Sifting Comparative Sequence:
Lipid Metabolism Genes and Regulation

Outline:

1) Apolipoprotein AI containing Gene Cluster

- **-Comparative Analysis: Example 1**
- **-Gene Regulation and Speciation Changes?**
- **-Novel Gene Identification**

2) Apolipoprotein E containing Gene Cluster

- **-Comparative Analysis: Example 2**
- **-Gene Regulatory Elements? Strategic Approaches.**

3) 7-alpha cholesterol hydroxylase (CYP7A) Gene

- **-Comparative Analysis: Example 3**
- **-Enhancer Identification: Transgenic Refinement.**
- **-Gene Regulation and Speciation Changes?**

Background

Majority of human genomic sequence is available.

- **-33% Finished**
- **-61% Draft**

Mouse genomic sequence is quickly becoming available (3X draft).

- **-2% Finished**
- **-11%Draft**

Raw Sequence----> Biological Function

Raw Sequence----> Biological Function outside of transcribe regions

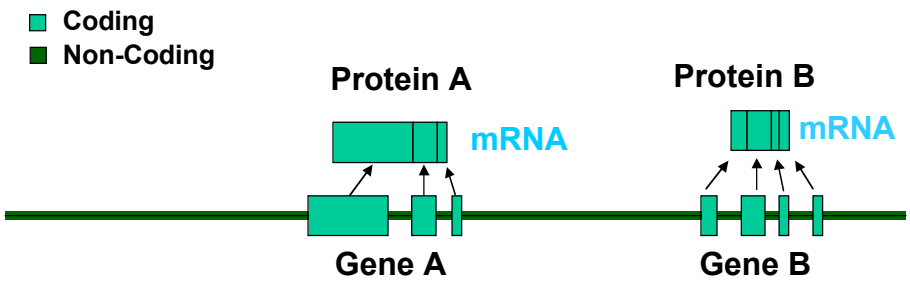
In general, functionally important sequences are conserved.

Conserved sequences are functionally important.

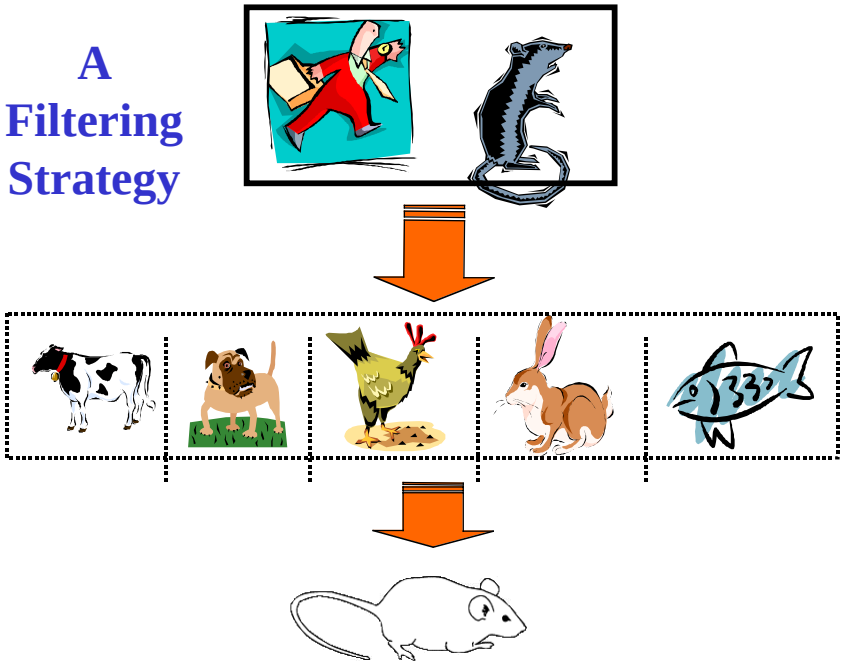
Categories of DNA

Coding ~5%

Non-coding ~95%



A Filtering Strategy



Welcome to the VISTA, or VISualization Tool for Alignments

VISTA is an integrated system for global alignment and visualization, designed for comparative genomic analysis.

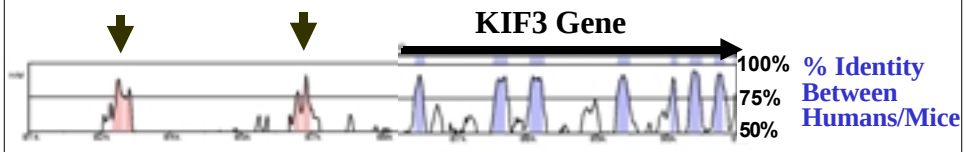
To Use VISTA:

WebServer	Input your sequences annotation in the required format described below, and we will send you a VISTA plot in PDF format.

