ESSAY 2

Answers must be written out in paragraph form. Outline form is not acceptable. Labeled diagrams may be used to supplement discussion, but a diagram without a written explanation will not receive credit. You must cite the source of all outside information you include. Include the page number of information from the course textbook or the web address of information found online.

The evolutionary relationships within the Blattellidae subfamily of cockroaches are still highly disputed. In an effort to reconstruct the phylogenetic relationships within this taxon, tandem mass spectrometry was used to obtain the amino acid sequences of CAPA neuropeptides from several species of cockroaches (Roth, Fromm, Gäde, & Predel, 2009).

Capa genes code for several peptide hormones, including CAPA-periviscerokinins (CAPA-PVK-1, CAPA-PVK-2, CAPA-PVK-3) and CAPA-pyrokinins (CAPA-PK), that each bind to different receptor types (Predel & Wegener, 2006) and are thought to help insects survive periods without water (desiccation) and tolerate cold temperatures (Terhzaz et al., 2015). These neuropeptide ligands were selected for analysis because the authors claim that the evolution of the coding regions of their genes is constrained and may be strongly conserved (Roth et al., 2009).

(a) **Explain** why the authors claim that the coding regions of genes for neuropeptide ligands are likely to be strongly conserved.

Tables 1–4 show the amino acid sequences of the CAPA-PVK-1, CAPA-PVK-2, CAPA-PVK-3, and CAPA-PK polypeptides, respectively. Each amino acid is represented by a one-letter abbreviation (Table 5), and the amino acid residues in the polypeptide chains are numbered from the amino (NH₂) end to the carboxyl (COOH) end. Shaded columns indicate positions where single amino acid polymorphisms exist between the species. The question mark (?) character in Table 1 represents that *Loboptera decipiens* and *Symploce pallens* did not express the CAPA-PVK-1 polypeptide. Figure 1 represents the universal genetic code.

		Relative Amino Acid Position									
Species	1	2	3	4	5	6	7	8	9	10	11
Blattella germanica	G	S	S	G	L	Ι	Р	Μ	G	R	V
Loboptera decipiens	?	?	?	?	?	?	?	?	?	?	?
Supella dimidiata	G	S	S	G	L	Ι	Α	Μ	Р	R	V
Supella longipalpa	G	S	S	G	L	Ι	Α	М	Р	R	V
Symploce pallens	?	?	?	?	?	?	?	?	?	?	?

		Relative Amino Acid Position									
Species	1	2	3	4	5	6	7	8	9	10	11
Blattella germanica	G	S	S	G	L	Ι	S	М	Р	R	V
Loboptera decipiens	G	S	S	G	L	Ι	S	М	Р	R	V
Supella dimidiata	G	S	S	G	L	Ι	S	М	Р	R	V
Supella longipalpa	G	S	S	G	L	Ι	S	М	Р	R	V
Symploce pallens	G	S	S	G	L	Ι	S	М	Р	R	V

TABLE 3. CAPA-PVK-3 POLYPEPTIDE AMINO ACID SEQUENCE

		Relative Amino Acid Position									
Species	1	2	3	4	5	6	7	8	9	10	11
Blattella germanica	G	S	S	G	Μ	Ι	Р	F	Р	R	V
Loboptera decipiens	G	S	S	G	Μ	Ι	Р	F	Р	R	V
Supella dimidiata	G	S	S	G	М	Ι	Р	F	Р	R	V
Supella longipalpa	G	S	S	G	Μ	Ι	Р	F	Р	R	V
Symploce pallens	G	S	S	G	Μ	Ι	Р	F	Р	R	V

TABLE 4. CAPA-PK POLYPEPTIDE AMINO ACID SEQUENCE

		Relative Amino Acid Position															
Species	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
Blattella germanica	Е	S	G	G	S	G	Е	Α	Ν	G	М	W	F	G	Р	R	L
Loboptera decipiens	G	S	G	G	S	G	Е	Α	Ν	G	Μ	W	F	G	Р	R	L
Supella dimidiata	G	G	G	S	S	G	Е	Т	Ν	G	Μ	W	F	G	Р	R	L
Supella longipalpa	G	G	G	S	S	G	Е	Т	Ν	G	Μ	W	F	G	Р	R	L
Symploce pallens	Е	G	G	S	S	G	E	Α	S	G	Μ	W	F	G	Р	R	L

TABLE 5. ONE-LETTER AND THREE-LETTER AMINO ACID ABBREVIATIONS

				1				1 1				
	Abbro	Abbreviations Amino Acid			Abbreviations		Amino Acid		Abbreviations		Amino Acid	
[А	Ala	Alanine		Ι	Ile	Isoleucine		R	Arg	Arginine	
[С	Cys	Cysteine		Κ	Lys	Lysine		S	Ser	Serine	
[D	Asp	Aspartic acid		L	Leu	Leucine		Т	Thr	Threonine	
	Е	Glu	Glutamic acid		М	Met	Methionine		V	Val	Valine	
[F	Phe	Phenylalanine		Ν	Asn	Asparagine		W	Trp	Tryptophan	
[G	Gly	Glycine		Р	Pro	Proline		Y	Tyr	Tyrosine	
[Η	His	Histidine		Q	Gln	Glutamine					
L												

