

Genotyping Tail Feather Discoloration in Indiana Eastern Wild Turkeys (Meleagris gallopavo silvestris)

Abstract

The feather coloration in birds is a product of melanin and carotene in the feathers. One of the major contributors to Avian melanin synthesis is the melanocortin-1 receptor gene (MC1R) (Van Grouw, 2013). The MC1R gene encodes for a protein receptor, which is primarily expressed in melanocytes of a developing feather (Mundy, 2005). Melanocytes produce melanin. If a mutation occurs in this gene, there may be a feather discoloration. The goal of this experiment is to determine if there are mutations in the *MC1R* gene of the turkeys that I harvested. (Meleagris gallopavo silvestris)



Fig. 1: First round of PCR. The BP lane contains bands of DNA of known lengths, in 500 bp increments. There are no bands under NC, which represents the negative control. A bright band can be seen at \approx 1000 bp under G (ground turkey), which represents the MC1R gene. The same band can be seen under W (wild), only more faint. This represents the *MC1R* gene, but at a lower concentration than in the ground.



Methodology

- Genomic DNA extraction using DNeasy Blood and Tissue Kit: Used to remove DNA from turkey tissues. Spectrophotometry: Used to quantify the amount of **DNA** extracted.
- Polymerase Chain Reaction to amplify the MC1R gene: Seeks out the MC1R gene from the DNA and replicates it.
- Gel electrophoresis: Used to visualize the presence and concentration of the gene.
- Ligation: Used to prepare PCR product to be inserted into a plasmid.
- Transformation into *E.coli* cells: **A plasmid containing** PCR product is inserted into E.coli DNA
- Genotyping of sequences by Purdue Genomics: Enables us to view each individual nucleotide in the MC1R gene.
- Analysis/comparison of sequences ID'ing mutations



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Results

1000 BP



Fig. 2: Second round of PCR using increased Wild template DNA. The BP lane contains bands of DNA of known lengths, in 500 bp increments. There are no bands under NC, which represents the negative control. A bright band can be seen at \approx 1000 bp's under G (ground turkey), which represents the *MC1R* gene. The same band can be seen under W (wild) with the same brightness. This represents the *MC1R* gene at the same concentration.



Fig. 3: Shown are 3 IPTG+AMP plates used to grow *E.coli* after transformation. As seen on the Wild plate, there is a lawn of growth instead of individual colonies. The Ground plate has no growth. The +Control plate shows a lawn of growth.

After successful transformation, the PCR product can be sequenced, allowing for comparison of MC1R sequences, identification of mutations.

References

Mundy, N.I. (2005). A window on the genetics of evolution: MC1R and feather colourationin birds. Proceedings Biology Sciences, 272(1573), 1633-1640. https://doi.org/10.1098/rspb.2005.3107.

Van Grouw, H. (2013). What colour is that bird? The causes and recognition of common colour aberrations in birds. *British Birds*, 106, 17–29.



Discussion

The beginning steps of this experiment were a success. DNA was successfully extracted from store bought and wild turkey breasts. As shown in Fig. 1, bands at ≈1000 bp are seen for both ground and wild turkey. The MC1R gene is roughly the same length, showing the MC1R gene was successfully amplified. A second PCR reaction was completed with a higher concentration of wild DNA, meaning there would be more PCR product, which can be seen with the brighter band in Fig. 2. The transformation step was unsuccessful, as seen in Fig. 3, Wild and pGAP (control) both have a lawn of *E. coli* growth, while the ground showed no growth. All plates should have individual colonies growing. This shows unsuccessful transformation of the pJET1.2 + MC1R plasmid into the E.coli.

Next Steps

