# Validity of morphological characters used to distinguish *Culex restuans* and *Culex pipiens*

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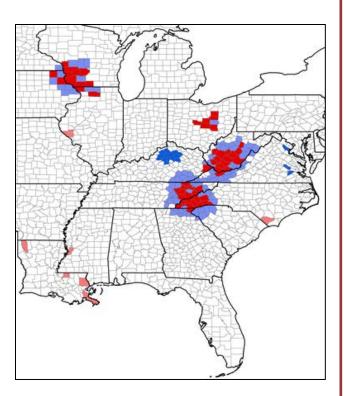
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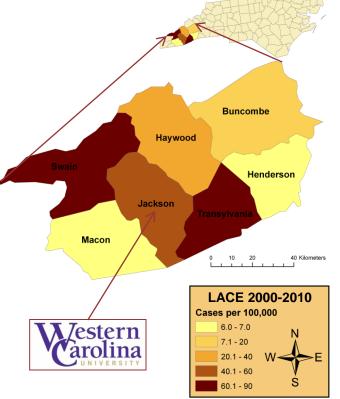




Haddow AD and Odoi A, The incidence risk, clustering, and clinical presentation of La Crosse virus infections in the eastern United States, 2003-2007. *PLoS One*. 2009 Jul 3;4(7):e6145.











La Crosse virus is endemic in Western North Carolina and is the major focus of our research. In particular, we are interested in the local scale ecology of three mosquitoes: the principal vector (Aedes triseriatus) and other potential vectors (Ae. albopictus and Ae. japonicus). Photo Credits: CDC



**Undergraduate Education (Environmental Health and Biology)** 

# Background

Culex pipiens and Culex restuans are primary vectors of West Nile virus in the eastern US

- Enzootic (avifaunal) transmission and magnification
- Primary vectors in epidemic transmission of WNV in many regions, but roles in transmission are complex and variable (Apperson 2002; Apperson 2004)
- Cx. pipiens is a more important vector than Cx. restuans





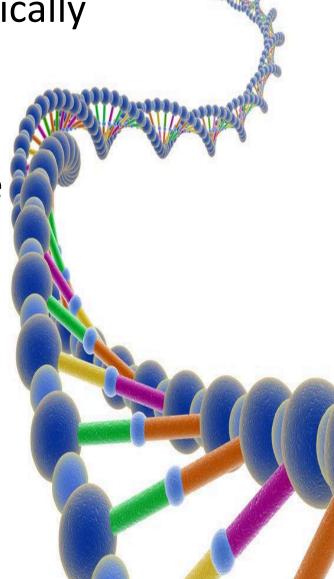
# Background

Difficult to differentiate morphologically

(NY) Harrington and Poulson 2008

Molecular ID by PCR more accurate

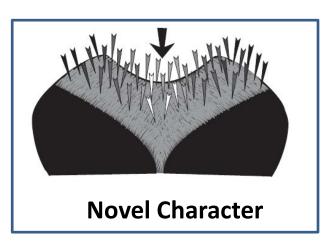
- Requires molecular techniques
- Resource intensive
- Contamination an issue



# Research Purpose

To determine the validity of 3 morphological characters used to identify *Cx. pipiens* and *Cx. restuans* 







Top of head with erect forked scales on median, and lateral areas of vertex and occiput black (Fig. 232); scutum with fine dark brown scales and usually 2 small round white spots (Fig. 233). . . . . . Cx. restuans

Top of head with several erect forked scales on median area of vertex and/or occiput light tan or pale, lateral erect forked scales dark brown (Fig. 234); scutum with coarse light tan scales, without small pale "Cx pipiens complex" Cx. quinquefasciatus (See Note 9)

