Reference List
Permethrin Spreadsheet
September, 2007

1. Abdel-Rahman, A.; Abou-Donia, S.; El-Masry, E.; Shetty, A., and Abou-Donia, M. Stress and combined exposure to low doses of pyridostigmine bromide, DEET, and permethrin produce neurochemical and neuropathological alterations in cerebral cortex, hippocampus, and cerebellum. Journal of Toxicology & Environmental Health. Part A. 2004; 67(2):163-192. ISSN: 1528-7394.
Abstract: Exposure to a combination of stress and low doses of the chemicals pyridostigmine bromide (PB), DEET, and permethrin in adult rats, a model of Gulf War exposure, produces blood-brain barrier (BBB) disruption and neuronal cell death in the cingulate cortex, dentate gyrus, thalamus, and hypothalamus. In this study, neuropathological alterations in other areas of the brain where no apparent BBB disruption was observed was studied following such exposure. Animals exposed to both stress and chemical exhibited decreased brain acetylcholinesterase (AChE) activity in the midbrain, brainstem, and cerebellum and decreased m2 muscarinic acetylcholine (ACh) receptor ligand binding in the midbrain and cerebellum. These alterations were associated with significant neuronal cell death, reduced microtubule-associated protein (MAP-2) expression, and increased glial fibrillary acidic protein (GFAP) expression in the cerebral cortex and the hippocampal subfields CA1 and CA3. In the cerebellum, the neurochemical alterations were associated with Purkinje cell loss and increased GFAP immunoreactivity in the white matter. However, animals subjected to either stress or chemicals alone did not show any of these changes in comparison to vehicle-treated controls. Collectively, these results suggest that prolonged exposure to a combination of stress and the chemicals PB, DEET, and permethrin can produce significant damage to the cerebral cortex, hippocampus, and cerebellum, even in the absence of apparent BBB damage. As these areas of the brain are respectively important for the maintenance of motor and sensory functions, learning and memory, and gait and coordination of movements, such alterations could lead to many physiological, pharmacological, and behavioral abnormalities, particularly motor deficits and learning and memory dysfunction.

2. Abdel-Rahman, A.; Dechkovskaia, A. M.; Goldstein, L. B.; Bullman, S. H.; Khan, W.; El-Masry, E. M., and Abou-Donia, M. B. Neurological deficits induced by malathion, DEET, and permethrin, alone or in combination in adult rats. Journal of Toxicology & Environmental Health. Part A. 2004; 67(4):331-356. ISSN: 1528-7394.
Abstract: Malathion (O,O-dimethyl-S-[1,2-carbethoxyethyl]phosphorodithionate), DEET (N,N-diethyl-m-toluamide), and permethrin [(+/-)-cis/trans-3-(2,2-dichloroethenyl)-2,2-dimethylcyclopropane carboxylic acid (3-phenoxyphenyl) methyl ester] are commonly used pesticides. To determine the effects of the dermal application of these chemicals, alone or in combination, the sensorimotor behavior, central cholinergic system, and histopathological alterations were studied in adult male Sprague-Dawley rats following a daily dermal dose of 44.4 mg/kg malathion, 40 mg/kg DEET, and 0.13 mg/kg permethrin, alone and in combination for 30 d. Neurobehavioral evaluations of sensorimotor functions included beam-walking score, beam walk time, inclined plane, and grip response assessments. Twenty-four hours after the last treatment with each chemical alone or in combination all behavioral measures were impaired. The combination of DEET and permethrin, malathion and permethrin, or the three chemicals together resulted in greater impairments in inclined performance than permethrin alone. Only animals treated with a combination of DEET and malathion or with DEET and permethrin exhibited significant increases in plasma butyrlcholinesterase (BChE) activity. Treatment with DEET or permethrin alone, malathion and permethrin, or DEET and permethrin produced significant increases in cortical acetylcholinesterase (AChE) activity. Combinations of malathion and permethrin or of DEET and permethrin produced significant decreases in midbrain AChE activity. Animals treated with DEET alone exhibited a significant increase in cortical m2 muscarinic ACh receptor binding. Quantification of neuron density in the dentate gyrus, CA1 and CA3 subfields of the hippocampus, midbrain, brainstem, and cerebellum revealed significant reductions in the density of surviving neurons with various treatments. These results suggest that exposure to real-life doses of malathion, DEET, and permethrin, alone or in combination, produce no overt signs of neurotoxicity but induce

significant neurobehavioral deficits and neuronal degeneration in brain.

3. Abdel-Rahman, A.; Shetty, A. K., and Abou-Donia, M. B. Disruption of the blood-brain barrier and neuronal cell death in cingulate cortex, dentate gyrus, thalamus, and hypothalamus in a rat model of Gulf-War syndrome. Neurobiology of Disease. 2002; 10(3):306-326. ISSN: 0969-9961.
Abstract: We investigated the effects of a combined exposure to restraint stress and low doses of chemicals pyridostigmine bromide (PB), N, N-diethyl-m-toluamide (DEET), and permethrin in adult male rats, a model of Gulf-War syndrome. Animals were exposed daily to one of the following for 28 days: (i) a combination of stress and chemicals (PB, 1.3 mg/kg/day; DEET, 40 mg/kg/day; and permethrin, 0.13 mg/kg/day); (ii) stress and vehicle; (iii) chemicals alone; and (iv) vehicle alone. All animals were evaluated for: (i) the disruption of the blood-brain barrier (BBB) using intravenous horseradish peroxidase (HRP) injections and endothelial barrier antigen (EBA) immunostaining; (ii) neuronal cell death using H&E staining, silver staining, and glial fibrillary acidic protein (GFAP) immunostaining; and (iii) acetylcholinesterase (AChE) activity and m2-muscarinic acetylcholine receptors (m2-AChR). Animals subjected to stress and chemicals exhibited both disruption of the BBB and neuronal cell death in the cingulate cortex, the dentate gyrus, the thalamus, and the hypothalamus. Other regions of the brain, although they demonstrated some neuronal cell death, did not exhibit disruption of the BBB. The neuropathological changes in the above four brain regions were highly conspicuous and revealed by a large number of HRP-positive neurons (21-40% of total neurons), a decreased EBA immunostaining (42-51 % reduction), a decreased number of surviving neurons (27-40% reduction), the presence of dying neurons (4-10% of total neurons), and an increased GFAP immunostaining (45-51 % increase). These changes were also associated with decreased forebrain AChE activity and m2-AchR (19-25% reduction). In contrast, in animals exposed to stress and vehicle or chemicals alone, the above indices were mostly comparable to that of animals exposed to vehicle alone. Thus, a combined exposure to stress and low doses of PB, DEET, and permethrin leads to significant brain injury. The various neurological symptoms reported by Gulf-War veterans could be linked to this kind of brain injury incurred during the war.

