

## **Practice Exercise**

### **Determining species identification from gel images**

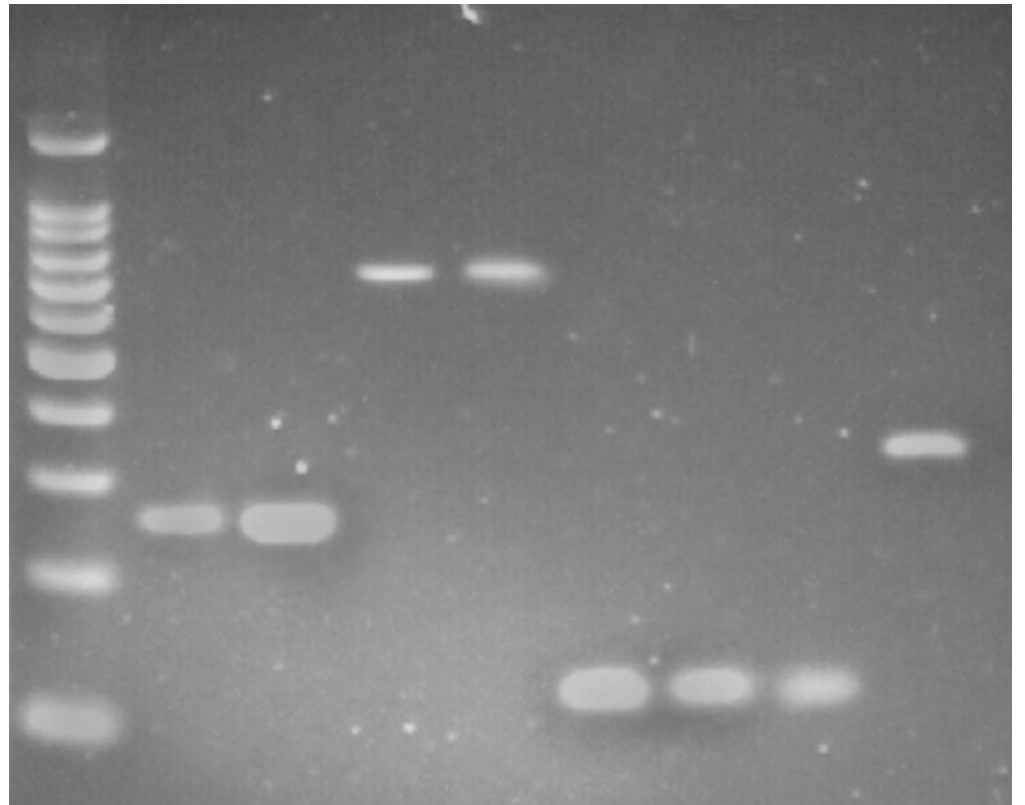
I have used 4 different PCR identification protocols on a set of 7 unknown shark samples. Using these gel images and the reference information provided please try to assign species identification to each sample.

- Samples 1-7 are in the same order on each gel and results from 2 or more gels can be used to assist in identification
- Sample 8 is a “negative control” (no tissue was added) and is there to check for DNA contamination in the reagents

# Shark Group Specific PCR

Caballero et al 2012

- 4 categories
  - *Isurus* spp. - 253bp
  - *Carcharhinus* spp.
    - Also *Prionace glauca*
  - *Alopias* spp. - 119bp
  - *Sphyrna* spp. - 334bp



# Caballero et al 2012 (Shark Group ID Primers)



Sample Identification

- 1 \_\_\_\_\_
- 2 \_\_\_\_\_
- 3 \_\_\_\_\_
- 4 \_\_\_\_\_
- 5 \_\_\_\_\_
- 6 \_\_\_\_\_
- 7 \_\_\_\_\_