			Second Das				
		U	С	А	G		
	U	UUU UUC UUA UUG	$\begin{bmatrix} UCU \\ UCC \\ UCA \\ UCG \end{bmatrix}$ Ser	UAU UAC UAA Stop UAG Stop	UGU UGC UGA Stop UGG Trp	U C A G	
First Base in Codon	С	CUU CUC CUA CUG	$\left. \begin{matrix} CCU\\ CCC\\ CCA\\ CCG \end{matrix} \right\}_{Pro}$	CAU CAC His CAA CAA Gln	CGU CGC CGA CGG	U C A G	Third Base in Codon
First Base	A	AUU AUC AUA AUG Met or Start	$ \begin{array}{c} ACU \\ ACC \\ ACA \\ ACG \end{array} \right\} Thr$	$ \begin{array}{c} AAU \\ AAC \end{array} Asn \\ \begin{array}{c} AAA \\ AAA \\ AAG \end{array} Lys $	$ \begin{array}{c} AGU \\ AGC \end{array} \\ \begin{array}{c} Ser \\ AGA \\ AGG \end{array} \\ \begin{array}{c} Arg \end{array} $	U C A G	Third Base
	G	GUU GUC GUA GUG	GCU GCC GCA GCG	GAU GAC GAA GAA GAG Glu	GGU GGC GGA GGG	U C A G	

Second Base in Codon

Figure 1. Universal genetic code

(b)

- (i) Justify the use of amino acid sequence comparison as a measure of evolutionary relatedness.
- (ii) Describe ONE advantage of selecting polypeptides encoded by highly conserved genes for this comparison.
- (iii) Calculate the number of amino acid differences in the CAPA-PK polypeptide sequences shown in <u>Table 4</u> between each pair of species and enter the values from your calculations in the unshaded boxes on the table provided (<u>Table 6</u>).
- (iv) **Pose a scientific question** to connect the lack of CAPA-PVK-1 expression in *Loboptera decipiens* and *Symploce pallens* to a mechanism of gene regulation.

(c)

- (i) Using the data in <u>Table 4</u>, **construct** a cladogram on the template provided (Figure 2).
- (ii) Justify the placement of the outgroup on the cladogram.

(d)

- (i) Using <u>Table 1</u> and <u>Figure 1</u>, **predict** the difference in the <u>template</u> strand of DNA coding for the amino acid in the seventh position of the CAPA-PVK-1 polypeptide between *Blattella germanica* and *Supella longipalpa*.
- (ii) **Propose** an explanation for why *Supella dimidiata* and *Supella longipalpa* are considered separate species even though they have identical amino acid sequences for all four CAPA polypeptides.
- (iii) **Propose** ONE mechanism that is likely to prevent the hybridization of *Supella dimidiata* and *Supella longipalpa*.

References

- Predel, R. & Wegener, C. (2006). Biology of the CAPA peptides in insects. *Cell and Molecular Life Sciences*, 63(21), 2477–2490.
- Roth, S., Fromm, B., Gäde, G., & Predel, R. (2009). A proteomic approach for studying insect phylogeny: CAPA peptides of ancient insect taxa (Dictyoptera, Blattoptera) as a test case. *BMC Evolutionary Biology*, 9(50).
- Terhzaz, S., Teets, N. M., Cabrero, P., Henderson, L., Ritchie, M.G., Nachman, R. J., ... Davies, S. A. (2015). Insect capa neuropeptides impact desiccation and cold tolerance. *Proceedings of the National Academy of Sciences of the United States of America*, 112(9), 2882–2887.

TABLE 6. NUMBER OF AMINO ACID DIFFERENCES IN CAPA-PK POLYPEPTIDE

	B. germanica	L. decipiens	S. dimidiata	S. longipalpa	S. pallens
B. germanica	-				
L. decipiens		-			
S. dimidiata			-		
S. longipalpa				-	
S. pallens					-

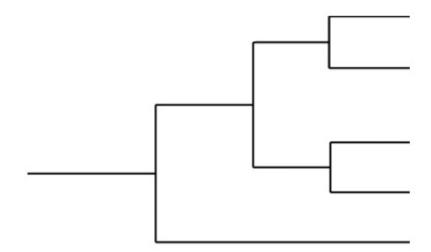


Figure 2. Cladogram representing the evolutionary relatedness of five cockroach species based on CAPA-PK polypeptide amino acid sequences.