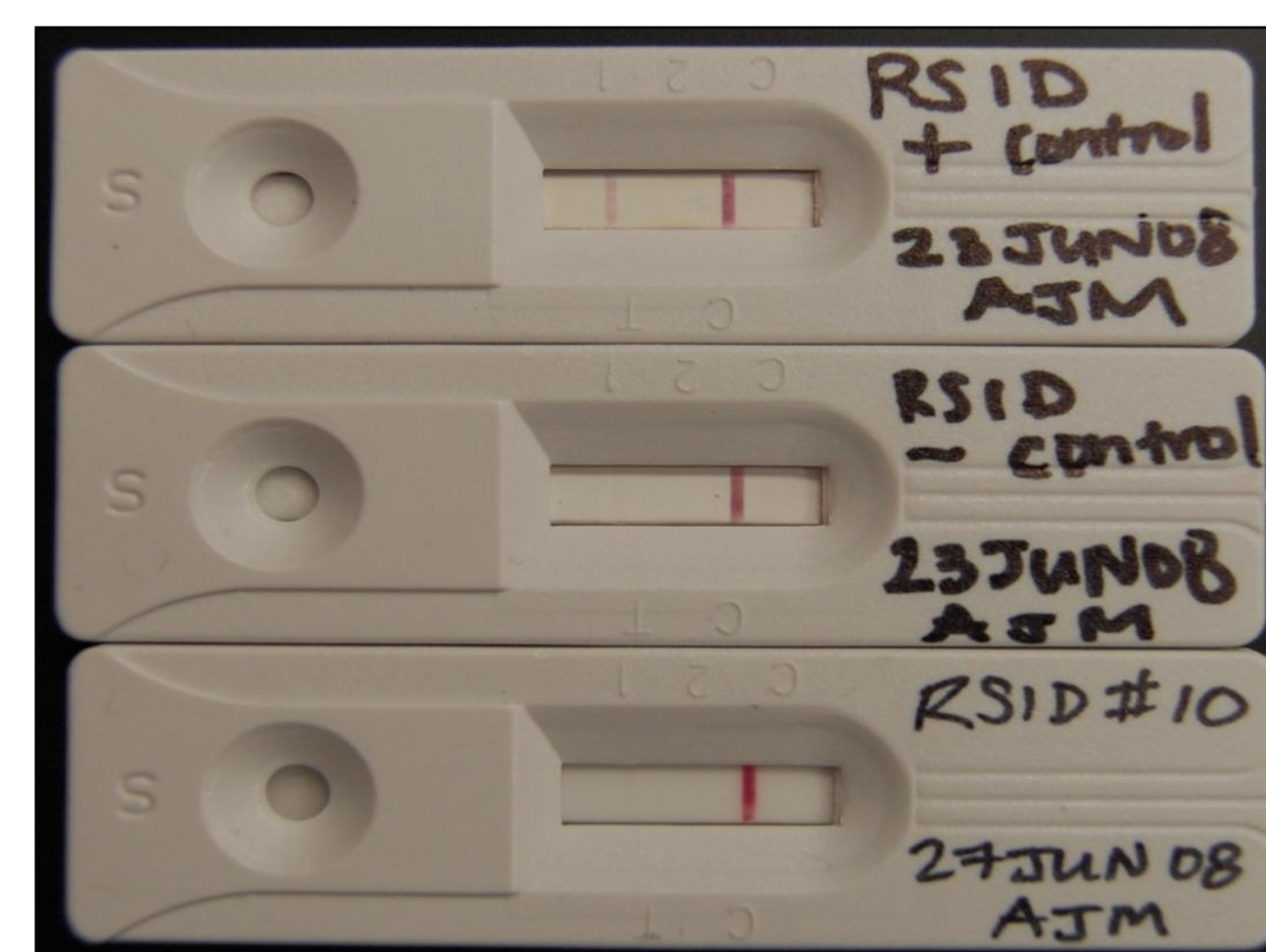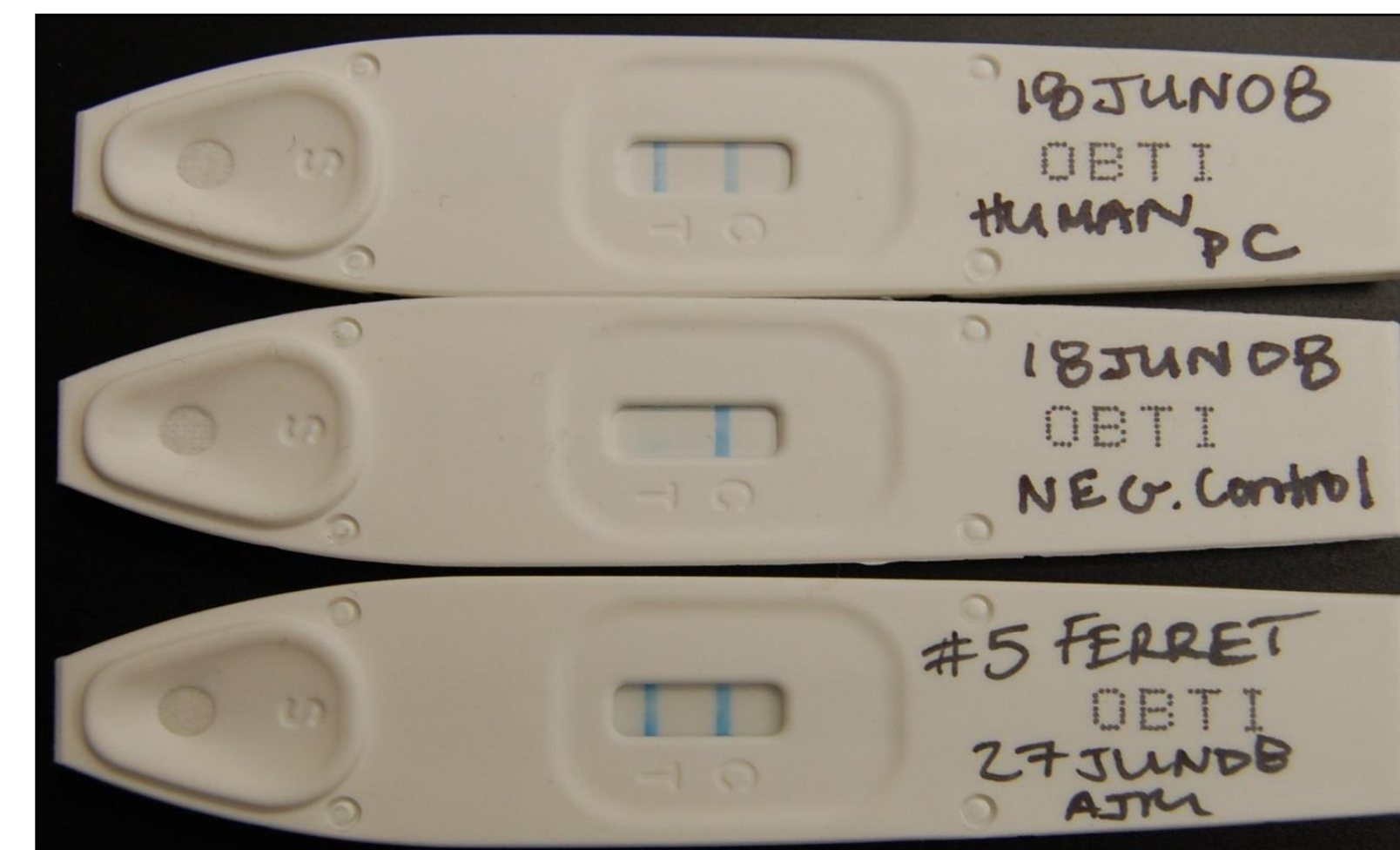**Fig.1: ABAcard® Hematrace® Test** showing positive results with ferret blood

Fig.2: Hexagon OBTI Test showing strong positive results with ferret blood