VISTA

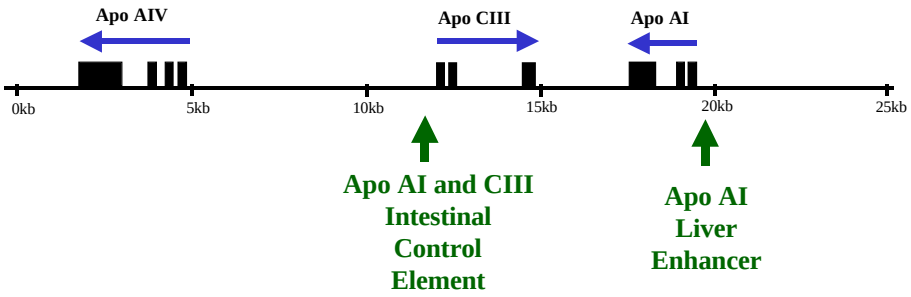
(VISual Tool for Alignment)

http://www-gsd.lbl.gov/vista



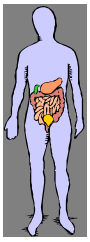
Human Sequence (horizontal axis)

Human Chromosome 11q23 Apolipoprotein Gene Cluster



Species Differences in Gene Expression

ApoAI Expression Sites:



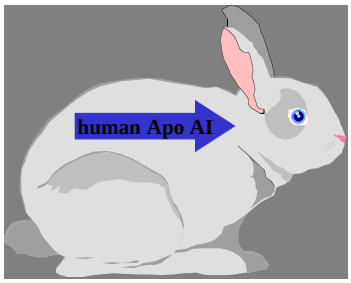
Intestine and Liver

Intestine and Liver

Intestine

Can Comparative Genomic Sequence Analysis Explain Differences in Gene Expression Between Species?

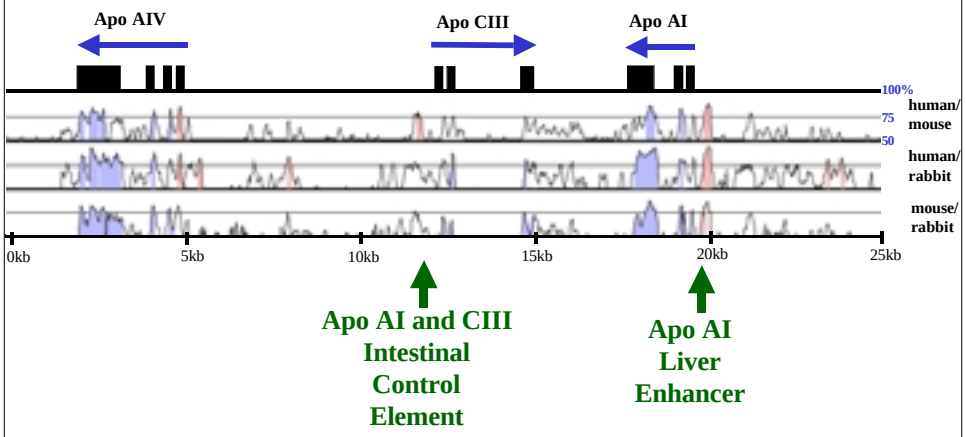
- All mammals express apo AI in liver and intestine except rabbits.
- Human apo AI transgenic rabbits express apo AI in liver and intestine.



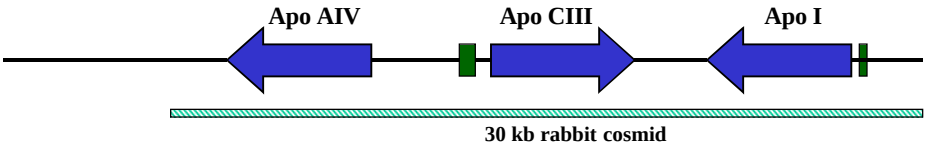
Hypothesis:
Cis regulatory difference(s) in rabbit Apo AI explain the lack of rabbit apo AI liver expression.

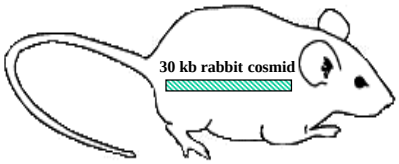
Comparative Genomic Sequence Analysis of Human/Mouse/Rabbit Apo AI

Do rabbits lack sequence conservation maintained in humans and mice?