4. ---. Subchronic dermal application of N,N-diethyl m-toluamide (DEET) and permethrin to adult rats, alone or in combination, causes diffuse neuronal cell death and cytoskeletal abnormalities in the cerebral cortex and the hippocampus, and Purkinje neuron loss in the cerebellum. Experimental Neurology. 2001; 172(1):153-171. ISSN: 0014-4886.
Abstract: N,N-Diethyl m-toluamide (DEET) and permethrin have been implicated as potential neurotoxic agents that may have played an important role in the development of illnesses in some veterans of the Persian Gulf War. To determine the effect of subchronic dermal application of these chemicals on the adult brain, we evaluated histopathological alterations in the brain of adult male rats following a daily dermal dose of DEET (40 mg/kg in 70% ethanol) or permethrin (0.13 mg/kg in 70% ethanol) or a combination of the two for 60 days. Control rats received a daily dermal dose of 70% ethanol for 60 days. Animals were perfused and brains were processed for morphological and histopathological analyses following the above regimen. Quantification of the density of healthy (or surviving) neurons in the motor cerebral cortex, the dentate gyrus, the CA1 and CA3 subfields of the hippocampus, and the cerebellum revealed significant reductions in all three treated groups compared with the control group. Further, animals receiving either DEET or permethrin exhibited a significant number of degenerating (eosinophilic) neurons in the above brain regions. However, degenerating neurons were infrequent in animals receiving both DEET and permethrin, suggesting that neuronal cell death occurs earlier in animals receiving combined DEET and permethrin than in animals receiving either DEET or permethrin alone. The extent of neuron loss in different brain regions was similar among the three treatment groups except the dentate gyrus, where neurodegeneration was significantly greater with exposure to DEET alone. The neuron loss in the motor cerebral cortex and the CA1 subfield of all treated groups was also corroborated by a significant decrease in microtubule associated protein 2-immunoreactive elements (15-52% reduction), with maximal reductions occurring in rats receiving DEET alone; further, the surviving neurons in animals receiving both DEET and permethrin exhibited wavy and beaded dendrites. Analysis of glial fibrillary acidic protein immunoreactivity revealed significant hypertrophy of astrocytes in the hippocampus and the cerebellum of all treated groups (24-106% increase). Thus, subchronic dermal application of DEET and permethrin to adult rats, alone or in combination, leads

to a diffuse neuronal cell death in the cerebral cortex, the hippocampal formation, and the cerebellum. Collectively, the above alterations can lead to many physiological, pharmacological, and behavioral abnormalities, particularly motor deficits and learning and memory dysfunction.

5. Abou-Donia, M. B.; Dechkovskaia, A. M.; Goldstein, L. B.; Abdel-Rahman, A.; Bullman, S. L., and Khan, W. A. Co-exposure to pyridostigmine bromide, deet, and/or permethrin causes sensorimotor deficit and alterations in brain acetylcholinesterase activity. Pharmacology, Biochemistry & Behavior. 2004; 77(2):253-262. ISSN: 0091-3057.
Abstract: Military personnel deployed in the Persian Gulf War (PGW) were exposed to a combination of chemicals, including pyridostigmine bromide (PB), DEET, and permethrin. We investigated the dose-response effects of these chemicals, alone or in combination, on the sensorimotor performance and cholinergic system of male Sprague-Dawley rats. Animals were treated with a daily dermal dose of DEET and/or permethrin for 60 days and/or PB (gavage) during the last 15 days. Neurobehavioral performance was assessed on day 60 following the beginning of the treatment with DEET and permethrin. The rats were sacrificed 24 h after the last treatment for biochemical evaluations. PB alone, or in combination with DEET, or DEET and permethrin resulted in deficits in beam-walk score and longer beam-walk times compared to controls. PB alone, or in combination with DEET, permethrin, or DEET and permethrin caused impairment in incline plane performance and forepaw grip strength. PB alone at all doses slightly inhibited plasma butyrylcholinesterase activity, whereas combination of PB with DEET or permethrin increased its activity. Brainstem acetylcholinesterase (AChE) activity significantly increased following treatment with combinations of either DEET or permethrin at all doses, whereas the cerebellum showed a significant increase in AChE activity following treatment with a combination of PB/DEET/permethrin. Co-exposure to PB, DEET, and permethrin resulted in significant inhibition in AChE in midbrain. PB alone or in combination with DEET and permethrin at all doses increased ligand binding for m2 muscarinic acetylcholine receptor in the cortex. In addition, PB and DEET together or a combination of PB, DEET, and permethrin significantly increased ligand binding for nicotinic acetylcholine receptor. These results suggest that exposure to various doses of PB, alone and in combination with DEET and permethrin, leads to sensorimotor deficits and differential alterations of the cholinergic system in the CNS.