# Hammerhead Species ID PCR

Control fragment ~860bp\*\*\*

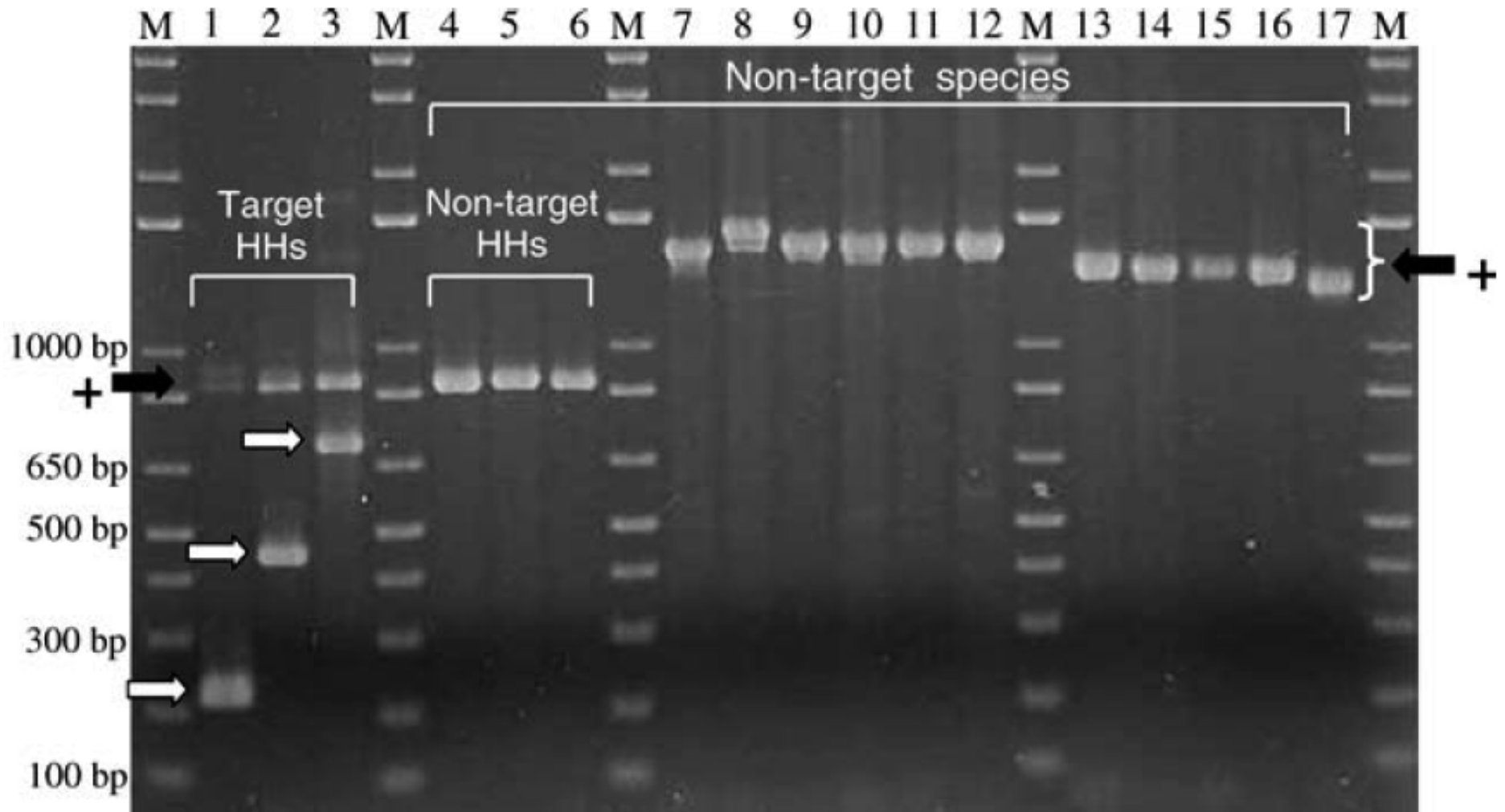
Abercrombie et al. 2005

*S. zygaena* - 249bp

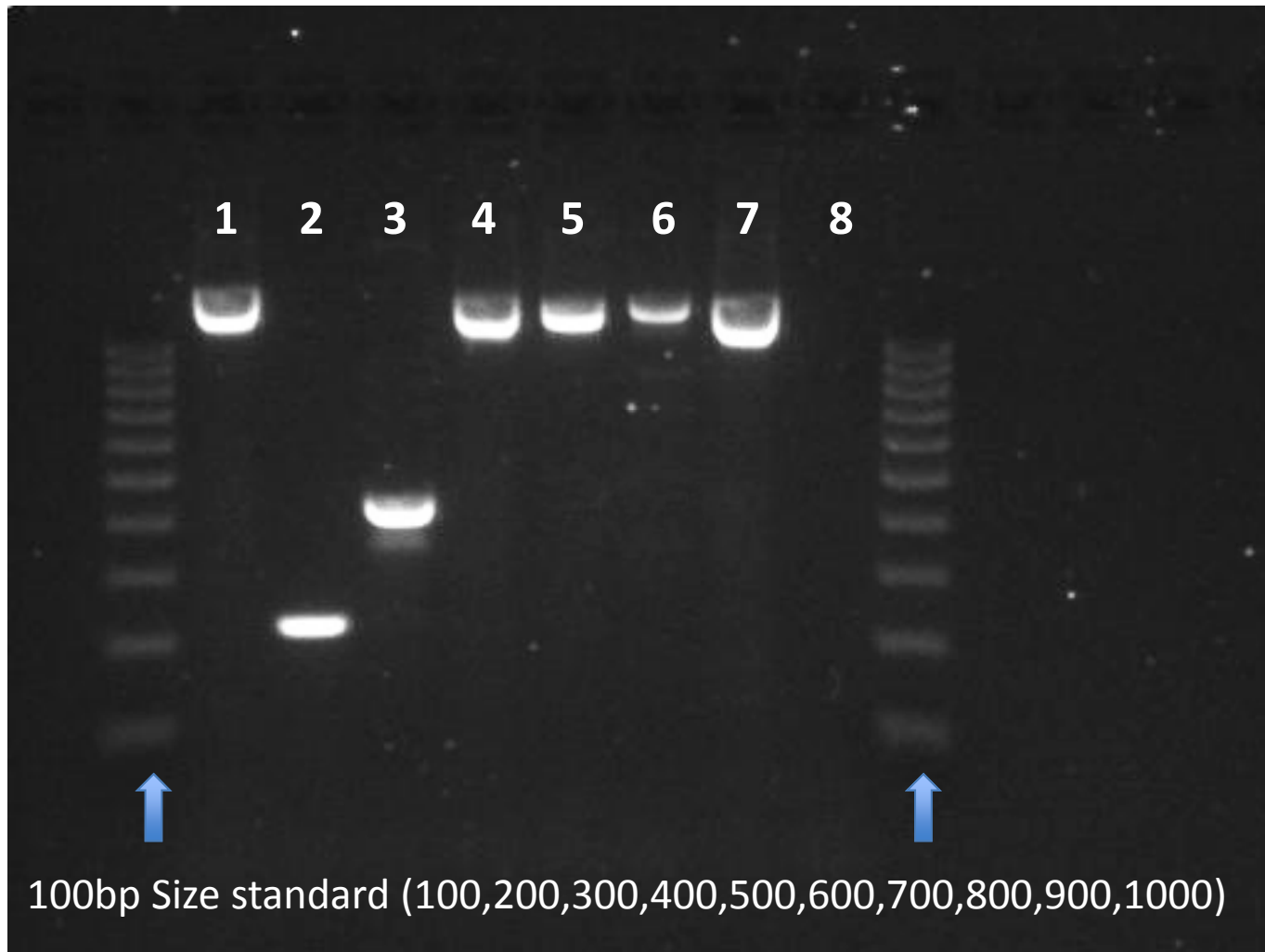