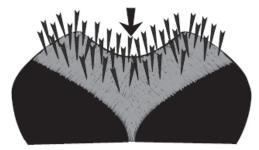


Fig. 232

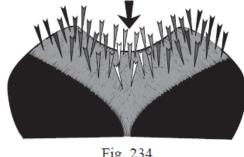


Fig. 234

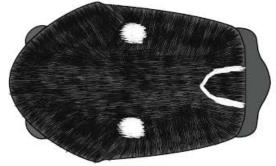


Fig. 233

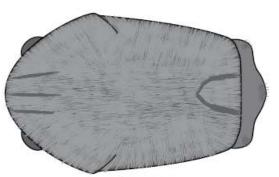
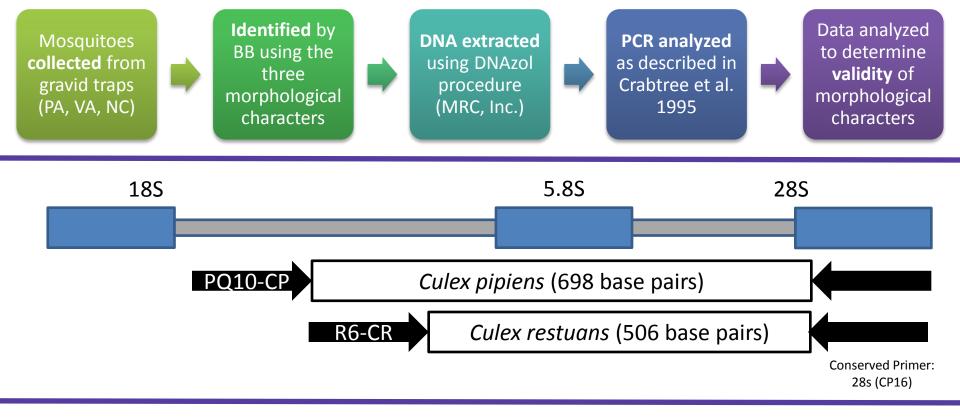
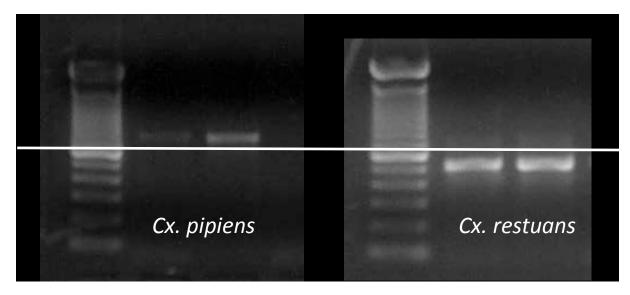


Fig. 235

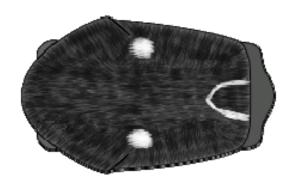
Keys to the Mosquitoes of the Mid-Atlantic Region, Harrison et al. (Unpublished)







# Scutal Spots Character



Culex restuans

	Culex restuans (PCR ID)		
	+	-	
Scutal Spots Present	30	0	
Scutal Spots Absent	28	58	

Sensitivity: 100% (95% CI: 88%-100%) Specificity: 67% (95% CI: 56%-77%)



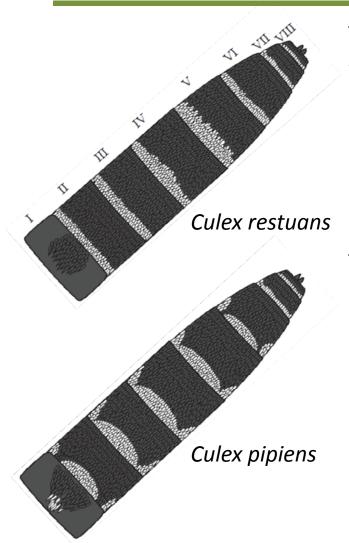
Culex pipiens

Although the specificity is low (67%), the sensitivity is perfect at 100%.