6. Abou-Donia, M. B.; Goldstein, L. B.; Dechovskaia, A.; Bullman, S.; Jones, K. H.; Herrick, E. A.; Abdel-Rahman, A. A., and Khan, W. W. Effects of daily dermal application of deet and permethrin, alone and in combination, on sensorimotor performance, blood-brain barrier, and blood-testis barrier in rats. Journal of Toxicology & Environmental Health. Part A. 2001; 62(7):523-541. ISSN: 1528-7394.
Abstract: DEET and permethrin were implicated in the development or illnesses in some veterans of the Persian Gulf War. This study was designed to investigate the effects of daily dermal application of these chemicals, alone or in combination, on the permeability of the blood-brain barrier (BBB) and blood-testes barrier (BTB) and on sensorimotor performance in male Sprague-Dawley rats. Groups of five rats were treated with a dermal daily dose of 4, 40, or 400 mg/kg DEET in ethanol or 0.013, 0.13, or 1.3 mg/kg permethrin in ethanol for 60 d. A group of 10 rats receive d a daily dermal dose or ethanol and served as controls. BBB permeability was assessed by injection of an iv dose of the quaternary ammonium compound [H-3]hexamethonium iodide. While permethrin produced no effect on BBB permeability, DEET alone caused a decrease in BBB permeability in brainstem. A combination of DEET and permethrin significantly decreased the BBB permeability in the cortex. BTB permeability was decreased by treatment with DEET alone and in combination with permethrin. The same animals underwent a battery of functional behavior tests 30, 45, and 60 d after exposure to evaluate their sensorimotor abilities. All treatments caused a significant decline in sensorimotor performance in a dose- and time-dependent manner. These results show that daily dermal exposure to DEET, alone or in combination with permethrin, decreased BBB permeability in certain brain regions, and impaired sensorimotor performance.

7. Abou-Donia, M. B.; Goldstein, L. B.; Jones, K. H.; Abdel-Rahman, A. A.; Damodaran, T. V.; Dechkovskaia, A. M.; Bullman, S. L.; Amir, B. E., and Khan, W. A. Locomotor and sensorimotor performance deficit in rats following exposure to pyridostigmine bromide, deet, and permethrin, alone and in

combination. Toxicological Sciences. 2001; 60(2):305-314. ISSN: 1096-6080.
Abstract: Since their return from Persian Gulf War (PGW), many veterans have complained of symptoms including muscle and joint pain, ataxia, chronic fatigue, headache, and difficulty with concentration. The causes of the symptoms remain unknown. Because these veterans were exposed to a combination of chemicals including pyridostigmine bromide (PB), DEET, and permethrin, we investigated the effects of these agents, alone and in combination, on the sensorimotor behavior and central cholinergic system of rats. Male Sprague-Dawley rats (200-250 gm) were treated with DEET (40 mg/kg, dermal) or permethrin (0.13 mg/kg, dermal), alone and in combination with PB (1.3 mg/kg, oral, last 15 days only), for 45 days. Sensorimotor ability was assessed by a battery of behavioral tests that included beam-walk score, beam-walk time, incline plane performance, and forepaw grip on days 30 and 45 following the treatment. On day 45 the animals were sacrificed, and plasma and CNS cholinesterase, and brain choline acetyl transferase, muscarinic and nicotinic acetylcholine receptors were evaluated. Animals treated with PB, alone or in combination with DEET and permethrin, showed a significant deficit in beam-walk score as well as beam-walk time as compared with controls. Treatment with either DEET or permethrin, alone or in combination with each other, did not have a significant effect on beam-walk score. All chemicals, alone or in combination, resulted in a significant impairment in incline plane testing on days 30 and 45 following treatment. Treatment with PB, DEET, or permethrin alone did not have any inhibitory effect on plasma or brain cholinesterase activities, except that PB alone caused moderate inhibition in midbrain acetylcholinesterase (AChE) activity. Treatment with permethrin alone caused significant increase in cortical and cerebellar AChE activity. A combination of DEET and permethrin or PB and DEET led to significant decrease in AChE activity in brainstem and midbrain and brainstem, respectively. A significant decrease in brainstem AChE activity was observed following combined exposure to PB and permethrin. Coexposure with PB, DEET, and permethrin resulted in significant inhibition in AChE in brainstem and midbrain. No effect was observed on choline acetyl transferase activity in brainstem or cortex, except combined exposure to PB, DEET, and permethrin caused a slight but significant increase in cortical choline acetyltransferase activity. Treatment with PB, DEET, and permethrin alone caused a significant increase in ligand binding for m2 muscarinic acetylcholine receptor (mAChR) in the cortex. Coexposure to PB, DEET, and permethrin did not have any effect over that of PB-induced increase in ligand binding. There was no significant change in ligand binding for nicotinic acetylcholine receptor (nAChR) associated with treatment with the chemical alone; a combination of PB and DEET or coexposure with PB, DEET, and permethrin caused a significant increase in nAChR ligand binding in the cortex. Thus, these results suggest that exposure to physiologically relevant doses of PB, DEET, and permethrin, alone or in combination, leads to neurobehavioral deficits and region-specific alterations in AChE and acetylcholine receptors.

8. Abou-Donia, M. B.; Suliman, H. B.; Khan, W. A., and Abdel-Rahman, A. A. Testicular germ-cell apoptosis in stressed rats following combined exposure to pyridostigmine bromide, *N,N*-diethyl *m*-toluamide (DEET), and permethrin. Journal of Toxicology & Environmental Health. Part A. 2003; 66(1):57-73.
Abstract: This study reports and characterizes the testicular apoptosis following daily exposure of male Sprague-Dawley rats to subchronic combined doses of pyridostigmine bromide (PB, 1.3 mg/kg/d in water, oral), a drug used for treatment of myasthenia gravis and prophylactic treatment against nerve agents during the Persian Gulf War, the insect repellent N,N-diethyl m-toluamide (DEET, 40 mg/kg/d in ethanol, dermal), and the insecticide permethrin (0.13 mg/kg in ethanol, dermal), with and without stress for 28 d. Combined. exposure to these chemicals was implicated in the development of illnesses including genitourinary disorders among many veterans of the Persian Gulf War. Previous studies from this laboratory have shown that exposure to combination of these chemicals produced greater toxicity compared to single components. Exposure to stress alone did not cause any significant histopathological alterations in the testes. Administration of combination of these chemicals induced apoptosis in rat testicular germ cells, Sertoli cells, and Leydig cells, as well as in the endothelial lining of the blood vessels. Testicular damage was significantly augmented when the animals were further exposed to a combination of chemicals and stress. Histopathological examination of testicular tissue sections showed that apoptosis was confined to the basal germ cells and spermatocytes, indicating suppression of spermatogenesis. Increased apoptosis of testicular cells coincided, in timing and localization, with increased expression of the apoptosis-promoting proteins

