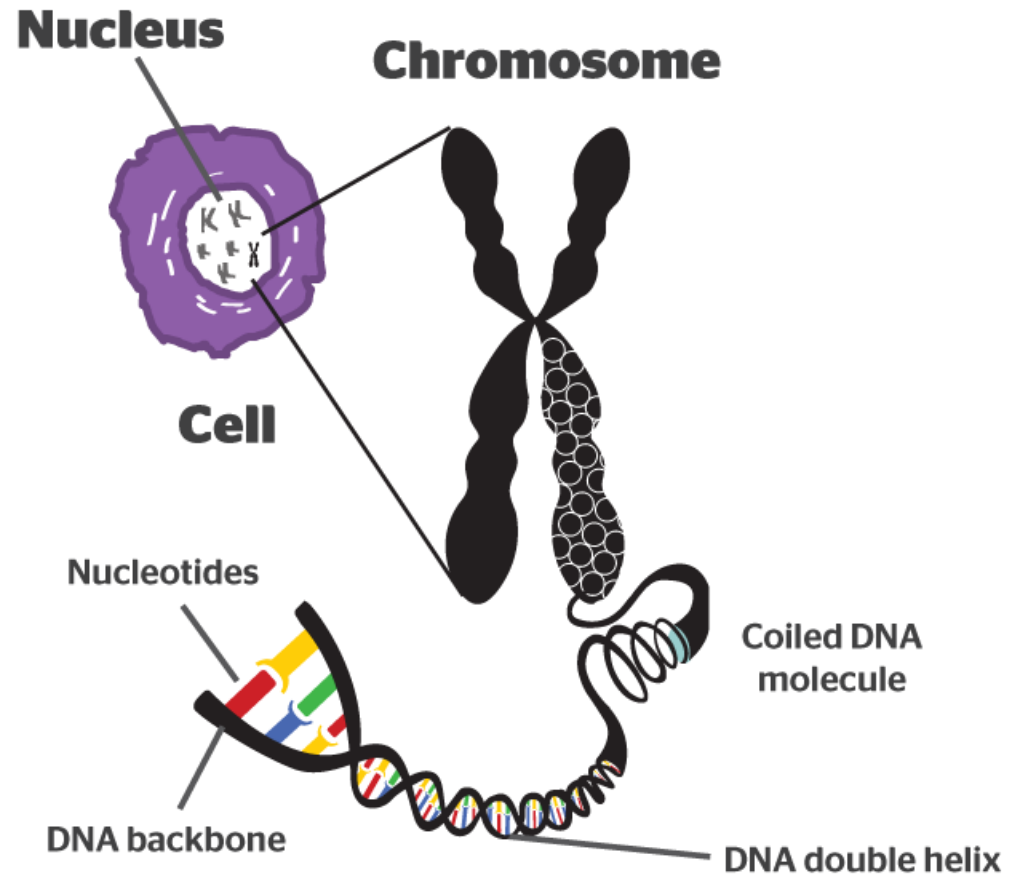


# TWO GENETIC DISEASES IN TOY MANCHESTER TERRIERS

**Nicole Tate, BS, Graduate Student**

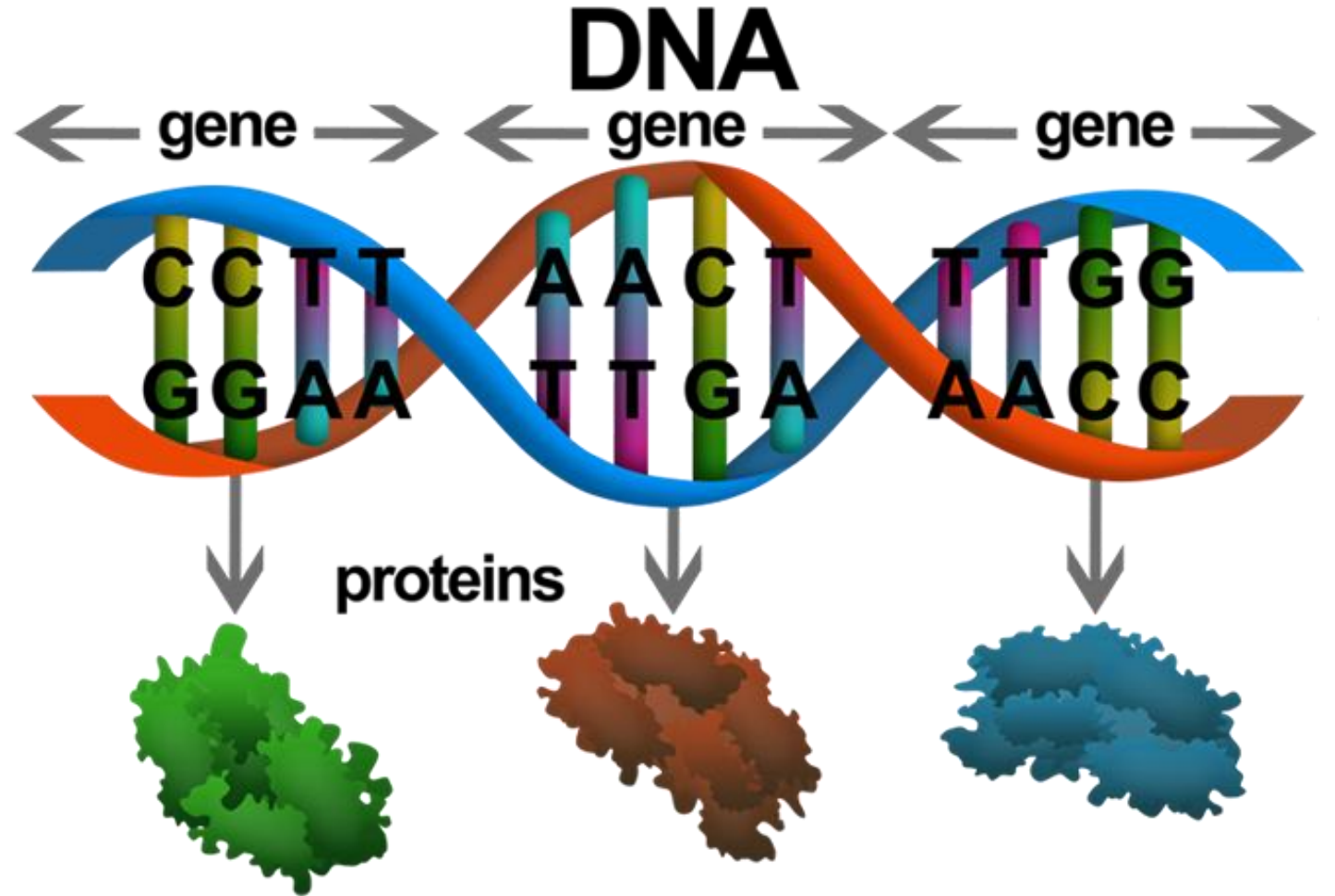
# Genetics Review

- Cell
- Genome
- Chromosomes
- DNA

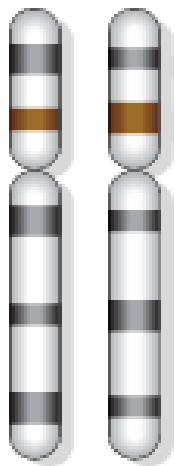
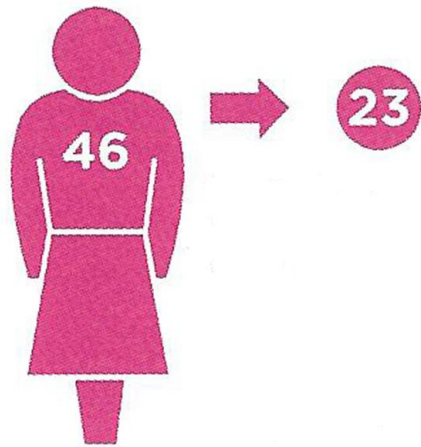