30 of 58 (52%) of the PCR confirmed *Culex restuans* had the dorsal spots on the scutum.

No Culex pipiens had these spots.

#### **Abdominal Band Character**



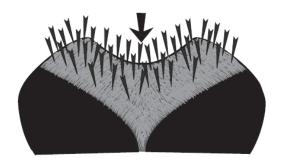
	Culex restuans (PCR ID)	
	+	-
Character Present (Straight Bands)	45	9
Character Missing (Centrally Enlarged)	21	70

Sensitivity: 83% (95% CI: 71%-92%) Specificity: 77% (95% CI: 70%-85%)

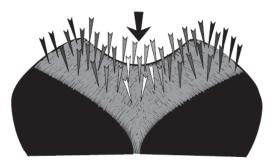
45 of 66 (68%) of PCR confirmed *Culex restuans* were identified with straight bands on the abdomen.

However, 70 of 79 (89%) *Culex pipiens* were identified correctly by the presence of a centrally enlarged band on the abdomen.

# Head (Vertex) Character



Culex restuans



Culex pipiens

ONLY PRESENT ON 72%
OF SPECIMENS

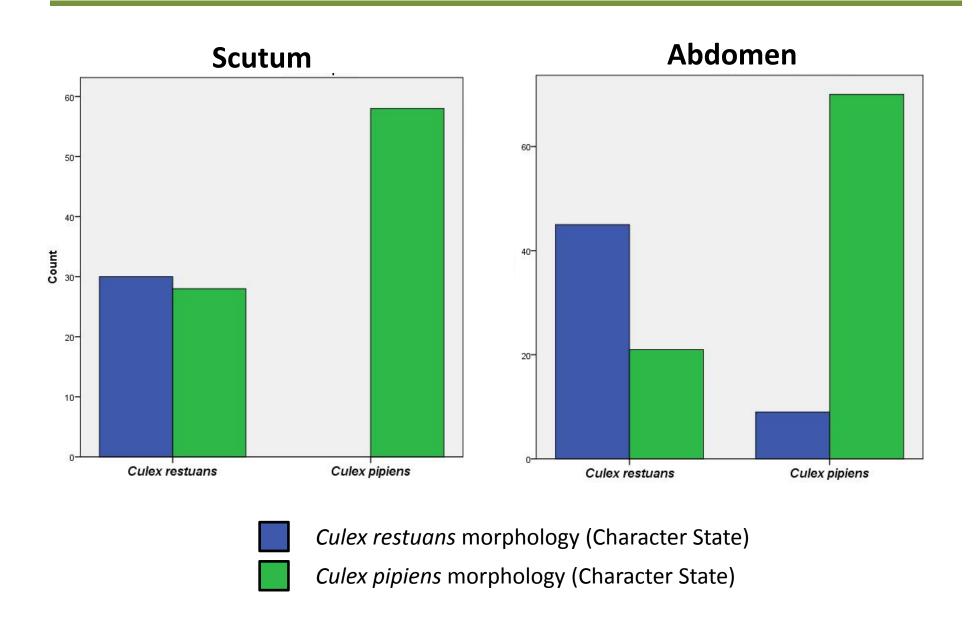
	Culex restuans (PCR ID)		
	+	-	
Character Present (Dark, erect, forked scales)	52	2	
Character Missing (Pale, erect, forked scales)	3	55	

Sensitivity: 96% (95% CI: 87%-99%) Specificity: 95% (95% CI: 86%-99%)

52 of 55 (95%) of the *Culex restuans* had dark erect forked scales on the vertex. Only 2 of 57(3.5%) of the *Culex pipiens* were misidentified by this character.