Bax and p53. Furthermore, significant increase of 3-nitrotyrosine immunostaining in the testis revealed oxidative and/or nitrosation induction of cell death. In conclusion, combined exposure to real-life doses of test compounds caused germ-cell apoptosis that was significantly enhanced by stress.

9. Abou-Donia, M. B.; Wilmarth, K. R.; Jensen, K. F.; Oehme, F. W., and Kurt, T. L. Neurotoxicity resulting from coexposure to pyridostigmine bromide, DEET, and permethrin: implications for Gulf War chemical exposures. Journal of Toxicology & Environmental Health. 1996; 48:35-56.
Abstract: Of the three-quarters of a million service personnel involved in the Persian Gulf War, approximately 30,000 have complained of neurological symptons of unknown etiology. One contributing factor to the emergence of such symptoms may be the simultaneous exposure to multiple agents used to protect the health of service personnel, in particular, the anti-nerve agent pyridostigmine bromide (PB; 3-dimtheylaminocarbonyloxy-N-methylpyridium bromide), the insect repellent DEET (N,N-diethyl-m-toluamide), and the insecticide permethrin (3-(2,2-dichloroethenyl)-2,2-dimethylcyclopropanecarboxylic acid (3-phenoxyphenyl)methyl ester). This study investigated neurotoxicity produced in hens by individual or simultaneous exposure to these agents (5 d/wk for 2 months to 5 mg/k/d PB in water, po; 500 mg/kg/d DEET, neat,sc; and 500 mg/kg/d permethrin in corn oil,sc). At these dosages, exposure to single compounds resulted in minimal toxicity. Combinations of two agents produced greater neurotoxicity than that caused by individual agents. Neurotoxicity was further enhanced following concurrent administration of all three agents. We hypothesize that competition for liver and plasma esterases by these compounds leads to their decreased breakdown and increased transport of the parent compound to nervous tissues. Thus, carbamylation of peripheral esterases by PB reduces the hydrolysis of DEET and permethrin and increases their availability to the nervous system. In effect, PB "pumps" more DEET and permethrin into the central nervous system. Consistent with this hypothesis, hens exposed to the combination of the three agents exhibited neuropathological lesions with several characteristics similar to those previously reported in studies of near-lethal doses of DEET and permethrin. If this hypothesis is correct, then blood and liver esterases play an important "buffering" role in protecting againse neurotoxicity in the population at large. It also suggests that individuals with low plasma esterase activity may be predisposed to neurologic deficits produced by exposure to certain chemical mixtures.

10. Abu-Qare, A. and Abou-Donia, M. Increased 8-hydroxy-2 '-deoxyguanosine, a biomarker of oxidative DNA damage in rat urine following a single dermal dose of DEET (n,n-diethyl-m-toluamide), and permethrin, alone and in combination. Toxicology Letters. 2000; 117(3):151-160.
Abstract: Levels of the biomarker of DNA oxidative damage 8-hydroxy-2'-deoxyguanosine (8-OHdG) in rat urine following dermal exposure to DEET (N,N'-diethyl-m-toluamide) and permethrin, alone and in combination have been determined. A group of five rats for each time point were treated with a single dermal dose of 400 mg/kg of DEET, 1.3 mg/kg of permethrin or their combination. Urine samples were collected 2, 4, 8, 16, 24, 48, and 72 h following application. Control urine samples of rats treated with ethanol were also collected at the same time intervals. Solid phase extraction coupled with high performance liquid chromatography (HPLC) with UV detection at 254 nm was used for determination of 2'-deoxyguanosine, and (8-OHdG). The limits of detection (LOD) were 0.5 ng of both 2'-deoxyguanosine and 8-OHdG. Their average percentage recoveries from urine samples were between 70-85%. A single dermal dose of DEET or in combination with permethrin significantly induced levels of (8-OHdG) that are excreted in the urine over the time course of the study compared to control urine samples. Permethrin did not cause significant increase in the amount of 8-OHdG in the urine. Levels of 8-OHdG in urine excreted at 24 h were 1009 +/- 342, 1701 +/- 321, 1140 +/- 316, and 1897 +/- 231 ng following treatment with ethanol, DEET, permethrin, and DEET + permethrin, respectively. The results indicate that dermal administration of DEET could generate free radical species hence cause DNA oxidative damage in rats.

11. Abu-Qare, A. W. and Abou-Donia, M. B. Binding of pyridostigmine bromide, N,N-diethyl-m-toluamide and permethrin, alone and in combinations, to human serum albumin. Archives of Toxicology. 2002; 76(4):203-208. ISSN: 0340-5761.