Fig.3: RSID™ Blood Test showing no reaction with ferret blood
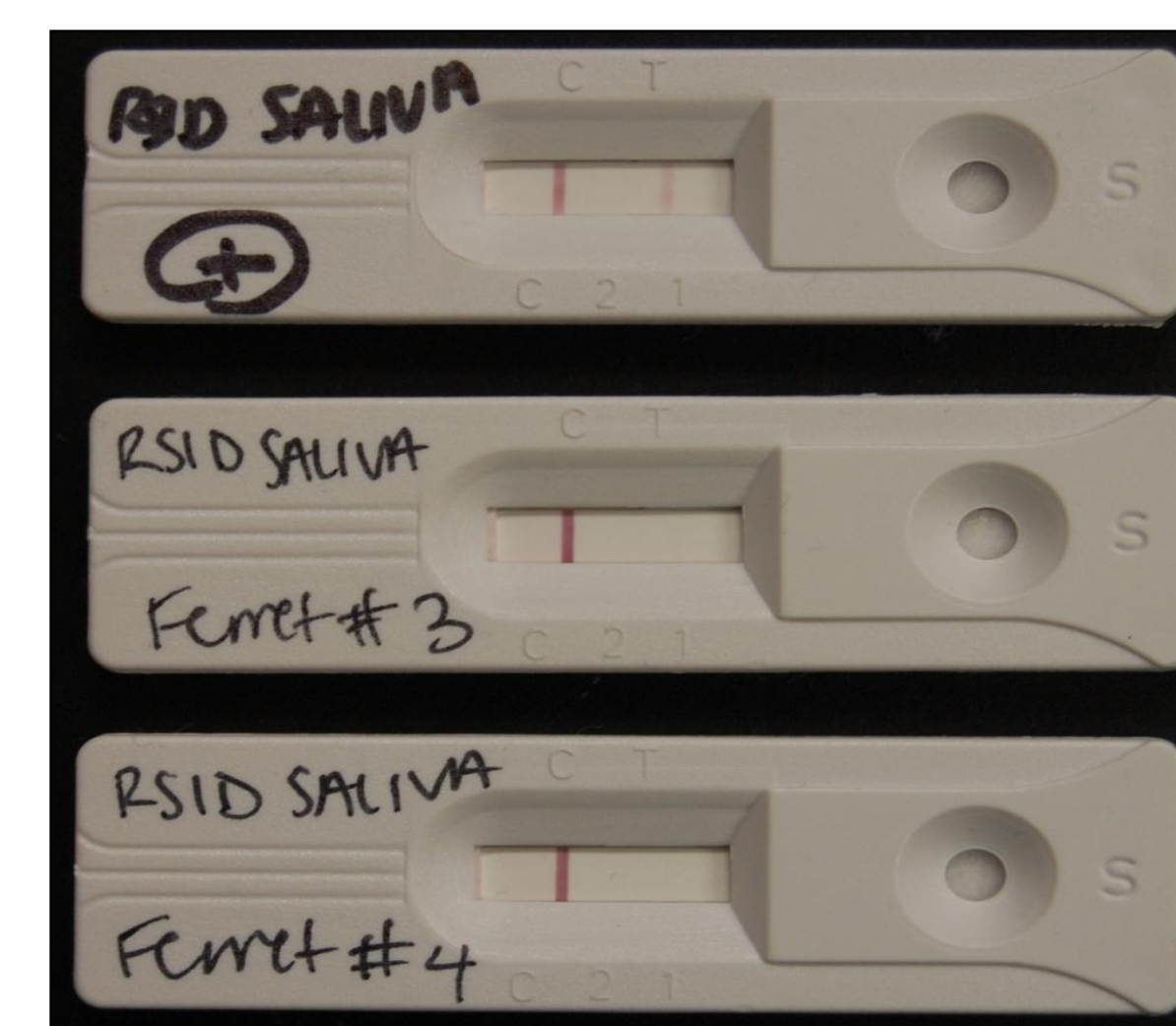

Fig.4: RSID™ Saliva Test showing no reaction with ferret saliva

Fig.5: SALIgAE® Tube Test showing no reaction with ferret saliva within ten minutes

No alleles were detected from 19 blood and saliva ferret samples analyzed by the AmpF/STR® Identifiler™ and AmpF/STR® Yfiler™ kits. A single allele, at the locus DYS365, was detected from one male ferret blood sample. All amplifications were performed in 12.5µl reaction volume.