# Genetics Review

- DNA
- Nucleotide
- Gene
- Proteins



# Homozygous, Heterozygous, What?!

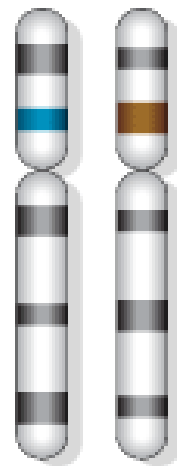


Individual B:  
homozygous

+

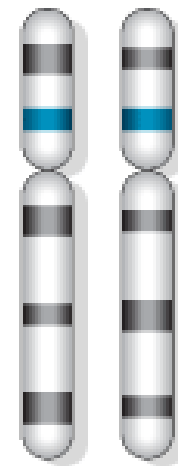


46



Individual A:  
heterozygous

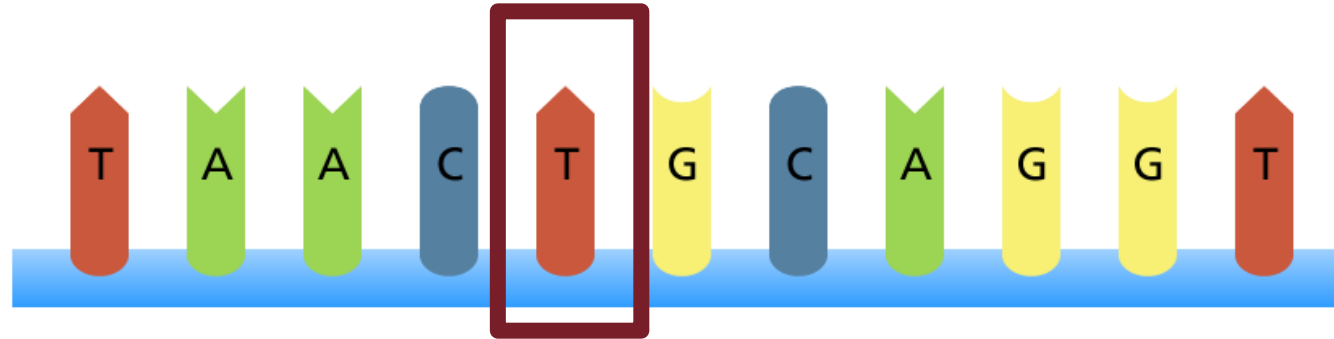
23 ♂



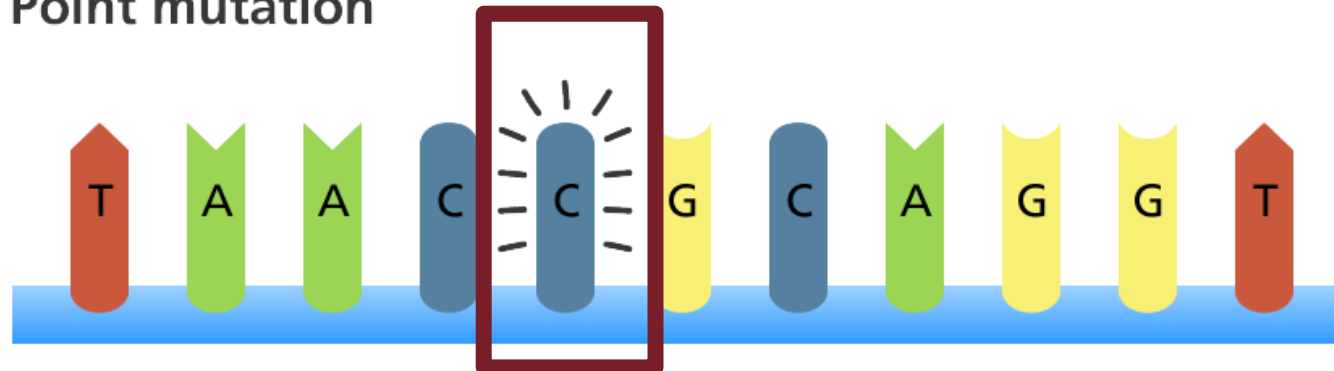
Individual C:  
homozygous

# Mutations

Original sequence



Point mutation

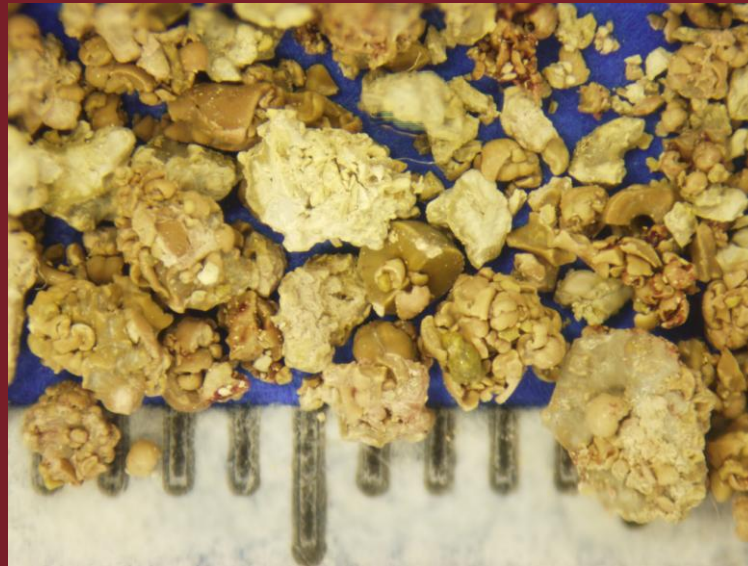


# Genetics Review

---

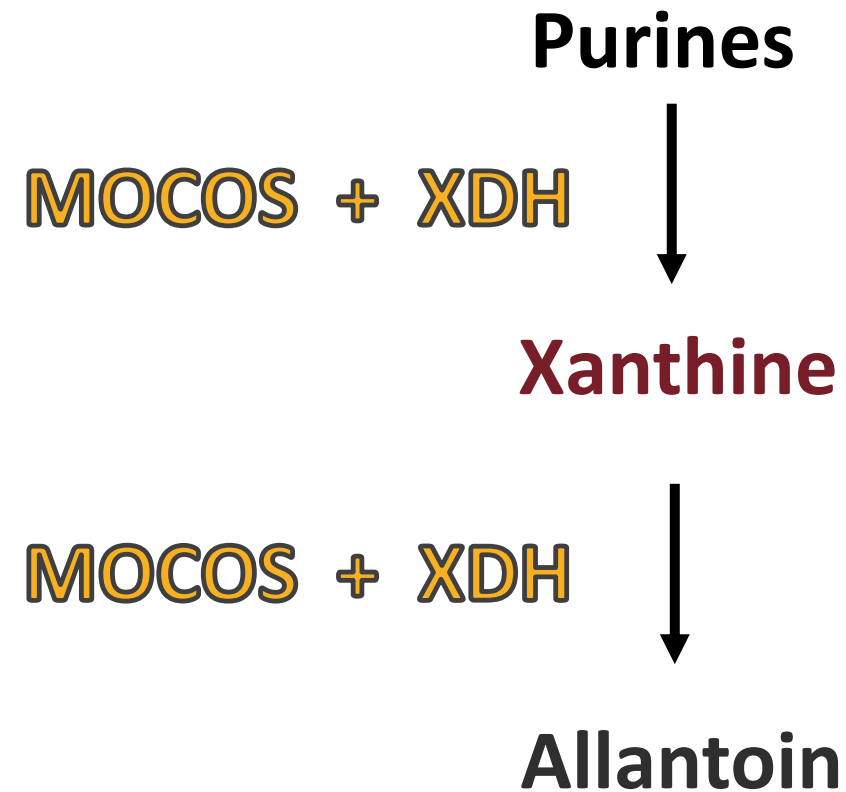


# XANTHINURIA: INTRODUCTION



# Where does xanthine come from?

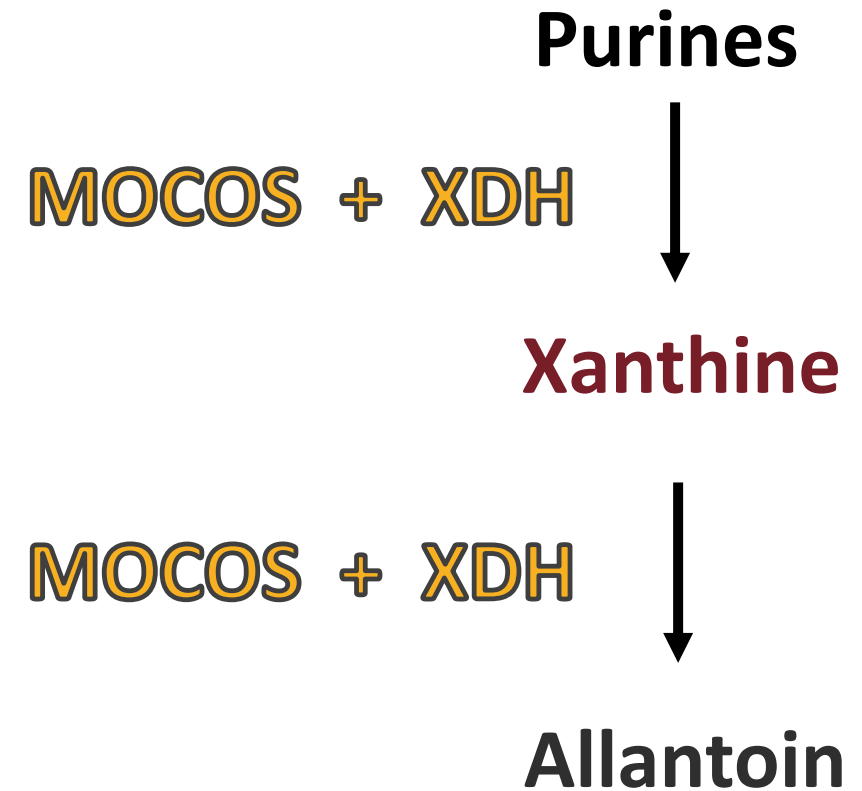
- Purine metabolism
  - ▣ Xanthine formation
- 2 enzymes
  - ▣ XDH
  - ▣ MOCOS
- Loss of function of enzymes
  - ▣ Increased urine levels