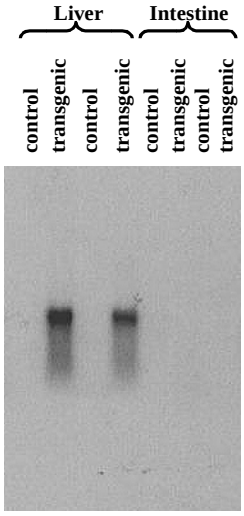
Rabbit ApoAI Transgenic Mice





Prediction:
Rabbit Apo AI should express in intestine and not liver.

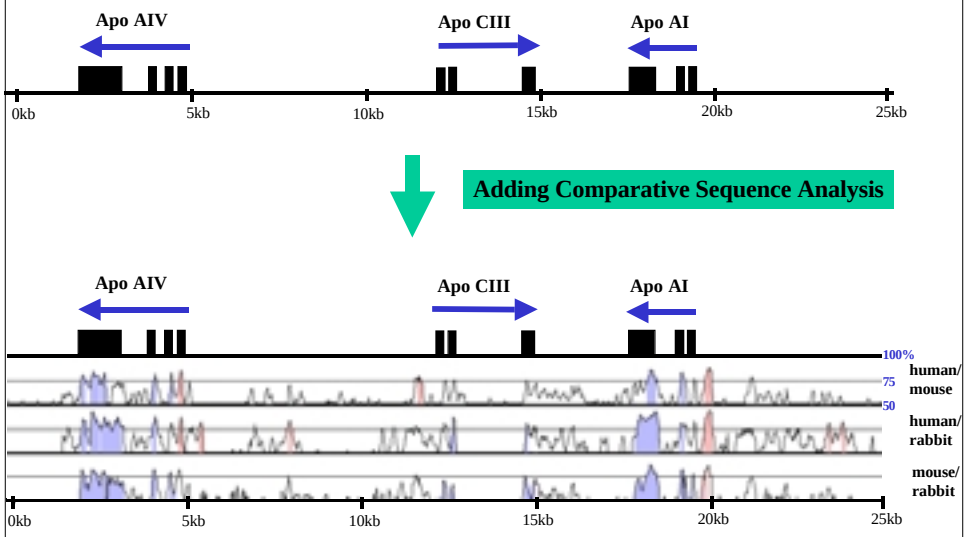
Northern analysis of rabbit apoAI in trangenic mice



Observation:
Rabbit Apo AI does express in transgenic mouse liver.

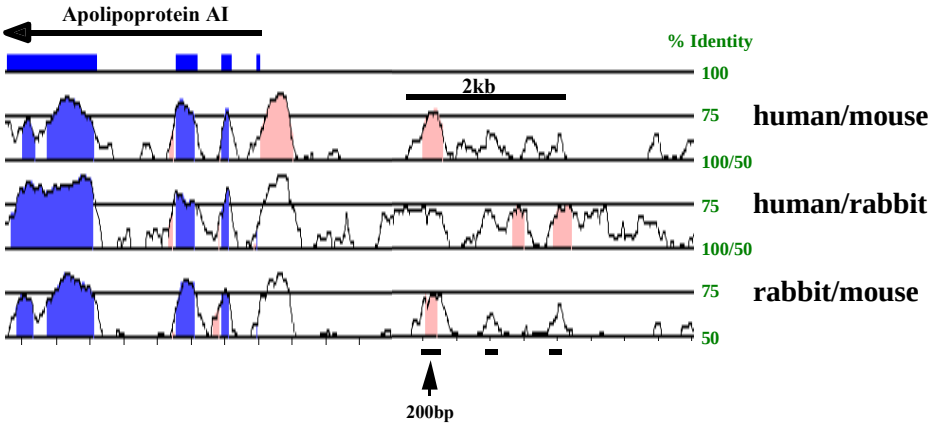
Conclusion:
Cis regulatory changes are unlikely to solely explain the lack of rabbit Apo AI liver expression.