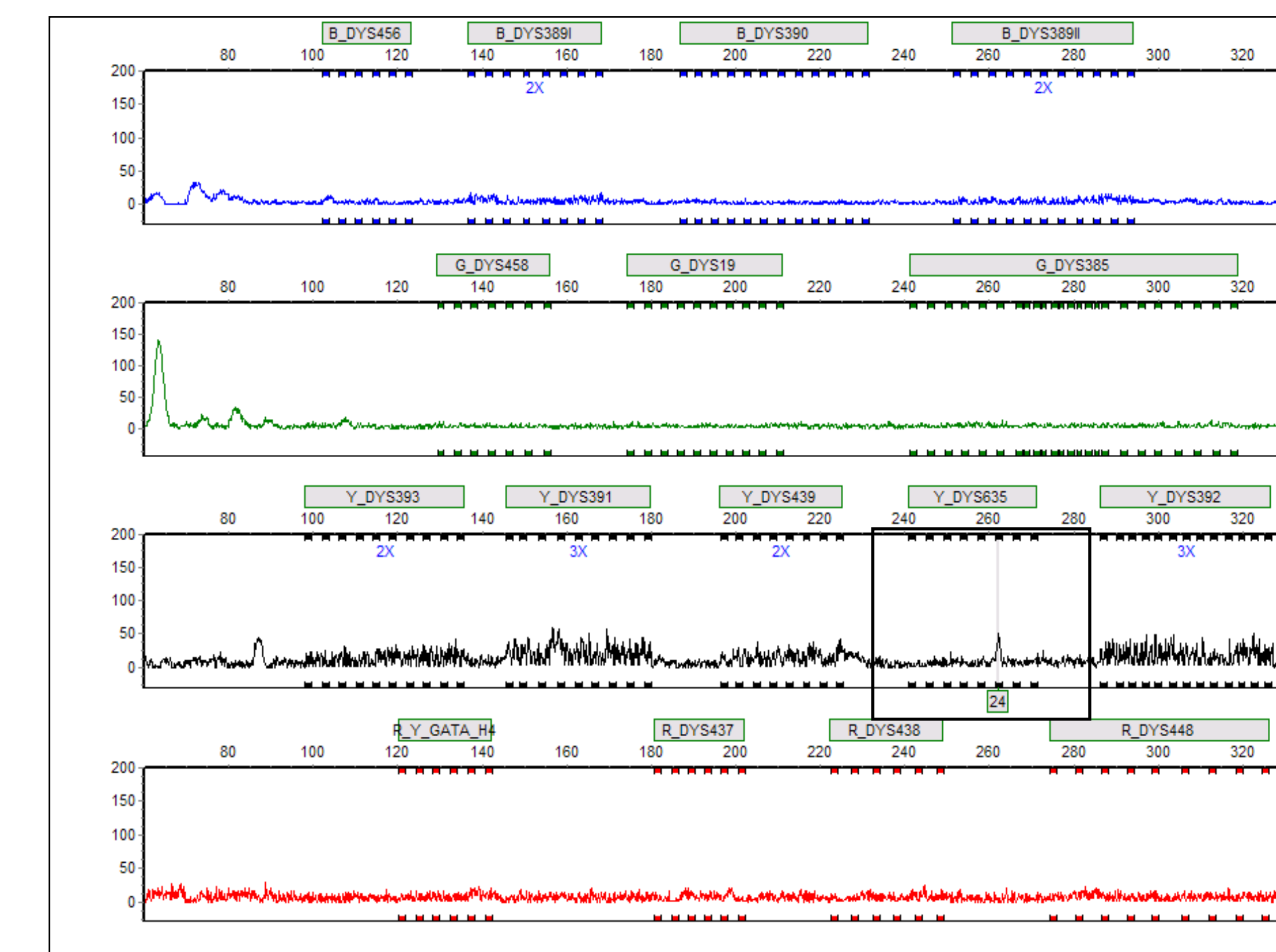

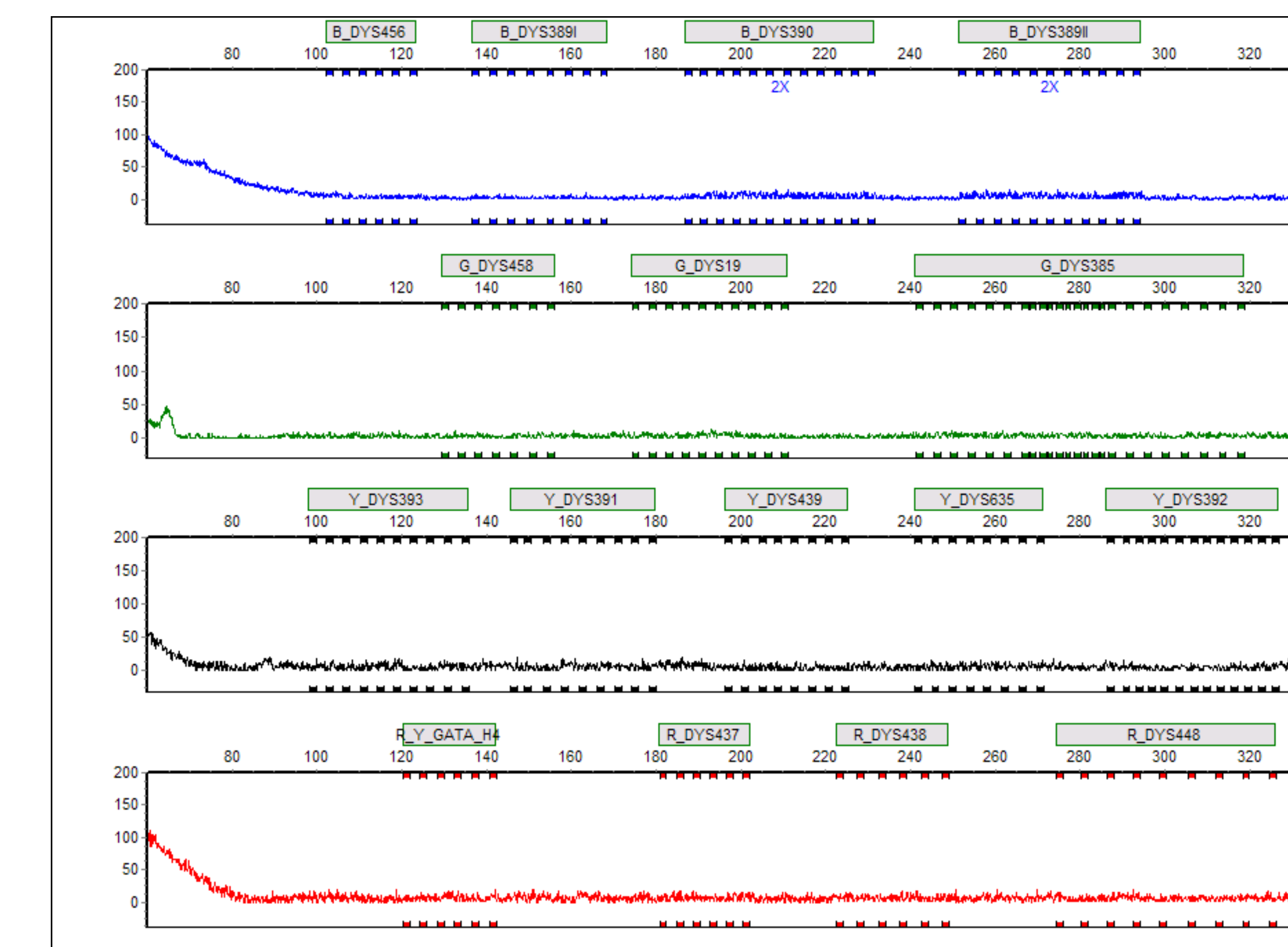**Fig.1:** Male ferret sample showing one allele at the DYS365 locus with Yfiler™ primers

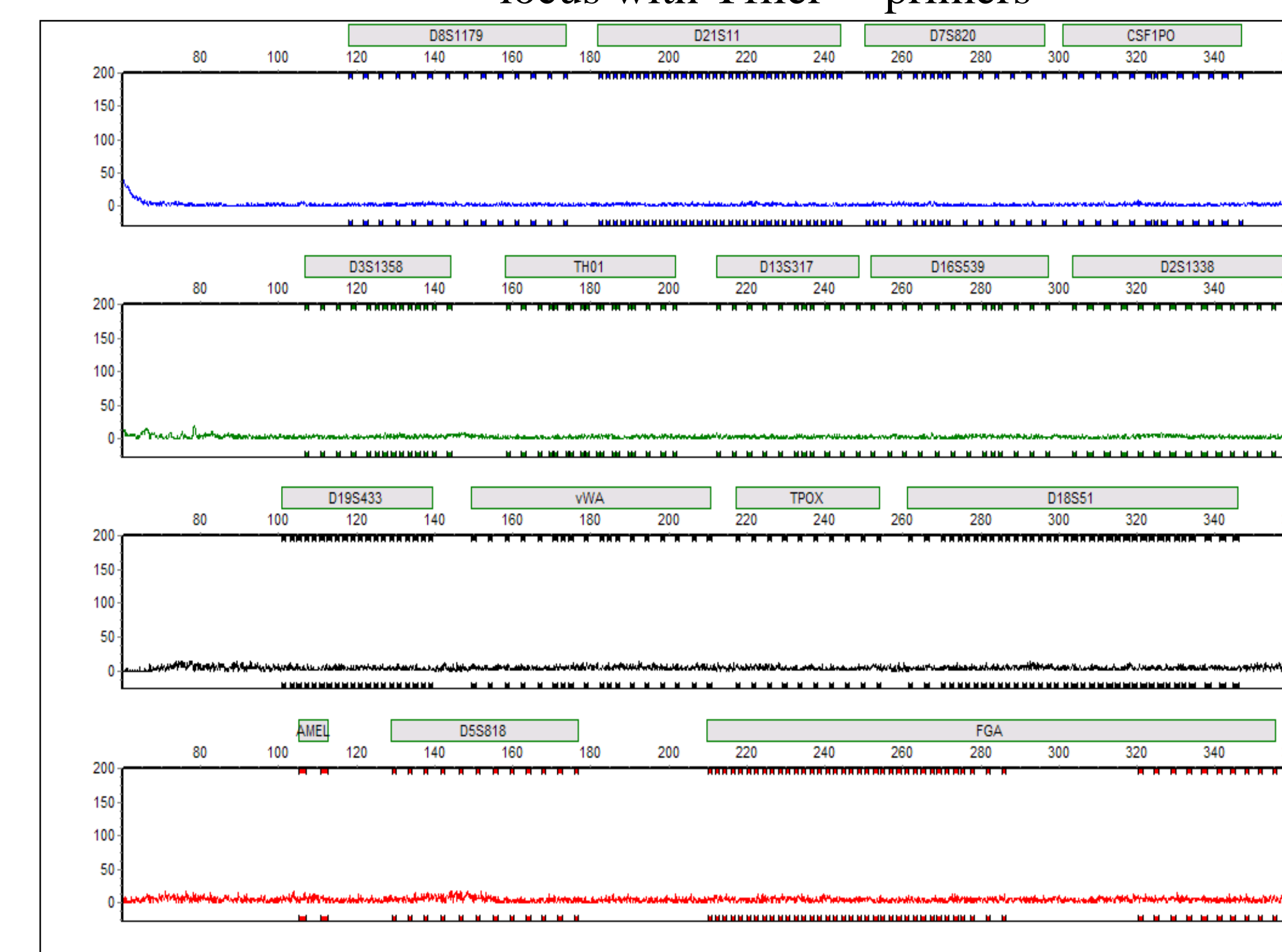**Fig.2:** Male