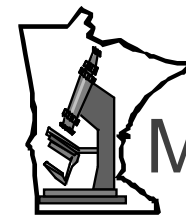
# Xanthinuria

- Two subtypes of hereditary xanthinuria
  - ▣ Type I - *XDH*
  - ▣ Type II - *MOCOS*



# Xanthine Urolithiasis

- Xanthine relatively insoluble
  - ▣ Uroliths (stones)
- Painful & obstructive
- Kidney disease



Minnesota Urolith Center

# Recommendations

- No treatment!
- Increased water intake
- Low purine diet
  - ▣ Prescription



# STUDY DESIGN

GENETIC INVESTIGATION OF CANINE XANTHINE UROLITHIASIS



# Study

---

- Aim:

- ▣ Uncover mutations underlying risk for canine xanthine urolithiasis via sequencing of *XDH* and *MOCOS* in genomic DNA from affected dogs.

# Affected Dogs and Test Populations

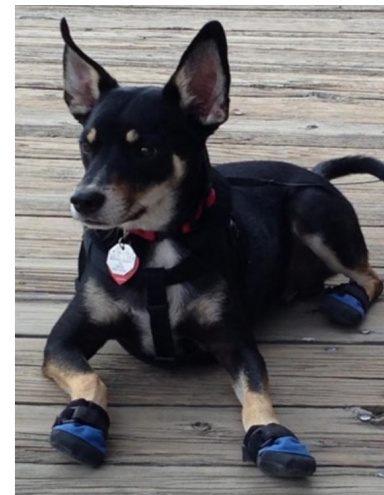
- Six dogs affected w/ xanthine urolithiasis
  - ▣ 2 Cavalier King Charles Spaniels (CKCS)
  - ▣ 2 Toy Manchester Terriers (TMT)
  - ▣ 1 mixed breed dog
  - ▣ 1 English Cocker Spaniel
- DNA extracted
- Sanger sequencing



Cavalier King Charles



Toy Manchester Terrier



Mixed Breed



English Cocker Spaniel

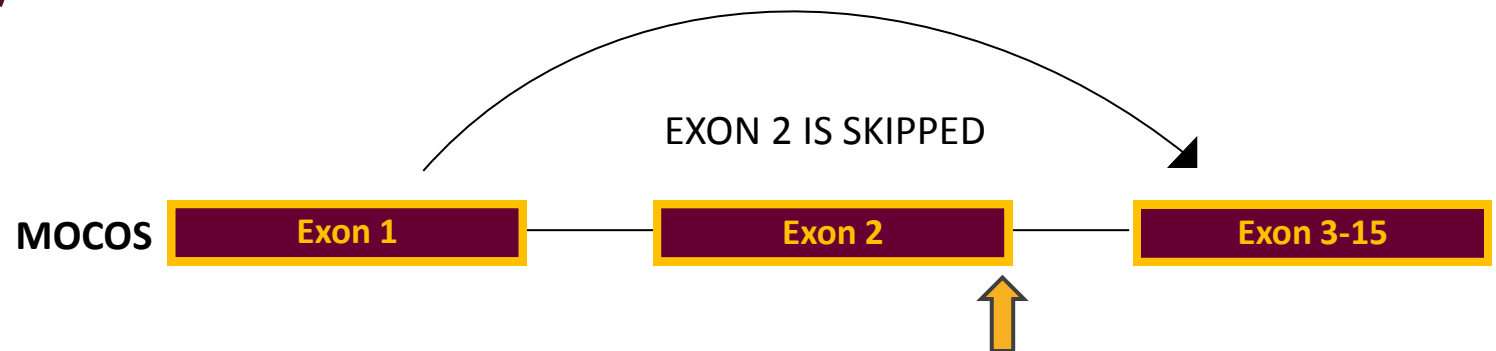
# TOY MANCHESTER TERRIERS

XANTHINURIA TYPE II<sub>a</sub>



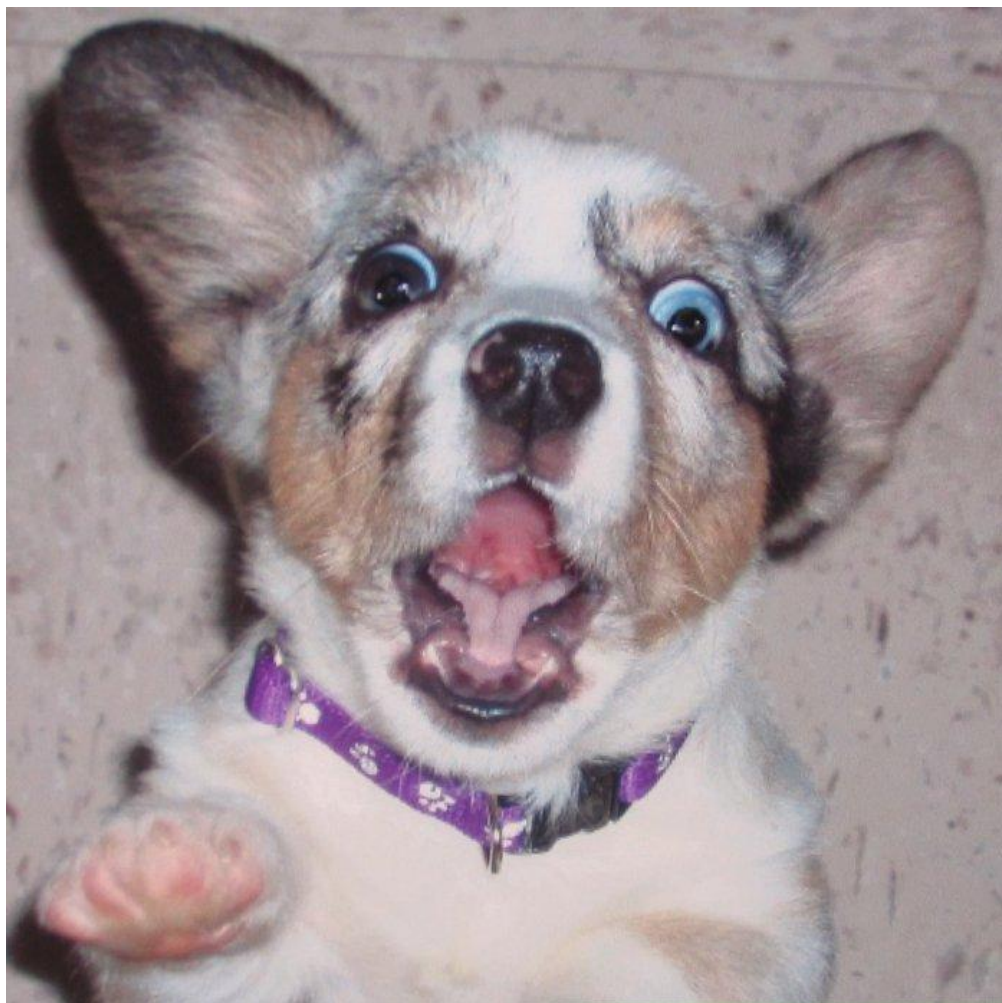
# Xanthinuria Type IIa

- No mutations in *XDH*
- Mutation in *MOCOS* (*last bp*)
  - ▣ Affected dogs homozygous
- Mutation predicted to affect splicing
  - ▣ Exon skipping (in frame)

