U.S. Wheat and Barley Scab Initiative
FY01 Final Performance Report (approx. May 01 – April 02)
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Cover Page

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Year:	**FY2001 (approx. May 01 – April 02)**
Grant Number:	**59-0790-9-060**
Grant Title:	**Fusarium Head Blight Research**
FY01 ARS Award Amount:	**$ 56,027**

Project

Program Area	Project Title	Requested Amount
Food Safety	Human Susceptibility to Trichothecene Mycotoxins	$ 59,594
	Total Amount Requested	**$ 59,594**

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Principal Investigator Date

Project 1: Human Susceptibility to Trichothecene Mycotoxins

1. What major problem or issue is being resolved and how are you resolving it?

Vomitoxin (VT or deoxynivalenol) and other trichothecenes are elaborated during head blight and thus pose a potential threat to human health. There have been several European studies that have suggested that a lower action level for VT be considered (120 ppb) rather than the 1-2 ppm being employed by most countries. The Joint Expert Committee on Food Additives (JEFCA) is releasing safety concerns for VT and other mycotoxins. I worked with this group and the International Life Sciences Research Institute to provide a state-of-the-art review on toxicological issues related to VT. In addition, the Japanese government has established new regulations for VT in food. I have collaborated with one of the Dr. Yohsiko Kinoshi who is Section Chief of Microbiology at the National Institute of Health Sciences in Tokyo relative to risk model and appropriate analytical methods.

Based on studies in the mouse immune system, we believe that the most critical step for VT toxicity induction is its action on cell signaling in leukocytes (white blood cells). We currently evaluating whether human leukocyte cytokine production and/or apoptosis induction are indeed targeted by the same levels of VT and related 8-ketotrichothecenes as are their mouse equivalents. If this is true, then the risk of low ppm levels of VT to humans will be extremely small when one considers the diversity of the human diet and the actual potential level of VT exposure in human tissues. Such evidence is critical because it would support the argument against establishing lower action levels than those currently set for VT.

2. What were the most significant accomplishments?

a. Completed evaluation of trichothecene effects on cytokine secretion and cell death in cloned human macrophage model (U-937 cells).
b. Identified underlying kinase mechanism for trichothecene-induced cell death in cloned human macrophage model (U-937 cells).
c. Completed evaluation of trichothecene effects on a cloned human T lymphocyte model (Jurkat cells).
d. Evaluated conditions for the primary culture of human leukocytes and conducted experiments on the effects of VT on cytokine secretion. Two culture approaches are being used. The first involves direct culturing of human blood obtained from volunteers. For the second approach, we have used human blood cells that are a by-product of donor blood processing. In both approaches, we have observed that VT will directly induce IL-6 and IL-8. We have determined that in both approaches that there is tremendous variability in how a donor's cells will respond. Some donors' white blood cells are highly sensitive to the VT whereas others appear to be completely resistant. Because cytokine receptors may impair our ability to fully detect a response, we have devised highly sensitive real-time PCR methods for measuring mRNAs of four cytokines. Again, in preliminary experiments with IL-8 mRNA, we have observed differential sensitivity to VT across different donor's blood samples. This is a critical observation because it *suggests some people may be resistant to VT whereas others are sensitiv*e. This implies that a environmental or genetic predisposition for VT sensitivity/resistance might exist which must be taken into account in hazard/risk assessments. We are expanding our donor pool and collecting multiple samples to validate these findings relative to multiple cytokines. In addition, we will also be conducting cytotoxicity assays to determine if there is a similar differential sensitivity.

Include below a list of the publications, presentations, peer-reviewed articles, and non-peer reviewed articles written about your work that resulted from all of the projects included in the grant. Please reference each item using an accepted journal format. If you need more space, continue the list on the next page.