ferret blood sample showing no results with Yfiler™ primers

Fig.3: Male ferret blood sample showing no results with Identifiler™ primers
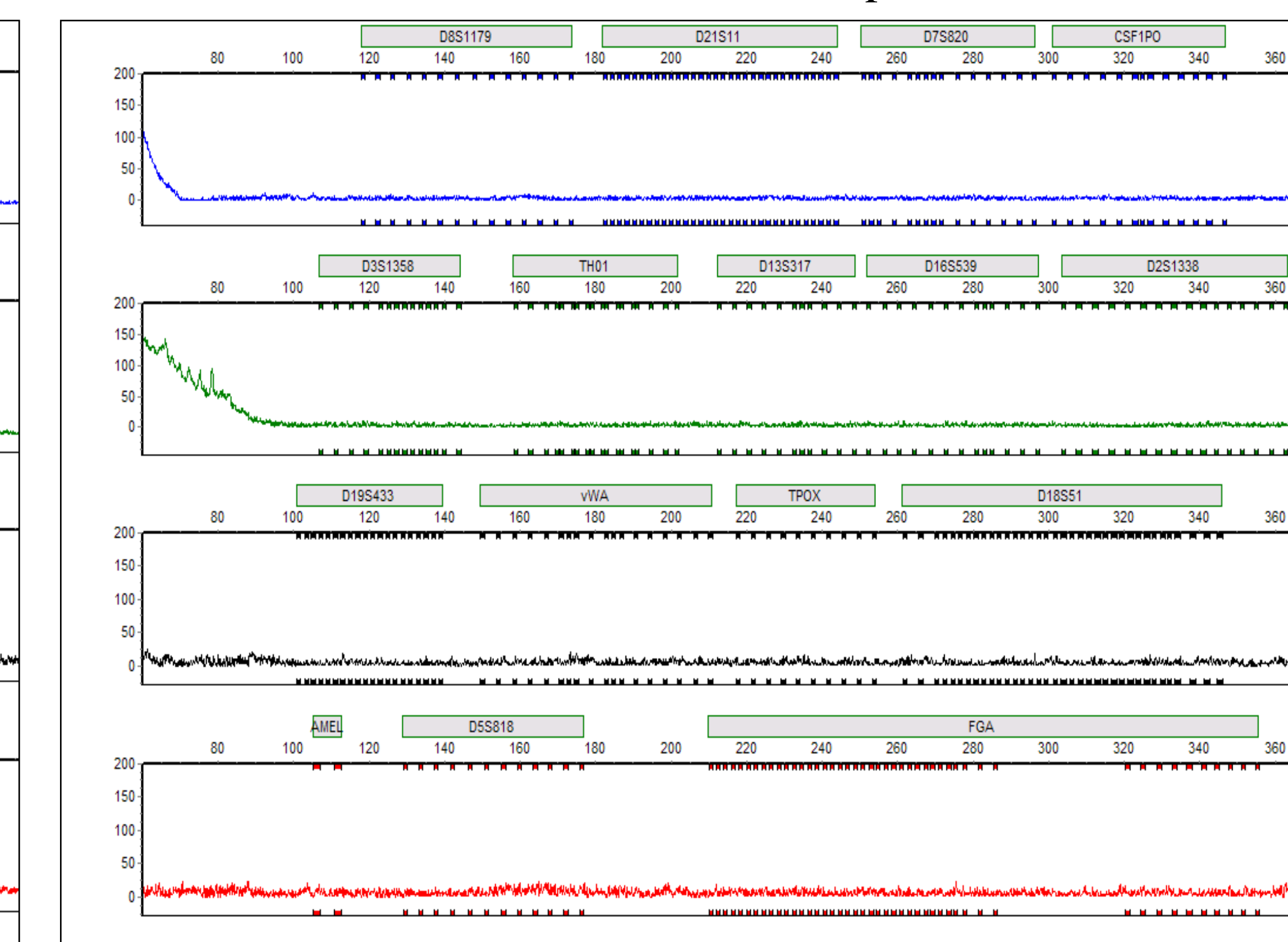

