

6. Housekeeping genes encode proteins involved in universally important processes such as transcription, translation, and glycolysis. Because these genes appear to be expressed in all cells at constant levels, the expression of housekeeping genes is often used as a control when comparing how the expression of other genes varies under different conditions.

Researchers studying the effect of pesticides on declining bee populations wanted to determine whether the expression of four housekeeping genes (*GAPDH*, *RPL32*, *RPS5*, and *TBP-AF*) was in fact constant in bees across different variables. The researchers collected samples of mRNA for each of the four genes and compared how their expression varied across the developmental stage of the bee, the sex of the bee, and the cell type from which the sample was taken. The mRNA from the samples was reverse transcribed to produce DNA copies of each gene. PCR was then used to amplify the DNA, and the Cq value was determined. The Cq value is the number of PCR cycles needed to produce a specified number of DNA copies. A high Cq value for a sample indicates the gene was expressed at a low level.

To analyze whether any of the examined variables affected expression of the housekeeping genes, researchers examined the range of Cq values for each gene in response to each variable. Genes with a wide range of Cq values were determined to be affected by the variable, while genes with a narrow range of Cq values were determined to be unaffected by the variable.

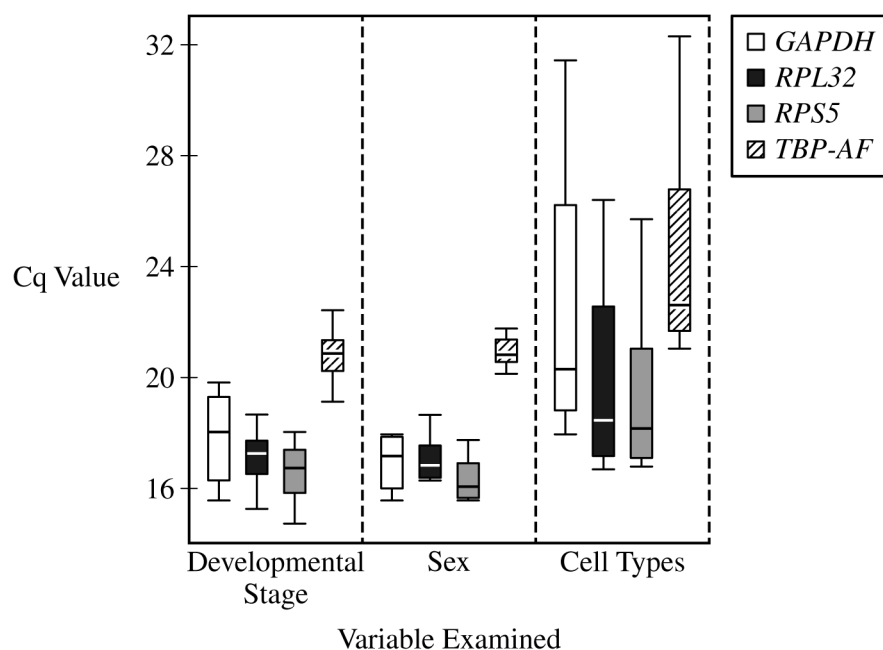


Figure 1. The effect of developmental stage, sex, and cell type on the Cq value of four housekeeping genes

- (a) Based on the data in Figure 1, **identify** the gene that had the lowest median Cq value when bees of different developmental stages were compared.
 - (b) The Cq value is inversely proportional to the amount of mRNA from that gene in the starting sample. Based on the data in Figure 1, **identify** the gene that has the lowest level of gene expression regardless of variable.
 - (c) The scientists investigated the effect of pesticides on the expression of other genes in one cell type of a group of bees containing males and females of the same developmental stage. They hypothesized that *TBP-AF* would serve as the best control gene for this experiment. Use the data to **evaluate** their hypothesis.
 - (d) **Explain** how expression of a gene such as *GAPDH* can vary from one cell type to another within the same bee.
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Write your responses to this question only on the designated pages in the separate Free Response booklet.

Question 6: Analyze Data**4 points**

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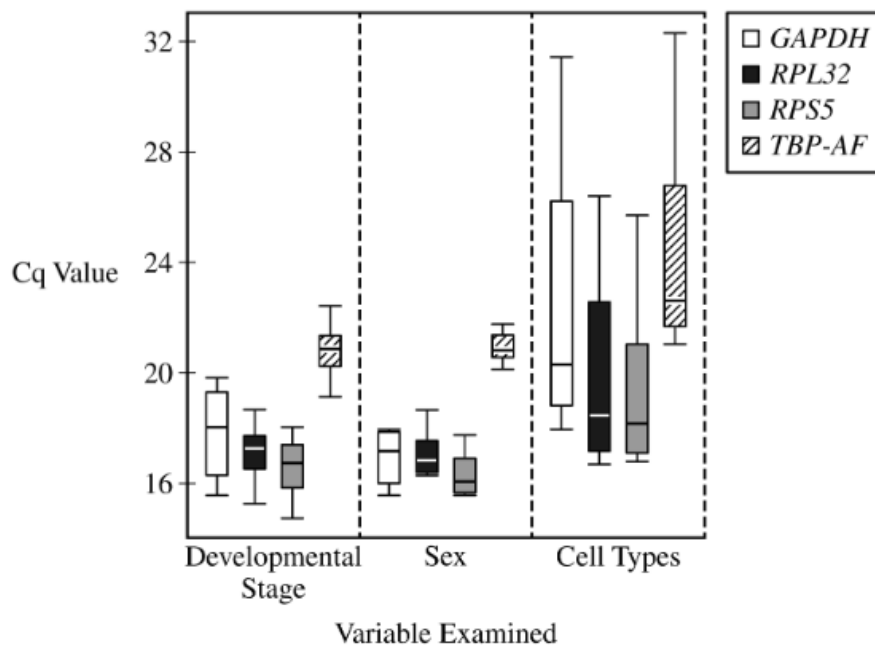


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(a)	Based on the data in <u>Figure 1</u> , identify the gene that had the lowest median Cq value when bees of different developmental stages were compared.	1 point
	<ul style="list-style-type: none">• <i>RPS5</i>	
(b)	The Cq value is inversely proportional to the amount of mRNA from that gene in the starting sample. Based on the data in <u>Figure 1</u> , identify the gene that has the lowest level of gene expression regardless of variable.	1 point
	<ul style="list-style-type: none">• <i>TBP-AF</i>	
(c)	The scientists investigated the effect of pesticides on the expression of other genes in one cell type of a group of bees containing males and females of the same developmental stage. They hypothesized that <i>TBP-AF</i> would serve as the best control gene for this experiment. Use the data to evaluate their hypothesis.	1 point
	<ul style="list-style-type: none">• Their hypothesis is supported because <i>TBP-AF</i> has the <u>smallest Cq range/most constant expression</u> (when comparing sexes).	
(d)	Explain how expression of a gene such as <i>GAPDH</i> can vary from one cell type to another within the same bee.	1 point
	<ul style="list-style-type: none">• Different cell types contain <u>different levels of/different</u> transcription factors, and therefore regulate the expression of genes in different ways.	
Total for question 6		4 points