Human Chromosome 11q23 Apolipoprotein Gene Cluster

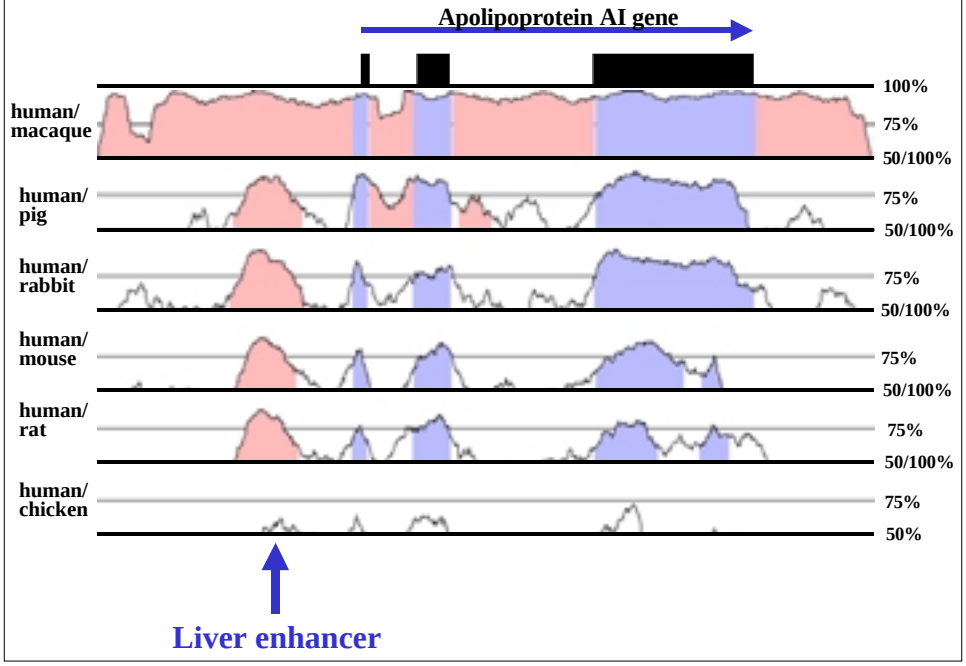


Three Species Sequence Comparison (Humans, Mice and Rabbits)

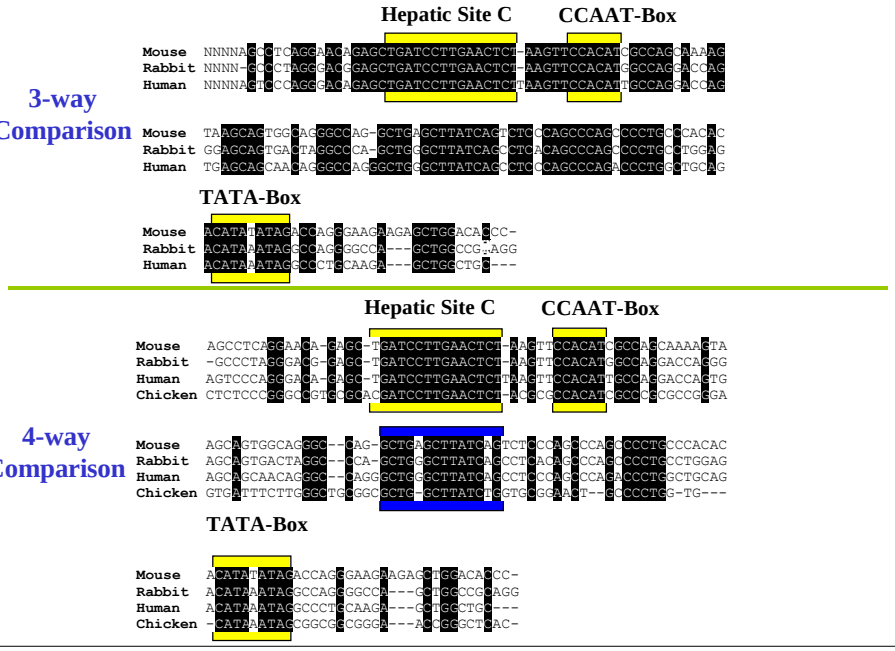
Aiding in the refinement of potential regulatory sequences



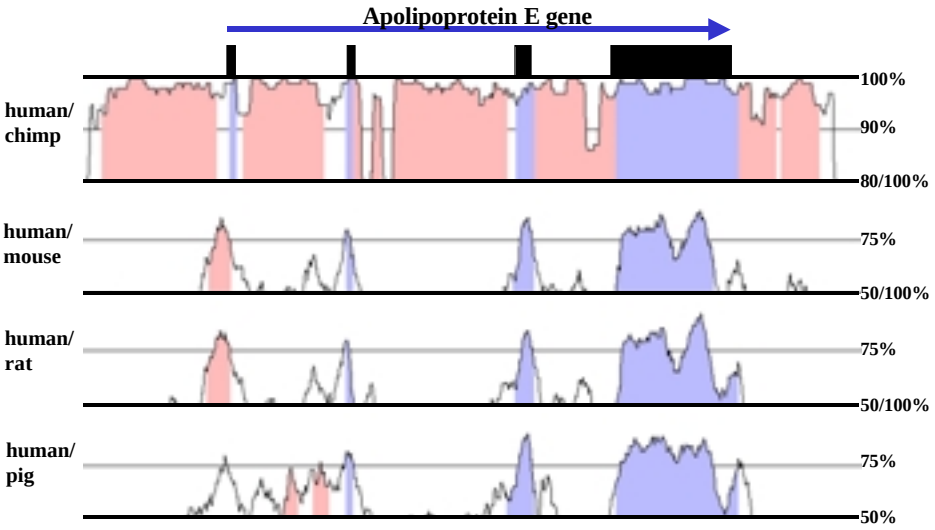
Multi-Species Comparative Analysis (VISTA)