Abstract: In this study we examined the interaction of the anti-nerve agent drug pyridostigmine bromide (PB, 3,3-dimethylaminocarbonyloxy-N-methylpyridiniyum bromide), the insect repellent DEET (N,N-diethyl-m-toluamide), and the insecticide permethrin [3-(2,2-dichloroethyl)-2,2-dimethylcyclopropanecarboxyfic acid (3-phenoxyphenyl)methyl ester] in binding to human serum albumin (HSA). Concentrations between 500 ng/ ml and 10 mug/ml PB, DEET and permethrin, alone or in combination, were incubated with HSA at 37degreesC for 60 min. Concentrations of PB, DEFT and permethrin were determined using high performance liquid chromatography (HPLC). The results showed that 81.2 +/- 4.2%, and 84.6 +/- 2.5% of the initial concentration of PB was bound to HSA when incubated alone or in combination with DEET or permethrin, respectively. DEFT and permethrin did not significantly interact with HSA after I h of incubation. Incubation of combinations of two or three compounds did not significantly alter the binding pattern of any of the compounds with HSA. These results showed that PB is highly bound to albumin protein, while the competition between PB, DEET and permethrin on binding sites of HSA as a possible site of interaction following combined administration in vivo is not likely.

12. ---. Combined exposure to DEET (*N,N*-diethyl-*m*-toluamide) and permethrin- induced release of rat brain mitochondrial cytochrome c. Journal of Toxicology & Environmental Health. Part A. 2001; 63(4):243-252. ISSN: 1528-7394.
Abstract: The release of cytochrome c from the mitochondrial intermembrane space can induce apoptosis. The levels of mitochondrial cytochrome c in rat brain following a single dermal dose of 400 mg/kg of DEET, and of 1.3 mg/kg of permethrin, alone or in combination were determined. Rats were sacrificed at a time interval of 0.5, 1, 2, 4, 8, 16, 24, 48, or 72 h after dosing. Brain mitochondria were isolated and the levels of cytochrome c were measured using reversed-phase high-performance liquid chromatography (HPLC) with ultraviolet (UV) detection. Average percentage recovery of cytochrome c spiked with control rat brain mitochondria was 83.2 +/- 8.9%. Limits of detection and quantitation were 1 and 5 ng, respectively. The results showed that a single dermal dose of a combination of DEET and permethrin significantly increased the release of brain mitochondrial cytochrome c starting 24 h after treatment. DEET and permethrin alone did not affect the release of cytochrome c. The results indicate that combined exposure to DEET and permethrin might induce the apoptotic processes in rat brain as seen by the release of cytochrome c.

13. ---. DEET (N,N-diethyl-m-toluamide) alone and in combination with permethrin increased urinary excretion of 6beta-hydroxycortisol in rats, a marker of hepatic CYP3A induction. Journal of Toxicology & Environmental Health. Part A. 2001; 64(5):373-384. ISSN: 1528-7394.
Abstract: In this study, the ratio of 6beta-hydroxycortisol (6beta-OHF) to free cortisol (F) was determined in urine following a single dermal dose of 400 mg/kg of DEET (N,N-diethyl-m-toluamide), and 1.3 mg/kg of permethrin, alone and in combination, in rats. Urine samples were collected at 2, 4, 8, 16, 24, 48, and 72 h after application. Recoveries of 6beta-OHF and cortisol (F) from control urine samples were between 75 and 85%, with limits of detection at 30 and 10 ng/ml for cortisol and 6beta-OHF, respectively. A single dermal dose of DEET alone and in combination with permethrin significantly increased urinary excretion of 6beta-hydroxycortisol 24 h after dosing. Permethrin did not significantly alter the urinary excretion of 6beta- hydroxycortisol. These results indicate that DEET, alone and in combination with permethrin, increased urinary excretion of 6beta-OHF in rats following a single dermal dose application.

14. Abu-Qare, A. W.; Suliman, H. B., and Abou-Donia, M. B. Induction of urinary excretion of 3-nitrotyrosine, a marker of oxidative stress: following administration of pyridostigmine bromide, deet (n,n-diethyl-m-toluamide) and permethrin, alone and in combination in rats. Toxicology Letters. 2001; 121(2):127-134. ISSN: 0378-4274.
Abstract: In this study, we determined levels of 3-nitrotyrosine in rat urine following administration of a single oral dose of 13 mg/kg pyridostigmine bromide (P B) (3-dimethylaminocarbonyloxy-N-methylpyridinum bromide), a single dermal dose of 400 mg/kg N,N-diethyl-m-toluamide (DEET) and a single dermal dose of 1.3 mg/kg permethrin, alone and in combination. Urine samples were collected from Ave treated and five control rats at 4, 8, 16, 24, 48, and 72 h following dosing. Solid-phase extraction coupled with high-performance liquid chromatography with ultraviolet detection at 274 nm was used for the determination of tyrosine and 3-nitrotyrosine. A single oral dose of PB and

a single dermal dose of DEET or their combination significantly ($P < 0.05$) increased levels of 3-nitrotyrosine starting 24 h after dosing compared with control urine samples. The maximum increase of 3-nitroytyrosine was detected 48 h after combined administration of PB and DEET. The ratio of 3-nitrotyrosine to tyrosine in urine excreted 48 h after dosing was 0.19 +/- 0.04, 0.20 +/- 0.05, 0.28 +/- 0.03, 0.32 +/- 0.04, 0.19 +/- 0.05, 0.42 +/- 0.04, 0.27 +/- 0.03, 0.36 +/- 0.04, and 0.48 +/- 0.04 following administration of water, ethanol, PB, DEET, permethrin, PB + DEET, PB + permethrin, DEET + permethrin, and PB + DEET + permethrin, respectively. The results indicate that an oral dose of PB and a dermal administration of DEET, alone and in combination, could generate free radical species, and thus increase levels of 3-nitrotyrosine in rat urine. Induction of 3-nitrotyrosine, a marker of oxidative stress, following exposure to these compounds could be significant in understanding the proposed enhanced toxicity following combined exposure to these compounds.