a. Moon, Y.S. and J.J. Pestka. 2002. Vomitoxin-induced cyclooxygenase-2 gene expression in macrophages mediated by activation of ERK and p38 but not JNK Mitogen-Activated Protein Kinase. Toxicol. Appl. Pharmacol. (in press)
b. Chung, Y.J., B.Jarvis, And J.J. Pestka. 2002. Modulation of lipopolysaccharide-induced proinflammatory cytokine production by satratoxins and other macrocyclic trichothecenes in the macrophage. J.Toxicol. Environ. Health (in press)
c. Pestka, J.J. and H.R. Zhou. 2002. Effects of tumor necrosis factor type 1 and 2 deficiencies on anorexia, growth and IgA dysregulation in mice exposed to the trichothecene vomitoxin. Food and Chem. Toxicol. (in press)
d. Yang, G-H., and J.J. Pestka. 2002. Vomitoxin (deoxynivalenol)-mediated inhibition of nuclear protein binding to NRE-A, an IL-2 promoter negative regulatory element, in EL-4 cells. Toxicology. 172:169-179
e. Wong, S.S., H-R. Zhou, and J.J. Pestka. 2002. Effects of vomitoxin (deoxynivalenol) on the binding of transcription factors AP-1, NF-?B and NF-IL6 in raw 264.7 macrophage cells. J. Toxicol. Environ. Health. 65:101-120
f. Pestka, J.J., H-R. Zhou, Q. Jia, and A.M. Timmer. 2002. Dietary fish oil suppresses experimental IgA nephropathy in mice. J. Nutrition. 132:261-269
g. Islam, Z., Y.S. Moon, H-R. Zhou, L.E. King, P.J. Fraker, and J.J. Pestka. 2002. Endotoxin potentiation of trichothecene-induced lymphocyte apoptosis is mediated by up-regulation of glucocorticoids. Toxicol. Appl. Pharmacol. 180:43-55.
h. Zhou, H-R., A.S. Lau, and J.J. Pestka. 2002. An essential role of double-stranded RNA-activated protein kinase PKR in vomitoxin-induced apoptosis in raw 264.7 cells. 41st. Society of Toxicology Annual Meeting, Nashville, TN.
i. Jia, Q., and J.J. Pestka. 2002. Dietary fish oil suppresses vomitoxin-induced IgA nephropathy in the mouse. 41st. Society of Toxicology Annual Meeting, Nashville, TN
j. Islam, Z., L.E. King, P.J. Fraker, and J.J. Pestka. 2002. Differential induction of apoptosis in murine lymphoid subpopulations in vivo following co-exposure to lipopolysaccharide and vomitoxin. 41st. Society of Toxicology Annual Meeting, Nashville, TN
k. Moon, Y., and J.J. Pestka. 2002. Relationship of trichothecene structure to Cox-2 induction in the macrophage model. 41st. Society of Toxicology Annual Meeting, Nashville, TN
l. Chung, Y., and J.J. Pestka. 2002. Enhancement of TNF alpha-mRNA stability by deoxynivalenol through the activation of p38 kinase. 41st. Society of Toxicology Annual Meeting, Nashville, TN.
m. Sugita-Kinoshi, Y and Pestka, J.J. Differential. 2001 Up-regulation of TNF-?IL-6 and IL-8 Production by Deoxynivalenol (Vomitoxin) and Other 8-Ketotrichothecenes in a Human Macrophage Model. J. Toxicol. Environ. Health. (in press)
n. Pestka, J.J. Deoxynivalenol: Toxicology and Potential Effects on Humans. 2001. Report submitted to the International Life Sciences Institute

o. Chung, Y-H., G-H. Yang, and J.J. Pestka. 2001. Up-regulation of macrophage inflammatory protein-2 and complement 3A receptor by trichothecenes. Abstr. 40th Ann. Meet. Soc. Toxicol. (San Francisco, CA).
p. Zhou, H.R. and J.J. Pestka. 2001. Activation of JNK, ERK and P38 mitogen-activated protein kinases by the trichothecene vomitoxin (deoxynivalenol): Role of translational arrest and double stranded RNA-dependent protein kinase R (PKR). Abstr. 40th Ann. Meet. Soc. Toxicol. (San Francisco, CA).
q. Hinton, D.M., M.J. Myers, A. Perltoni, F. Hines, R. Raybourne, R.E. Sotomayor, J. Shaddock, A. Warbritton, M. Chou and J.J. Pestka 2001. Enhanced histopathology with morphometry, immuno- and in-situ staining in immunotoxicity studies of the mycotoxins aflatoxin B_1 and deoxynivalenol. Abstr. 40th Ann. Meet. Soc. Toxicol. (San Francisco, CA).
r. Uzarski, R. and J.J. Pestka. 2001. Upregulation of mitogen-activated protein kinases (MAPKs) and caspase-3 by vomitoxin (deoxynivalenol) in Jurkat T cells. Abst. 40th Ann. Meet. Soc. Toxicol. (San Francisco, CA)
s. Zhou, H.R. and J.J. Pestka. 2001. Essential role of non-receptor tyrosine kinase Hck in vomitoxin-induced phosphorylation of JNK, ERK, and p38 mitogen-activated protein kinases. Abstr. Soc. Exp. Biol. Ann Meet. (Orlando, FL).
t. Islam, Z. and J.J. Pestka. 2001. Role of corticosteroids in LPS-mediated potentiation of trichothecene-induced lymphocyte apoptosis. Abstr. Soc. Exp. Biol. Ann. Meet. (Orlando, FL).