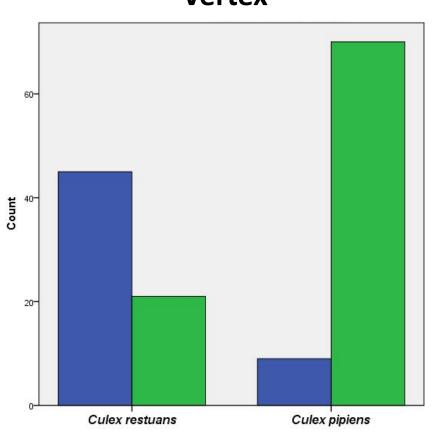
This character is <u>highly sensitive and specific when present</u>. However, of the 158 mosquitoes with PCR confirmed identities, this character was only present on 112 specimens (71%).

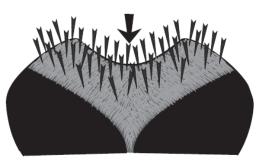
### "Traditional" Characters



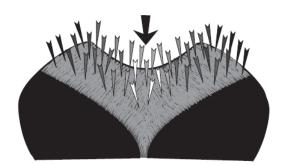
### "Novel" Character

#### **Vertex**





Culex restuans



Culex pipiens

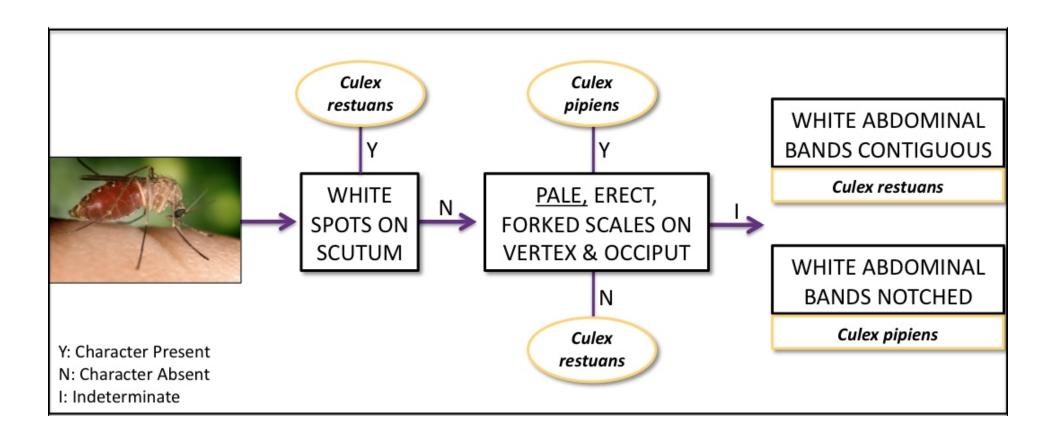
Culex restuans morphology (Character State)

Culex pipiens morphology (Character State)

# Take Home Messages

- The novel vertex character (pale vs black scales) works very well
  - Sensitivity= 96% for Cx. restuans
  - Better than the abdominal character
  - However, not always present (missing ~30% of the time)
- Scutal spots, when present, are 100% sensitive for *Cx. restuans* 
  - Of 58 Culex restuans, 28 (48%) were missing the pale scales.
  - Specificity is low (67%); absence does not identify Cx. pipiens
- The abdominal band character appears to be the least predictive
  - Perhaps does a better job at identifying Cx. pipiens than Cx. restuans
- Public health professionals would be best served by a hierarchy of multiple characters

# Take Home Messages



#### Selected References

- Crabtree, M.B., Savage, H.M. and B.R. Miller. 1995. Development of a Species-diagnostic Polymerase Chain Reaction Assay for the Identification of *Culex* Vectors of St. Louis Encephalitis Virus Based on Interspecies Sequence Variation in Ribosomal DNA Spacers. *American Journal of Tropical Medicine and Hygiene*. 53(1): 105-109.
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# Acknowledgements

#### **Funding:**

WCU College of Health and Human Sciences
WCU Honors College
Virginia Commonwealth University

### **Specimens:**

Ryan Harrison (Forsyth County, NC)

Questions?