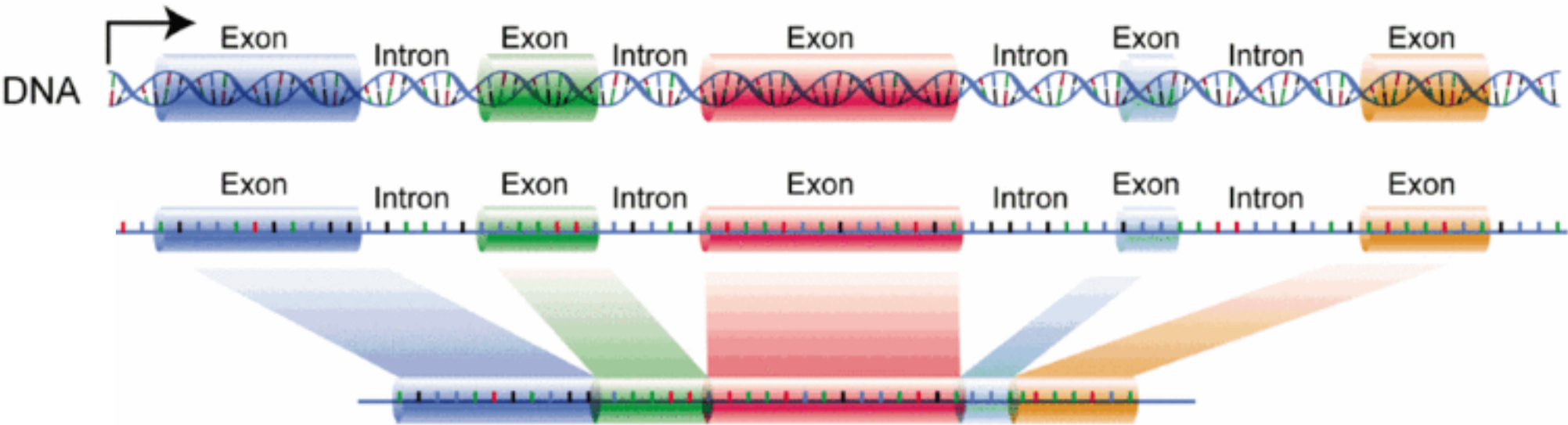




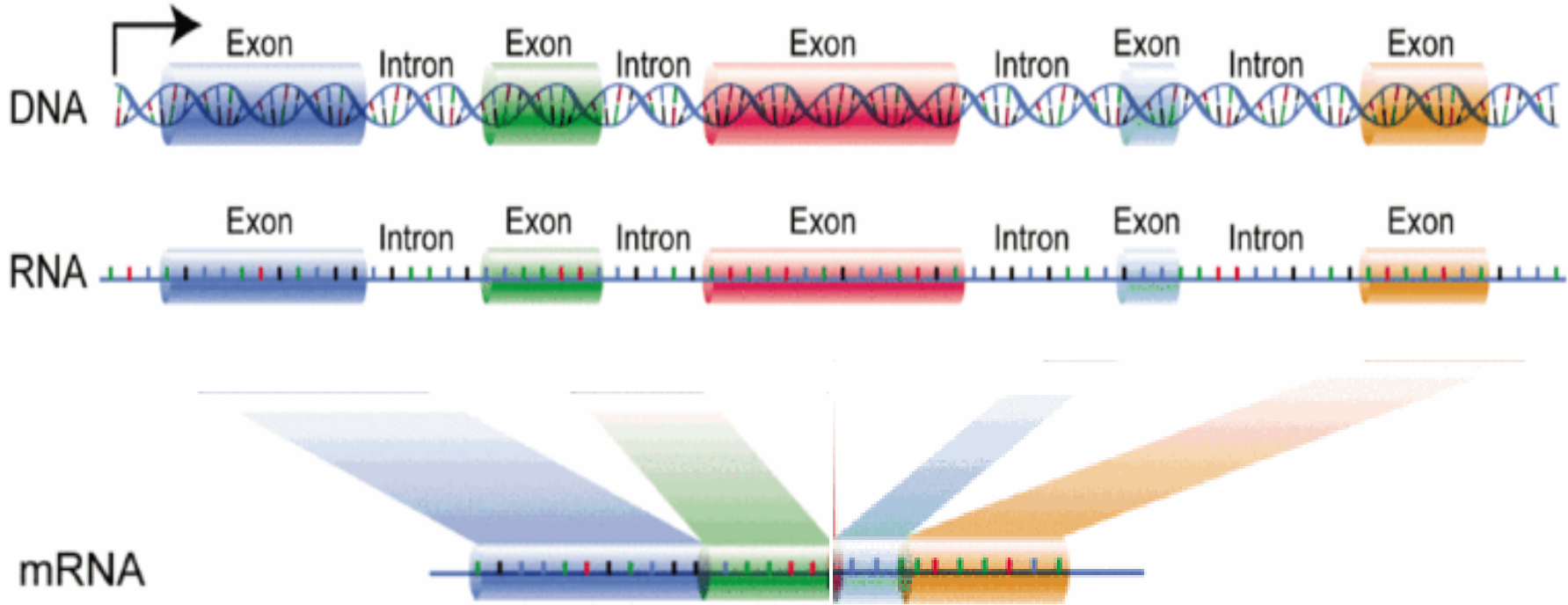
# Splicing?!



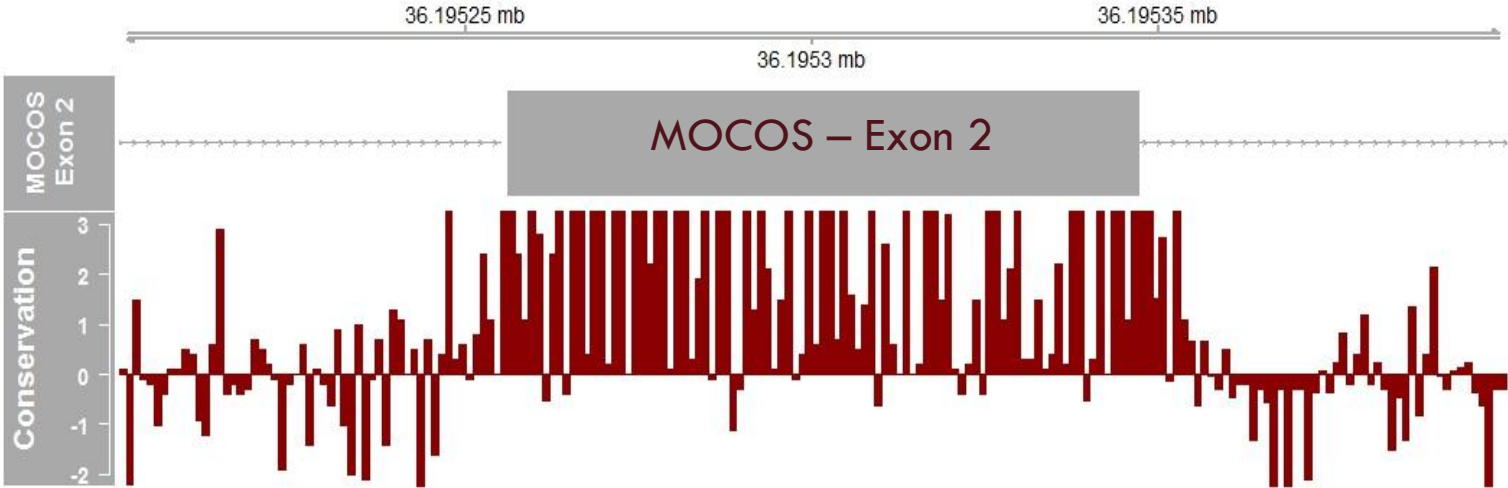
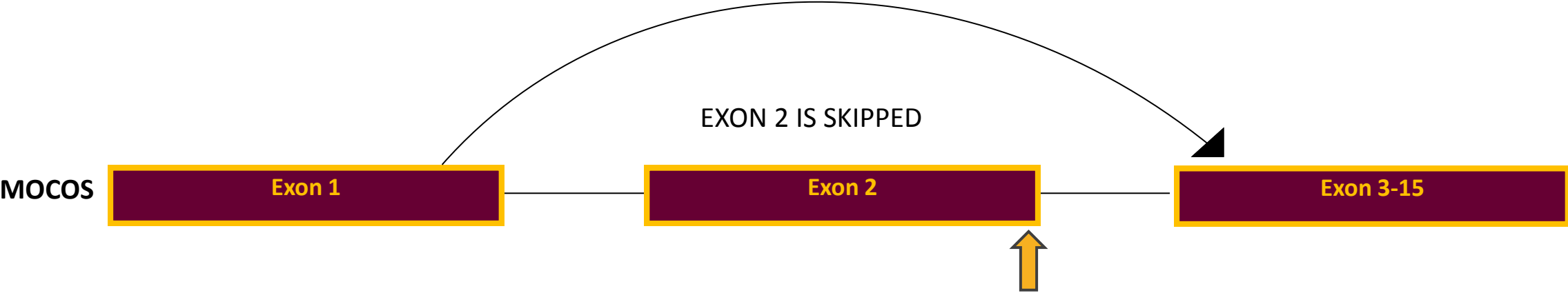
# Splicing?