15. Aldridge, W. N. An assessment of the toxicological properties of pyrethroids and their neurotoxicity. Critical Reviews in Toxicology. 1990; 21(2):89-104.
Abstract: II. Pyrethroids in biological systems. Includes mode of action and toxicokinetics. III. Specific neurotoxicological problems. IV. Peripheral nerve damage. Includes functional deficit and increase in beta-glucuronidase and beta-galactosidase and axonal degeneration. V. Delayed neuropathy. VI. Skin effects in humans and experimental studies. Includes human experience and experimental studies. VII. Poisoning and possible treatment. Includes poisoning cases and treatment of poisoning. VIII. Possibilities for further research.

16. Anadon, A.; Diez, M. J.; Sierra, M.; Sanchez, J. A., and Teran, M. T. Microsomal enzyme induction by permethrin in rats. Veterinary & Human Toxicology. 1988; 30(4):309-312. ISSN: 0145-6296.
Abstract: The synthetic pyrethroid, permethrin, was evaluated for its ability to alter hepatic microsomal drug-metabolizing function. The influence of permethrin (25:75 cis-trans) on plasma antipyrine kinetics and gamma-glutamyl transpeptidase (gamma-GTP) activity were studied in rats. After 3 days of administration of 90 mg permethrin/kg/day, there was no significant change in the antipyrine half-life and the area under the curve, while the apparent volume of distribution and clearance were significantly increased. Treatment with 190 mg permethrin/kg/day for 3 days decreased antipyrine half-life and the area under the curve, and increased the apparent volume of distribution and the clearance significantly. The gamma-GTP activity was significantly increased within 21 days and 14 days after the start of permethrin administration, at doses of 90 and 190 mg permethrin/kg/day, respectively. The antipyrine kinetics results indicate that permethrin is capable of producing a dose-dependent marked enzyme-inducing effect.

17. Anadon, A.; Martinez-Larranaga, M. R.; Diaz, M. J., and Bringas, P. Toxicokinetics of permethrin in the rat. Toxicology & Applied Pharmacology. 1991; 110:1-8.
Abstract: The toxicokinetics of permethrin after single 460 mg/kg oral and 46 mg/kg intravenous doses were studied in male Sprague-Dawley rats. Serial blood samples after oral and intravenous dosage, and brain, medulla oblogata, sciatic nerve, and liver samples after oral administration were collected. Plasma, hypothalamus, cerebellum, frontal cortex, caudate putamen, hippocampus, medulla oblongata, sciatic nerve, and liver concentrations of permethrin qand its metabolites, m-phenoxybenzyl alcohol and m-phenoxybenzoic acid, were determined by high performance liquid chromatographic assay. The permethrin plasma profile could be adequately described by a two compartment open model. For permethrin, the elimination half-life (t½beta) and the mean residence time from plasma were 8.67 and 11.19 hr after iv and 12.37 and 17.77 hr after po administration. The total plasma clearance was not influenced by dose concentration or route and reached a value of 0.058 liter/hr. After a single oral dose, permethrin was absorbed slowly with a Tmax of 3.52 hr. The maximum plasma concentration was 49.46 ug/ml. The oral bioavailability of permethrin was found to be 60.69%. The plasma concentration-time data for permethrin and its metabolites as well as the tissue concentration-time data for permethrin and its metabolites after an oral dose of permethrin were found to fit a one-compartment open model. The elimination half-life (t½el) of permethrin was greater for the hippocamus, medulla oblongata, frontal cortex, and sciatic nerve (23.10, 22.36, 13.86, and 16.27 hr, respectively) than for plasma (t½beta, 12.37 hr). The maximum amounts of permethrin in cerebellum, hippocampus,caudate putamen, frontal cortex, hypothalamus, and sciatic nerve were about 1.5, 2, 2, 2.7, 4.8, and 7.5 times higher than in the plasma, respectively, indicating an

accumulation of perethroid by nervous tissue itself. Nervous tissue accumulation of permethrin was also reflected by the area under the concentration curve ratios of tissue/plasma (1.16, 3.71, 1.57, 4.27, 3.48, and 8.77, respectively). The metabolites of permethrin, m-phenoxybenzyl alcohol and m-phenoxybenzoic acid, were detected in plasma and in all selected tissues for 48 hr after dosing, suggesting that a combination of metabolism by the tissues and diffusion into it from the blood may be present.

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Abstract: Simultaneous exposure to DEFT and permethrin was recently proposed to be associated with the "Gulf War Syndrome." However, no studies have reported the percutaneous absorption of DEET and permethrin when applied simultaneously to the skin as a mixture, the relevant route of exposure in the Persian Gulf, The present study quantitates percutaneous absorption of DEET and permethrin after coadministration to rodent and pig skin in vitro, Dosing solutions were also prepared with either acetone. dimethyl sulfoxide (DMSO), or ethanol to compare vehicle effects on percutaneous absorption of permethrin and DEET, The influence of DEET on carbaryl absorption and dermal disposition was also assessed in pig studies to statistically demonstrate DEET effects in acetone or DMSO and different solvent concentrations, Topical application of permethrin + DEET resulted in absorption of DEET (1-20% dose), but no permethrin, Permethrin (1.2-1.7% dose) was detected only when mouse skin was dosed solely with permethrin, a finding suggesting that DEET decreased permethrin absorption, DEET also inhibited carbaryl absorption in acetone mixtures, but had no effect on DMSO mixtures, Irrespective of solvent, DEET did not enhance carbaryl penetration into skin, For DEETI absorption was greater in mouse skin (10.7-20.6% dose) than in rat skin (1.1-5.2% dose) and pig skin (2.8% dose), The extent of DEET absorption was greater with DMSO and acetone than with ethanol in rat and mouse skin. These studies support DEET, but not permethrin or carbaryl, as hating sufficient systemic exposure to potentially cause signs of toxicity when simultaneously applied with pesticides, Furthermore, these studies demonstrated that DEET does not necessarily enhance dermal absorption of all toxicants as was originally hypothesized.