*S. lewini* - 445bp

*S. mokarran* - 782bp

\*\*\*note - control fragment in non *Sphyrna* species is ~1100-1200bp



# Abercrombie et al. 2005 (HammerHead Shark ID Primers)



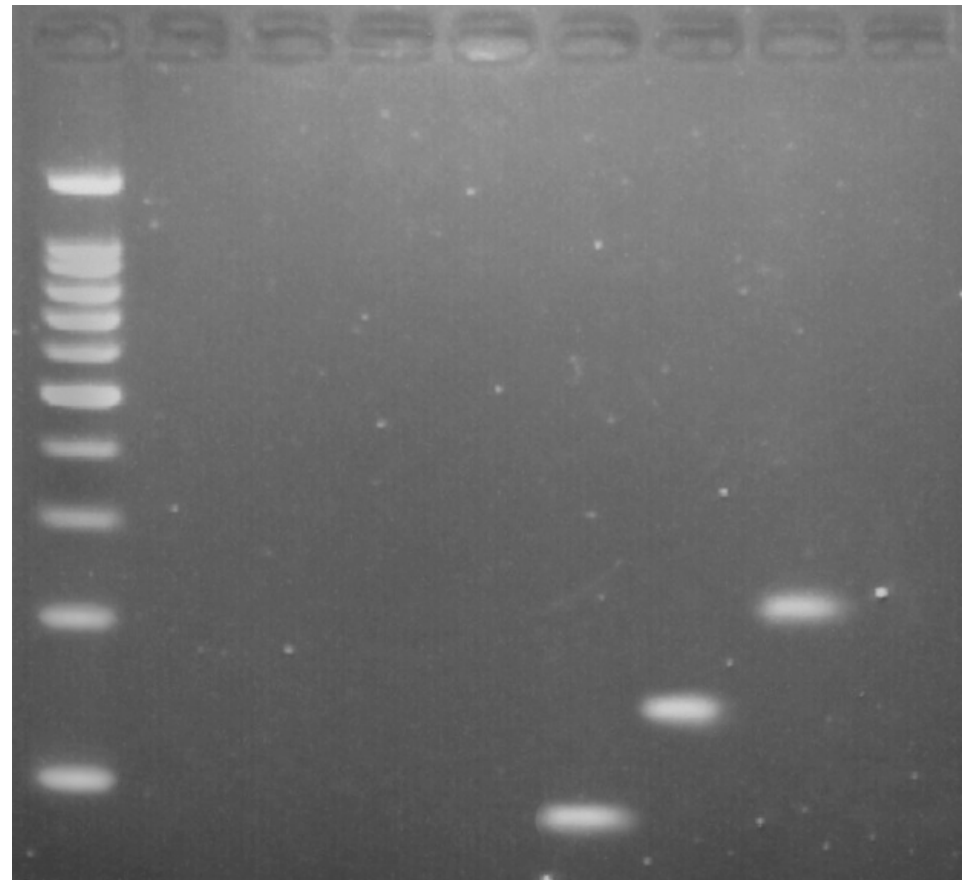
Sample Identification

- 1 \_\_\_\_\_
- 2 \_\_\_\_\_
- 3 \_\_\_\_\_
- 4 \_\_\_\_\_
- 5 \_\_\_\_\_
- 6 \_\_\_\_\_
- 7 \_\_\_\_\_