# Splicing?

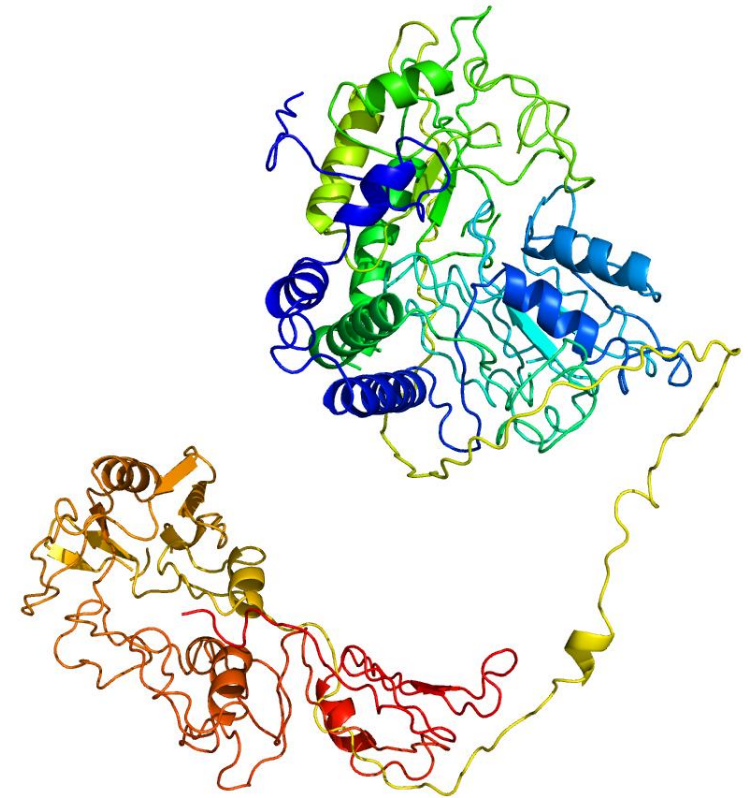
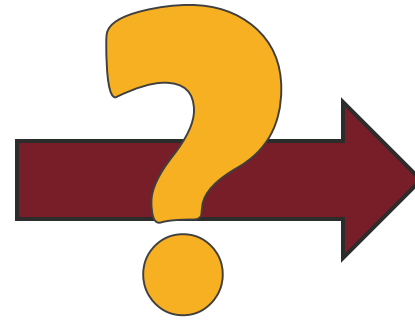
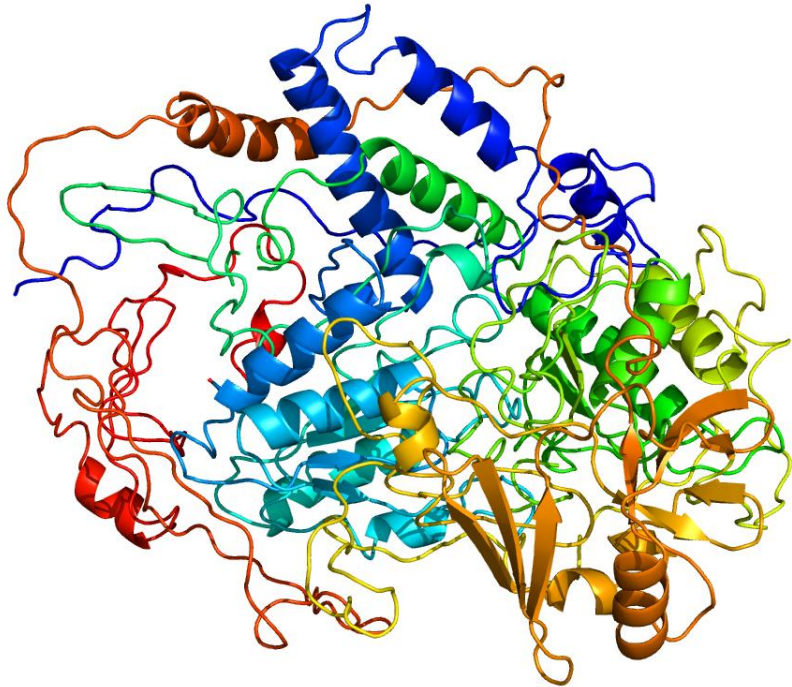


# Exon Skipped



# Aberrant Splicing

- ▣ MOCOS Protein Prediction (Phyre2)



# Xanthinuria Type IIa - Frequency

## □ Test Population

### ▣ 241 Toy Manchester Terriers

■ 194 clear of the mutation

■ 44 carriers = ~18%

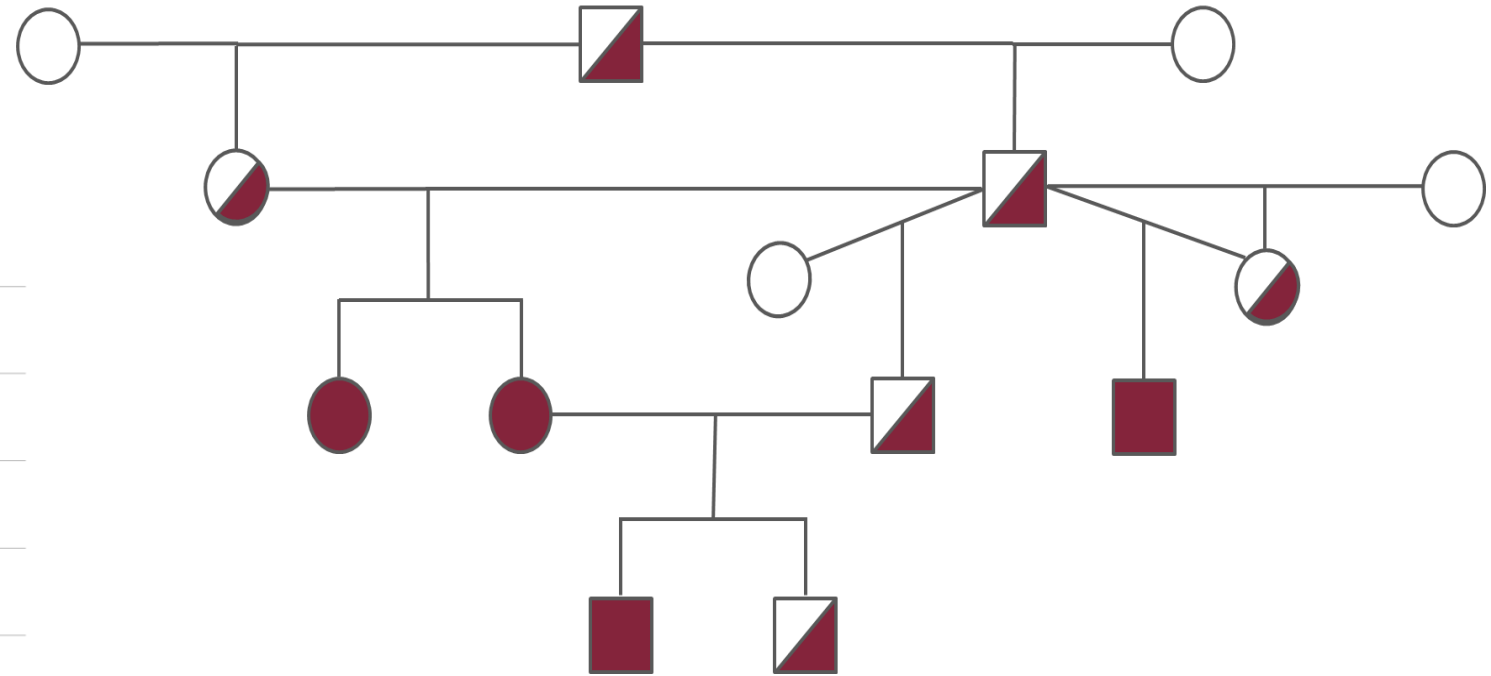
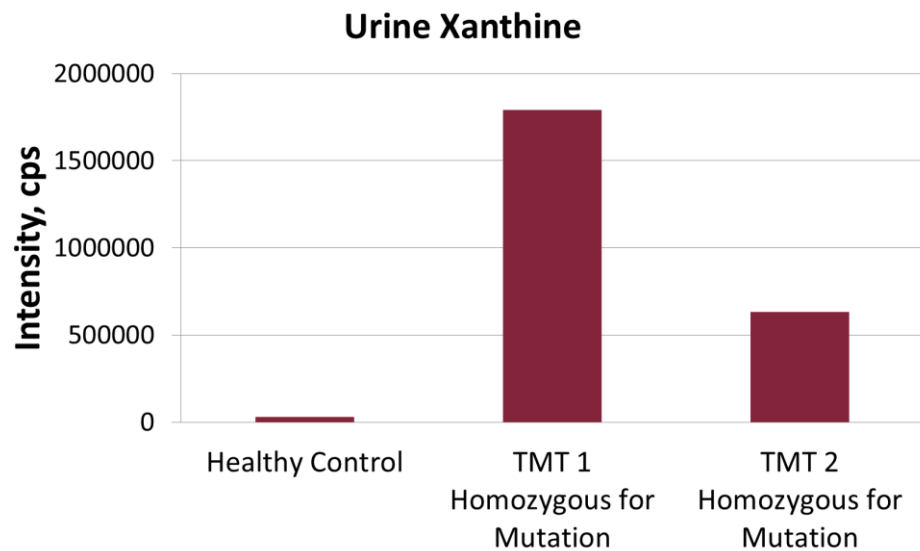
■ **6 homozygous**

