Title	Detection of <i>Rhyzopertha dominica</i> larvae in stored wheat using ELISA: The impact of
	myosin degradation following fumigation
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Citation	Journal of Stored Products Research, Volume 43, Issue 2, 2007, Pages 156-159
Keywords	Rhyzopertha dominica; Insect fragments; Stored-grain

Abstract

Hard red winter wheat kernels were infested with eggs of *Rhyzopertha dominica*. After 20 d, when the larvae reached the fourth instar, they were killed by exposing the infested kernels to phosphine gas for 24 h. The infested kernels were then divided into four portions and treated as follows: one portion was immediately frozen at -80 °C to avoid myosin degradation; the other three portions were kept at 32 °C and 65% relative humidity, and then frozen at -80 °C after 14, 28, and 56 d post-fumigation, respectively. Each treatment was replicated five times. Myosin was measured using a commercial enzyme linked immunosorbent assay (ELISA) method that specifically detects this protein (Biotect[®], Austin, TX). Myosin degradation was most rapid in the first 2 weeks after the larvae were killed, decreasing from 1.672 to 0.695 ng/well during this period (a 58.4% reduction). There were no significant differences in myosin degradation between samples that were 14, 28, and 56 d post-fumigation. Grain is often fumigated to control insects. Frequently, this occurs many weeks before the grain is milled and may be repeated during the storage period. Therefore, estimates using the ELISA test may underestimate internal insect infestation because of myosin degradation. Insect fragment estimates for previously fumigated grain could be underestimated by as much as 58%.