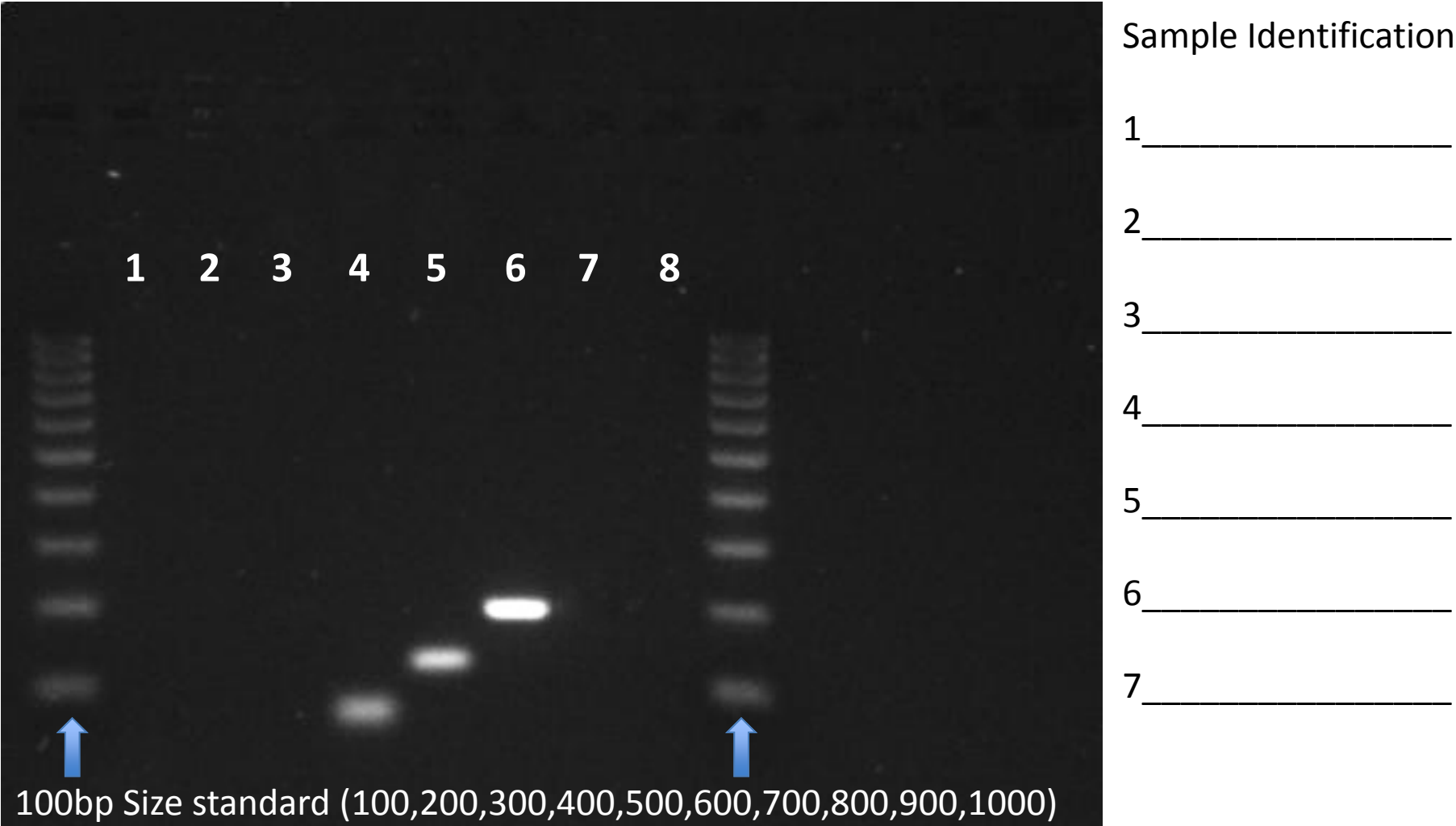
# Thresher Shark Specific PCR

Caballero et al 2012

- *Alopias* species
  - *Alopias vulpinus* - 76bp
  - *A. pelagicus* - 198bp
  - *A. superciliosus* - 264bp
- No amplification in non target species



Caballero et al 2012 (Thresher Shark ID Primers)