■ **3 with no history of stones!**



# Three Homozygous “Controls”

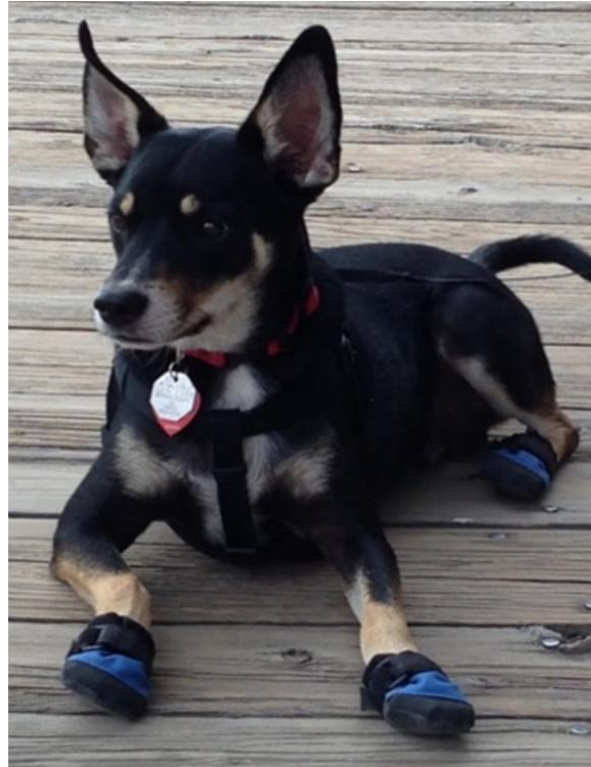
- 2 spayed females
- 1 male puppy
- Relatives of affected



# What about those other breeds?



Cavalier King Charles



Mixed Breed





# Questions??



# JUVENILE DILATED CARDIOMYOPATHY: INTRODUCTION



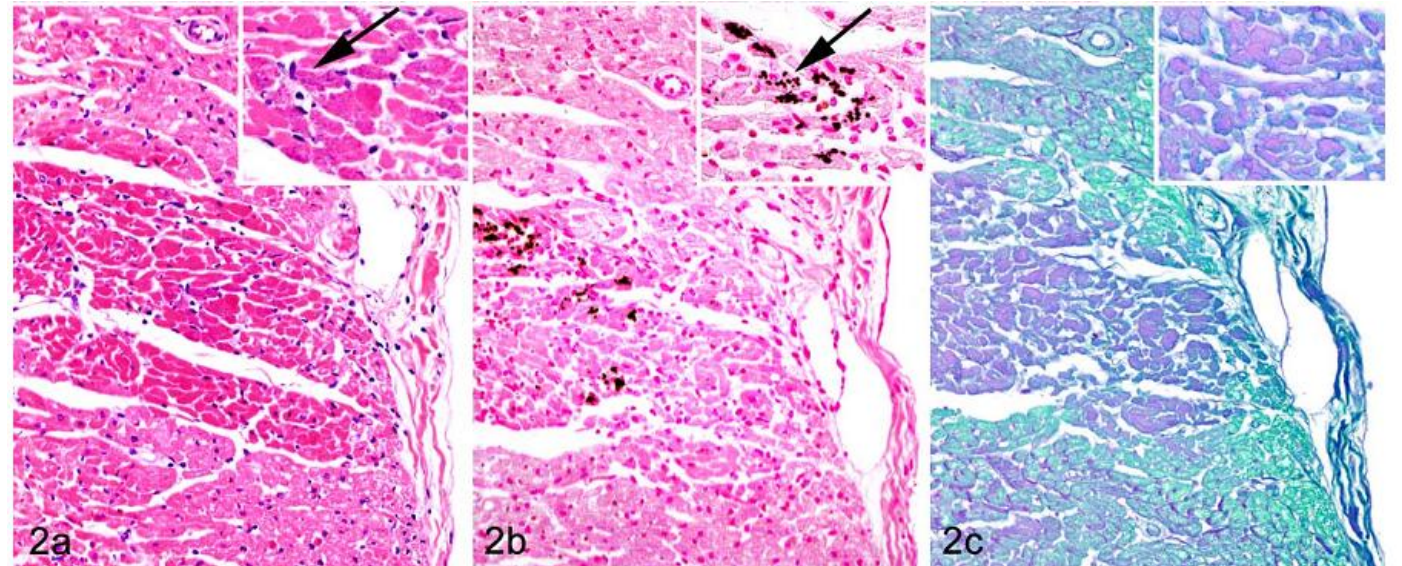
# JDCM in Toy Manchester Terriers

- Young age
  - ▣ 10 weeks to 1 year
- Appear healthy
  - ▣ Males cryptorchid
- Death sudden
  - ▣ Anesthesia/surgery
  - ▣ Exercise
- Presumed fatal arrhythmia



# Histology and Necropsy

- Mild enlargement of heart
- Degeneration and scarring of tissue
- Inflammation of tissue
- Mild
- Widespread

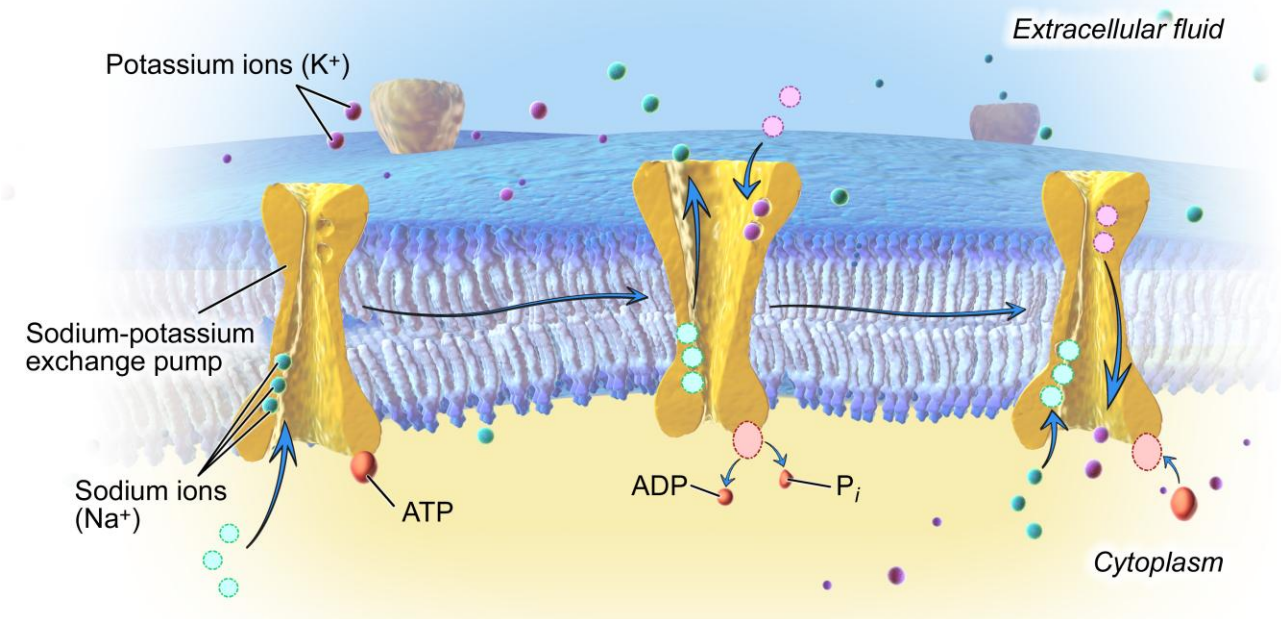


# Genome Wide Association Study

- Numerous cardiomyopathy genes
- Compare cases to controls and look for markers that are in a higher frequency in cases.
  - ▣ 170K “markers”
- 12 Cases
- 36 Controls