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Abstract: The cause of the Gulf War Syndrome may be related to soldiers being exposed to insecticides (e.g., permethrin. (P)), insect repellents (e.g., N,N-diethyl-m-toluamide (DEET)), an organophosphate nerve agent simulant (e.g., diisopropyl fluorpohosphate (DFP)), and/or prophylactic treatment (e.g., pyridostigmine bromide (PB)) against potential nerve gas attacks. The purpose of this study was to assess the dermal disposition of [C-14]permethrin in ethanol or ethanol:water (3:2) in the isolated perfused porcine skin flap (IPPSF) model with simultaneous dermal exposure to DEET or DFP. These IPPSFs were also simultaneously perfused arterially with or without PB, DFP, or DFP + PB. The results indicated that DFP + PB significantly increased [C-14]permethrin absorption compared to controls (1.06% dose vs 0.14% dose). PB significantly increased [C-14]permethrin disposition in the stratum corneum (SC) in aqueous mixtures only (9.40 vs 3.35% dose), while topical DEET or topical DFP reduced [C-14]permethrin levels in the SC especially in nonaqueous mixtures. PB also significantly enhanced [C-14]permethdn penetration into all skin tissues and perfusate in aqueous mixtures, while DEET reversed this effect. PB appeared to influence [C-14]permethrin disposition in flowthrough diffusion cells, suggesting that the mechanism of this interaction may be associated predominantly with epidermal permeability, although muscarinic effects in the vasculture in IPPSFs should not be ruled out and requires further investigation. These experiments suggest that intraarterial perfusion of PB and/or DFP and topical application of DFP or DEET can alter the disposition of [C-14]permethrin in skin and possibly its bioavailability in soldiers simultaneously exposed to these chemicals.

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774.
Abstract: This study extends data suggesting potential immunotoxic effects of pyrethroid insecticides by defining for the first time specific cellular immune alterations in mice. Functions requiring specific antigen recognition and/or effector function (MLR, CTL, NK) were decreased while nonspecific mitogen stimulations and body and organ weights were not effected. There is a potential for alteration in immune function in mammalian systems.

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Abstract: ABSTRACT: The effects of permethrin, its cis and trans isomers, and deltamethrin on operant behavior and food intake were examined. Male Sprague-Dawley rats were trained to respond on a VI20 second schedule of food reinforcement. Rats were injected intraperitoneally with pyrethroids or their Emulphor vehicle 20 minutes prior to testing. Technical grade permethrin (15-60 mg/kg) produced a dose-related decrease in operant response rate. The 60 mg/kg dose decreased rates by 60%. Lower doses of cis-permethrin (30 mg/kg) and deltamethrin (2 mg/kg) also produced significant decreases in response rate. A 30 mg/kg dose of trans-permethrin was without effect. Food intake was also measured for 1.5 and 24 hr periods after tech-permethrin treatment. Food intake was decreased over both intervals by the 60 mg/kg dose. The results of these studies indicate that subconvulsive doses of pyrethroid insecticides can have significant effects on learned behavior and food intake.

NOTES: In this study rats were injected intraperitoneally with pyrethroids. Permethrin (15-60 mg/kg) did produce a dose-related decrease in operant response rate. Food intake was decreased over the 1.5 and 24 hour intervals with the 60 mg/kg dose. The results show that subconvulsive doses of pyrethroid insecticides has a significant effect on learned behavior and food intake. The pyrethroids persist in the field for about 30 days. In the mouse pyrethroids (permethrin) have produced hyperactvity, increased sensitivity to external stimuli and whole body tremor, ultimately leading to prostration and death.

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Abstract: A degeneration of the nigrostriatal pathway is a primary component of Parkinson's disease (PD), and we have investigated the actions of insecticides on this pathway. For in vivo exposures, C57BL/6 mice were treated three times over a 2-week period with heptachlor, the pyrethroids deltamethrin and permethrin, or chlorpyrifos. One day after the last treatment, we observed that heptachlor and the pyrethroids increased maximal [3H]dopamine uptake in striatal synaptosomes from treated mice, with dose-dependent changes in Vmax displaying a bell-shaped curve. Western blot analysis confirmed increased levels of dopamine transporter (DAT) protein in the striatum of mice treated with heptachlor and permethrin. In contrast, we observed a small, but statistically significant decrease in dopamine uptake by 100 mg/kg chlorpyrifos. For heptachlor, doses that upregulated DAT expression had little or no effect on serotonin transport. Permethrin did cause an upregulation of serotonin transport, but required a 30-fold greater dose than that effective on dopamine uptake. Other evidence of specificity was found in transmitter release assays, where heptachlor and deltamethrin released dopamine from striatal terminals with greater potency than other transmitter types. These findings confirm that insecticides possess specificity for effects on striatal dopaminergic neurotransmission.

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Abstract: A single case is described of congenital leukaemia with 11q23/MLL rearrangement in a preterm female newborn. Because of arachnophobia, the mother had heavily abused aerosolised permethrin, a widely used household insecticide. Permethrin is considered comparatively safe, but, in view of the mother's history, its potential to induce cleavage of the MLL gene in cell culture was tested. Incubation of the BV173 cell line with 50 muM permethrin readily induced MLL cleavage.

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Abstract: Although organophosphorus and pyrethroid pesticides are considered environmental contaminants, their estrogenic potentials are still ubiquitous and unclear. The present study was undertaken to evaluate the estrogenic activities of nine pesticides (phoxim, malathion, monocrotophos, dimethoate, opunal, fenvalerate, cypermethrin, permethrin, and deltamethrin) using three in vitro methods [E-Screen assay, estrogen receptor (ER) competitive binding assay, and pS2 expression assay]. All the pyrethroid pesticides tested induced MCF-7 cell proliferation significantly, while organophosphorus pesticides did not. The estrogenic potency were ranked as permethrin > fenvalerate > cypermethrin > deltamethrin. The proliferation induced by cypermethrin, permethrin, and deltamethrin was blocked by ICI 182.780, while fenvalerate only partly inhibited it. In addition, pyrethroid pesticides inhibited the binding of [3H]estradiol to ER, while the organophosphorus failed to do so. Fenvalerate, permethrin, and cypermethrin induced pS2 mRNA expression with varying potency, while there were no significant effects in deltamethrin-treated groups. Our findings provide evidence to support the idea that pyrethroid pesticides tested produce an ER-specific, agonist response. Fenvalerate induced MCF-7 cell proliferation by a mechanism not involving ER-mediated pathway. Organophosphorus pesticides tested showed no estrogenic potential. Compared with the pS2 expression assay, E-Screen was a more sensitive and useful assay for screening of the xenoestrogenic chemicals.