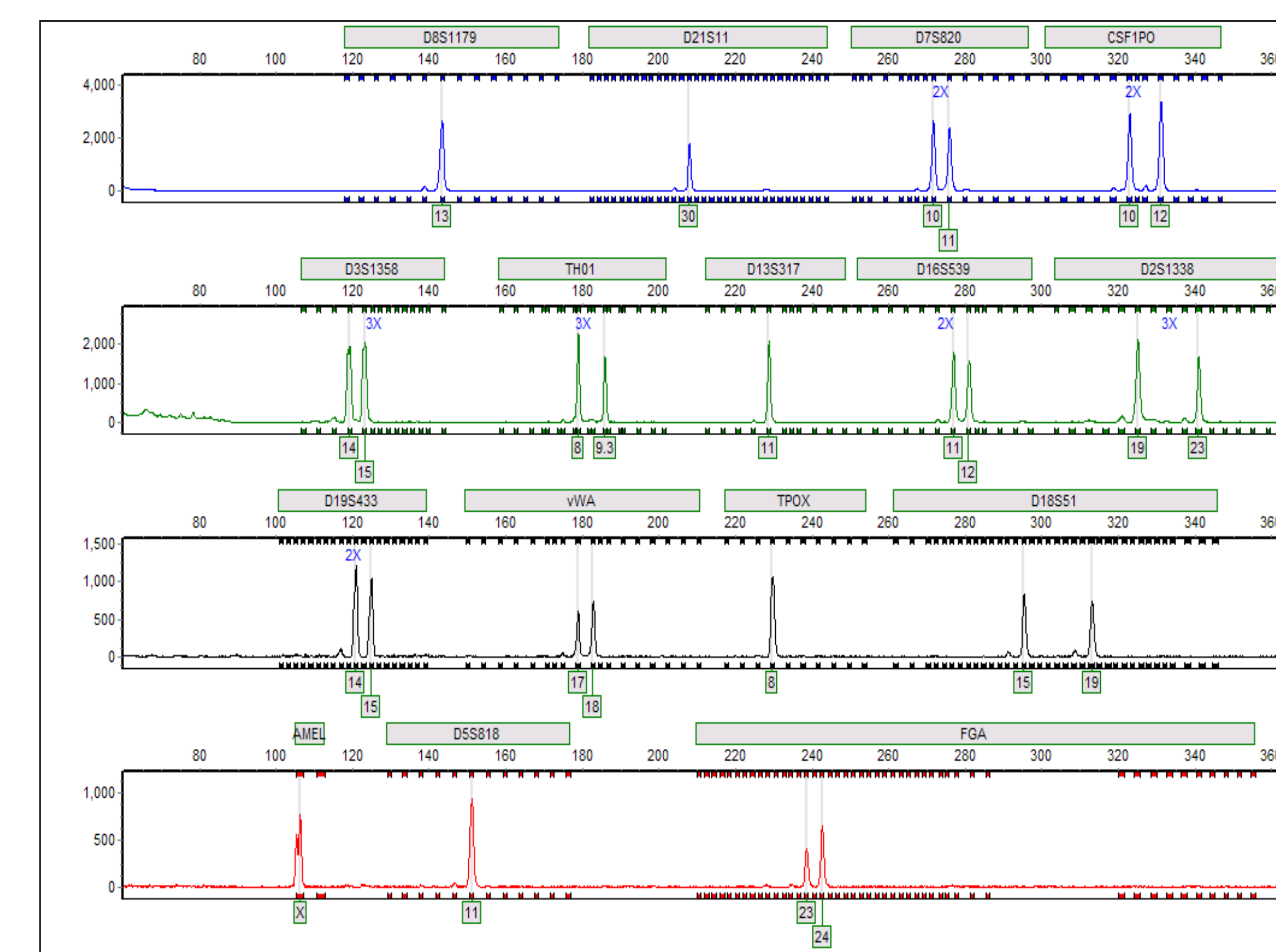**Fig.4:** Female ferret blood sample showing no results with Identifiler™ primers

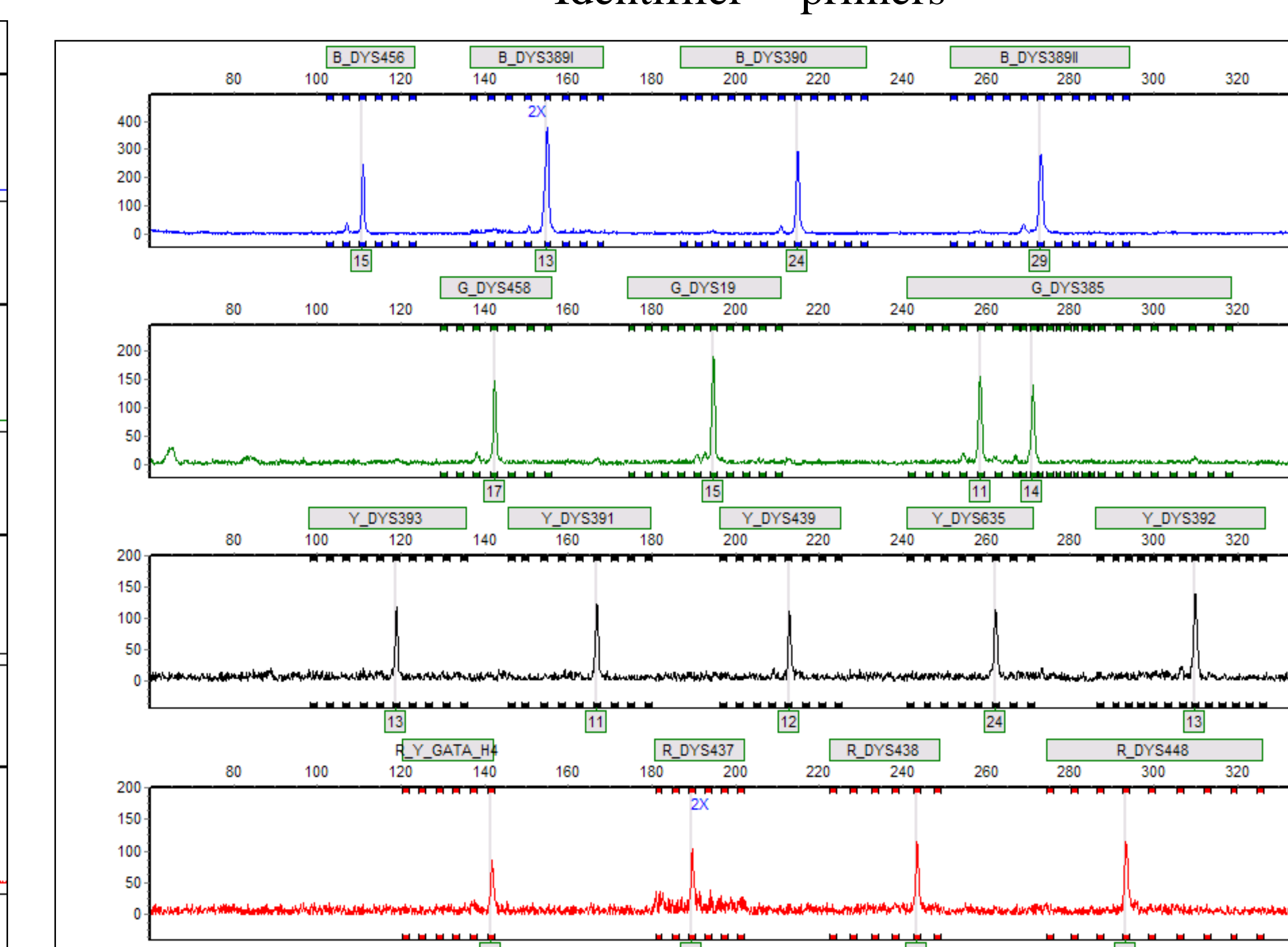**Fig.5:** Human positive control with Identifiler™ primers

Fig.6: Human positive control with Yfiler™ primers

CONCLUSION

While little to no reaction occurred between DNA from ferrets and human specific primers, a human profile could easily be distinguished from profiles obtained from ferret samples. Even though it is rare to encounter ferret blood at most crime scenes, it is possible to differentiate human bloodstains from ferrets and other animals using a combination of techniques.