# Region of Interest

- Genes associated with cardiomyopathy
- Potassium Channels
  - ▣ Regulate the electrical activity of cardiac muscle cells
  - ▣ Dysfunction can cause arrhythmias



# JDCM in Toy Manchester Terriers - Frequency

## □ Test Population

### ▣ 191 Toy Manchester Terriers

- 133 clear of the mutation
- 43 carriers = ~23%
- **15 homozygous**



# TESTING & BREEDING DECISIONS





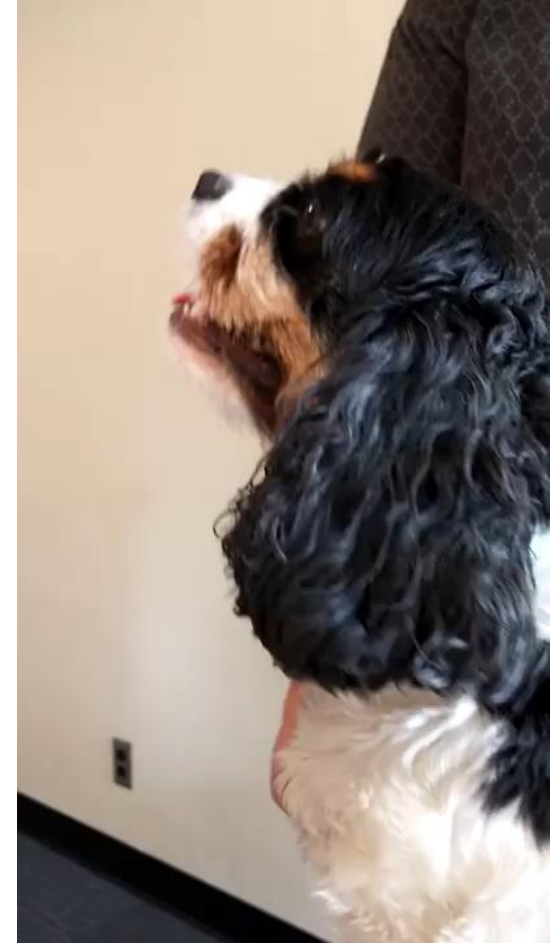
# Submitting Samples

- Types
  - ▣ Blood
  - ▣ Cheek Swab
  - ▣ Dew Claw & Tail Docking
  - ▣ Semen
  
- Information and submission forms can be found on our websites:  
<http://z.umn.edu/jdcm>
- <http://z.umn.edu/xanthinuria>



# Submitting Cheek Swabs

- Place completed swabs directly into a paper envelope and seal it.
- Submission form and envelope with swabs go in a larger envelope.
- Complete instructions on our websites



# Test Results

## University of Minnesota Juvenile Dilated Cardiomyopathy (JDCM)

*Canine Genetics Laboratory*

612-624-5322

295 Animal Science/Veterinary Medicine

Fax: 612-625-0204

1988 Fitch Avenue

e-mail: [cgl@umn.edu](mailto:cgl@umn.edu)

St. Paul, MN 55108

<http://z.umn.edu/caninegenetics>

**Sample Number:** TMT 000

**Owner:** TMT Fan  
Address

**Report Generated:** Sep 16, 2016

**Registered Name:** Cutest TMT Ever

**Breed:** Toy Manchester Terrie

**With Registration Number:**

**Sex:** F      **DOB:** 09/16/16

**Permanent ID:**

**ID verified:**

**Juvenile Dilated Cardiomyopathy (JDCM):** Affected (D/D) - Two copies of the JDCM mutation

Print Form

### **Orthopedic Health Foundation (OFA) Database:**

To register these results with OFA, make a copy of this result page, sign below, enclose fee (if any) and send to:

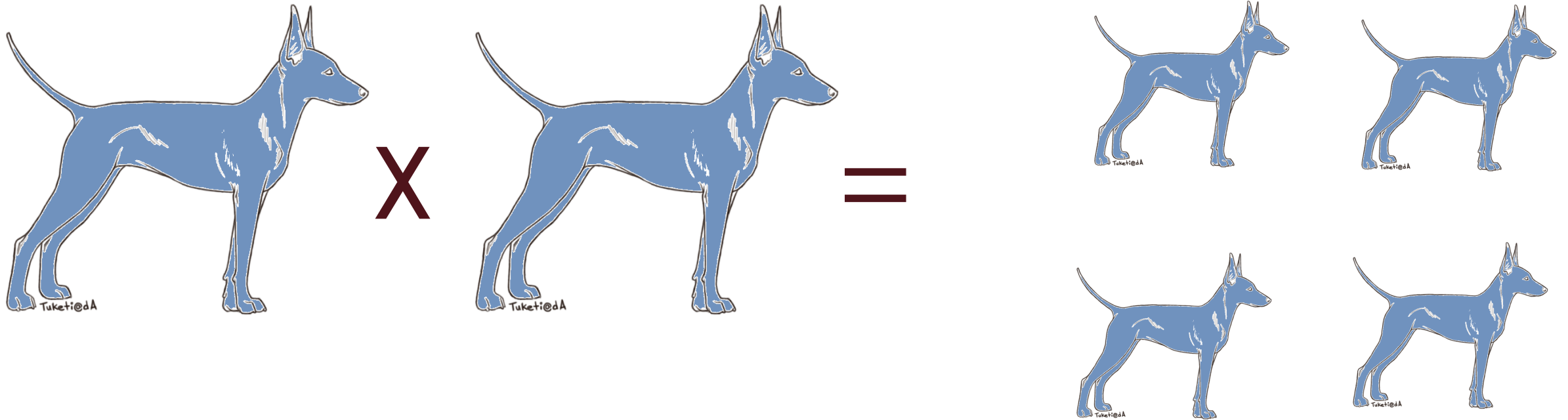
# Test Results

---

- Clear (N/N), Carrier (D/N), Affected (D/D)
- Fully penetrant
- Recessive
  
- So now what?

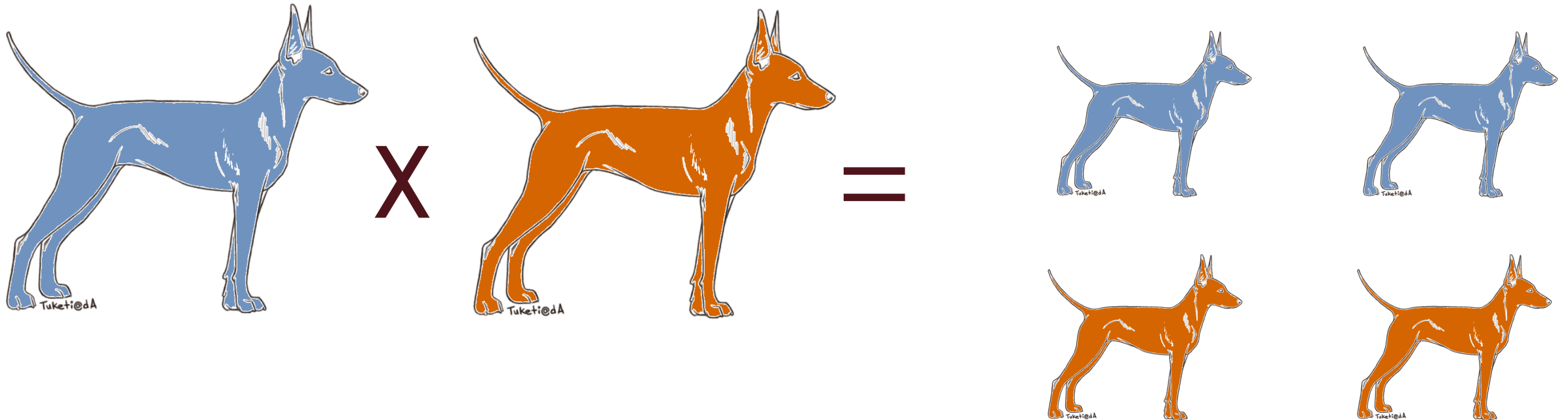
# Breeding Outcomes

- Clear (N/N) x Clear (N/N) = 100% Clear (N/N)