# Multiple Shark Species ID PCR

Shivji et al 2002

Control fragment ~ 1100-1200bp \*\*\*

*Isurus paucus* - 418bp

*Carcharhinus obscurus* - 480bp+++

*Lamna nasus* - 554bp

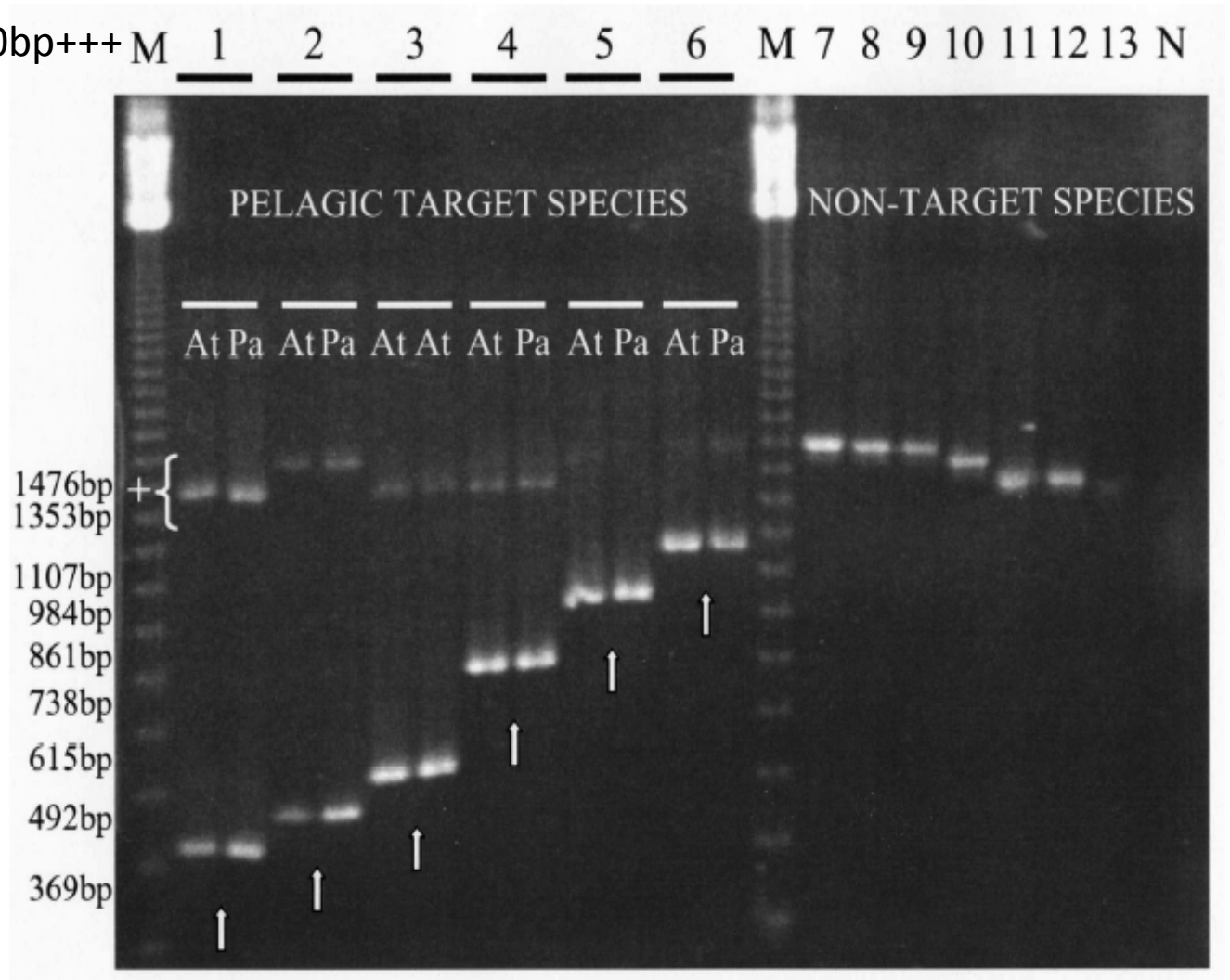
*Isurus oxyrinchus* - 771bp

*Prionace glauca* - 929bp

*C. falciformis* - 1085bp

\*\*\*note - control fragment in *Sphyrna* spp. is ~ 860bp

+++note - *C. obscurus* primer also amplifies in *C. galapagensis* and *C. longimanus*





# Shivji et al. 2002 (Multiple Shark Species ID Primers)