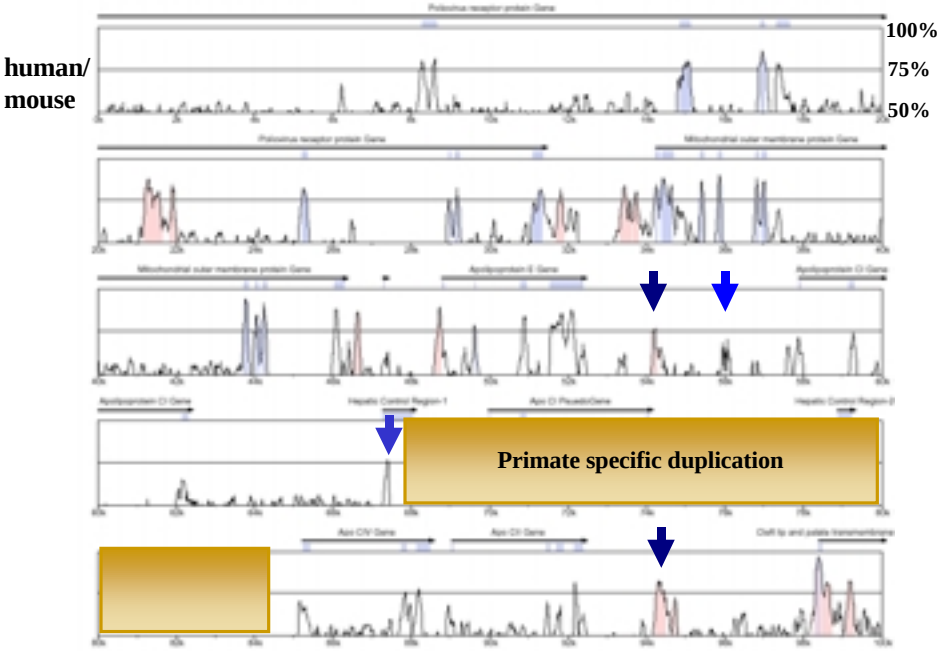
Power of Deep Sequence Alignments



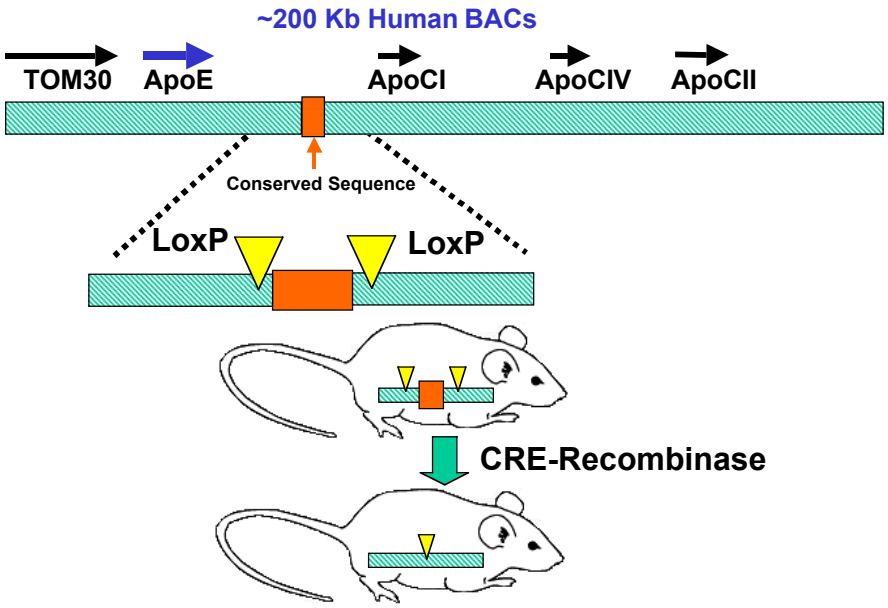
Multi-Species Comparative Analysis (VISTA)



Apolipoprotein E Conservation and Gene Regulation



Characterizing Conserved Sequence Function



Towards A Better VISTA

Information from a Single Sequence Alone

Multi-Organism High Quality Sequences