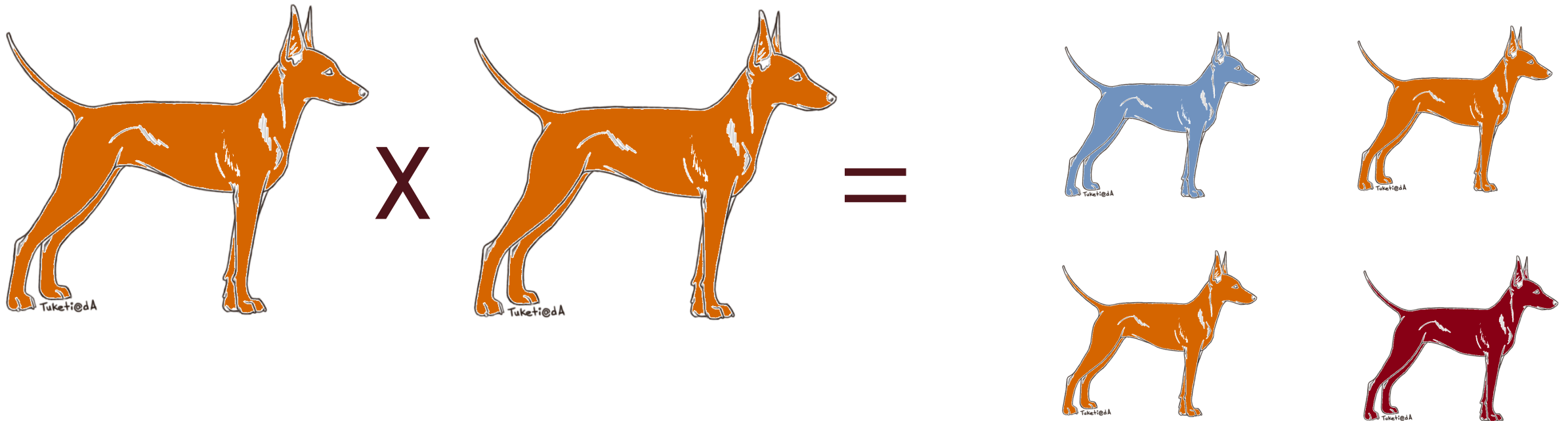
# Breeding Outcomes

- Clear (N/N) x Carrier (D/N) = 50% Clear (N/N), 50% Carrier (D/N)

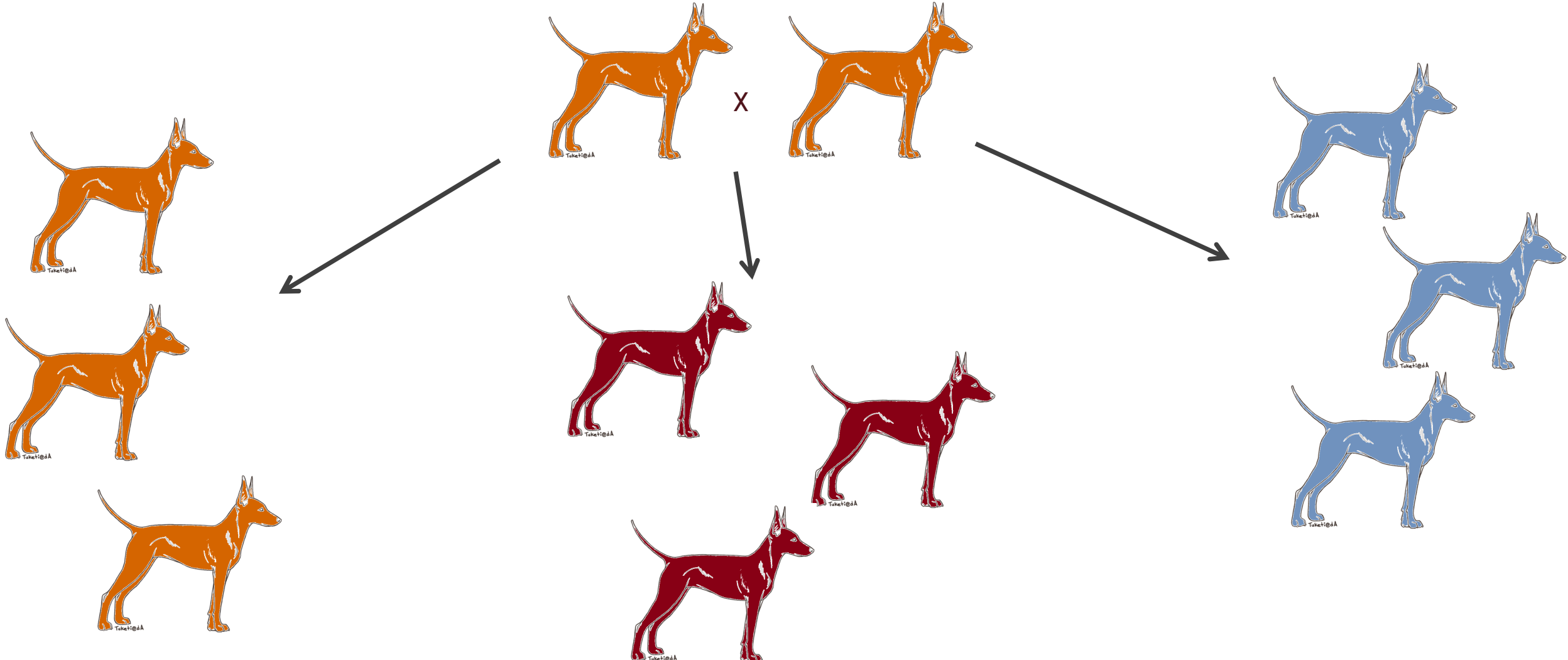


# Breeding Outcomes

- Carrier (D/N) x Carrier (D/N) = 25% Clear (N/N), 50% Carrier (D/N), 25% Affected (D/D)



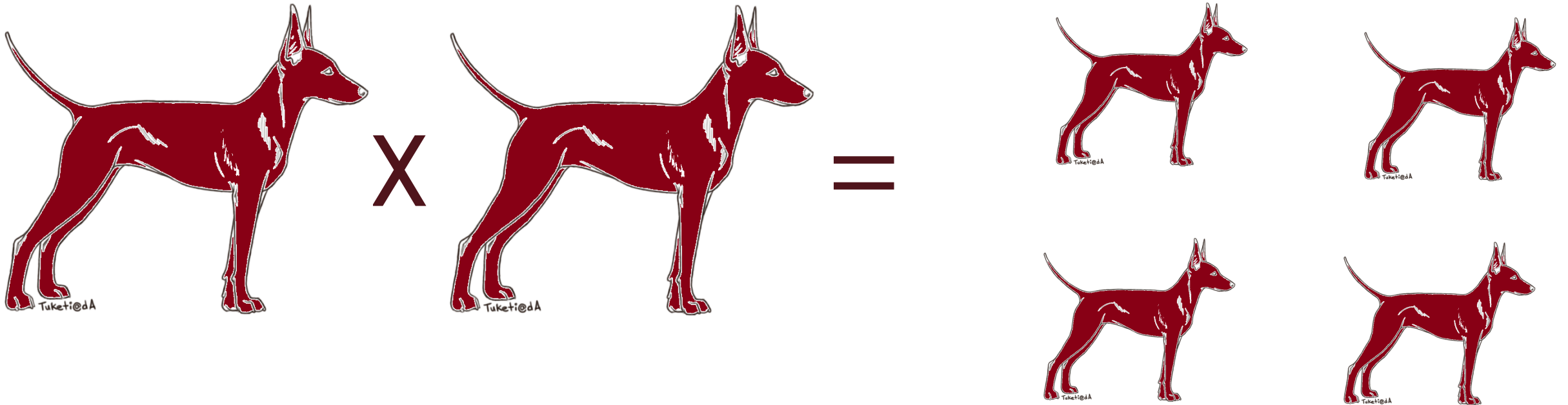
# In Reality...





# Breeding Outcomes

- Affected (D/D) x Affected (D/D) = 100% Affected (D/D)



# Take Home Message

- 18% carriers for Xanthinuria
- 23% carriers for JDCM
- Can't just use clear dog
  - ▣ small number of breeding stock
- Breed conditions out over many generations
  - ▣ Test and replace
  - ▣ Keep carriers

# Acknowledgements

- Dr. Eva Furrow
- University of Minnesota Canine Genetics Laboratory
  - ▣ Katie Minor
  - ▣ Kasey Petersen
  - ▣ Shannon Larrabee
  - ▣ Dr. Jim Mickelson
  - ▣ Dr. Jody Lulich
- Veterinarians that submitted samples
  - ▣ Dr. JD Foster
  - ▣ Dr. Michael Koch
  - ▣ Dr. Kate Byrne
  - ▣ Dr. Tim Kuhnmuench
- Dr. Paula Henthorn
- Dr. Shannon Martinson
- Dog Biomedical Variant Database Consortium
- Study participants, owners, breeders and breed associations



# Questions?