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Abstract: Recent data have demonstrated that the in vivo effects of low dosages of two pyrethroids, cismethrin and deltamethrin, can be differentiated. Two behavioral tests, locomotor activity and the acoustic startle response (ASR), were utilized to separate the behavioral actions of Type I and II pyrethroids using permethrin, RU11679, cypermethrin, RU26607, fenvalerate, cyfluthrin, flucythrinate, fluvalinate and p,p'-DDT. Dosage-effect functions for all compounds were determined for both figure-eight-maze activity and the ASR in the rat. All compounds were administered po in 1 ml/kg corn oil 1.5-3 hr prior to testing. All compounds produced dosage-dependent decreases in locomotor activity. The Type I compounds, permethrin and RU11679, along with p,p'-DDT, increased amplitude and had no effect on latency to onset of the ASR. In contrast, the Type II pyrethroids, cypermethrin, cyfluthrin, and flucythrinate, decreased amplitude and increased the latency to onset of the ASR. Fenvalerate increased the amplitude, had no effect on latency, but unlike the other compounds tested, increased ASR sensitization. Fluvalinate had no effect on any measure of the ASR. These data provide further evidence of the differences between the in vivo effects of low dosages of Type I and II pyrethroids, and extend the findings of our previous work to other representatives of the two classes of pyrethroids.

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Abstract: The acute administration of 1R,cis, alpha S-cypermethrin, deltamethrin fenvalerate and permethrin produced a dose-dependent lowering of the dose of pentylenetetrazol required to elicit a seizure in rats. The proconvulsant action of cypermethrin displayed stereospecificity in that the 1R, cis, alpha S isomer of cypermethrin was the most potent compound tested, while the non-insecticidal isomer, 1S,cis, alpha R-cypermethrin, was devoid of proconvulsant activity. Pretreatment of rats with PK 11195, an antagonist of the peripheral-type benzodiazepine binding site, elicited a complete reversal of the proconvulsant actions of both deltamethrin and permethrin. In contrast, pretreatment with phenytoin did not alter the pyrethroid-induced proconvulsant activity. These results suggest that the effects of pyrethroids on pentylenetetrazol seizure threshold are mediated via an interaction with peripheral-type benzodiazepine binding sites.

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Abstract: 1. Type I (permethrin and allethrin) or type II (cypermethrin and fenvalerate) pyrethroids caused 23-37% increases in the striatal content of the dopamine metabolite 3,4-dihydroxyphenylacetic acid (DOPAC). 2. Toxicity symptoms and increases in DOPAC were associated with higher brain concentrations for type I (2.6-5.8 micrograms/gm) than type II pyrethroids (0.4-0.6 micrograms/gm). 3. No specific difference in the interaction between type I and II pyrethroids and the striatal dopaminergic system were recognized.

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Abstract: Parkinson's disease (PD) is a progressive neurodegenerative disease affecting the nigrostriatal dopaminergic pathway. Several epidemiological studies have demonstrated an association between pesticide exposure and the incidence of PD. Studies from our laboratory and others have demonstrated that certain pesticides increase levels of the dopamine transporter (DAT), an integral component of dopaminergic neurotransmission and a gateway for dopaminergic neurotoxins. Here, we report that repeated exposure (3 injections over 2 weeks) of mice to two commonly used pyrethroid pesticides, deltamethrin (3 mg/kg) and permethrin (0.8 mg/kg), increases DAT-mediated dopamine uptake by 31 and 28%, respectively. Using cells stably expressing DAT, we determined that exposure (10 min) to deltamethrin and permethrin (1 nM-100 muM) had no effect on DAT-mediated dopamine uptake. Extending exposures to both pesticides for 30 min (10 muM) or 24 h (1, 5, and 10 muM) resulted in significant decrease in dopamine uptake. This reduction was not the result of competitive inhibition, loss of DAT protein, or cytotoxicity. However, there was an increase in DNA fragmentation, an index of apoptosis, in cells exhibiting reduced uptake at 30 min and 24 h. These data suggest that up-regulation of DAT by in vivo pyrethroid exposure is an indirect effect and that longer-term exposure of cells results in apoptosis. Since DAT can greatly affect the vulnerability of dopamine neurons to neurotoxicants, up-regulation of DAT by deltamethrin and permethrin may increase the susceptibility of dopamine neurons to toxic insult, which may provide insight into the association between pesticide exposure and PD.

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Abstract: Permethrin, a type I synthetic pyrethroid insecticide, was evaluated through assessment of the behavioral development of F1 progeny of mice. Groups each of 30 male and 30 female ICR (CD-1) mice, as F0-generation, were given 0, 4.9, 9.8, and 19.6 mg/kg/d permethrin by gavage for 4 weeks before mating. Behavioral endpoints of motor reflexes, motor coordination, and activity were evaluated in 171 progeny. Clinical signs of toxicity including salivation, hyperactivity, and liquid feces which attributed to permethrin were observed in the F0-mice treated with 9.8 and 19.6 mg/kg/d. Reduction of body weight became evident only during gestation and lactation periods for the middle and high dose groups. Significant differences in the development of reflexes, swimming ability, and open field activity were evident in the offspring for the 9.8 and 19.6 mg/kg/d dose groups compared to the control group. These results show that permethrin at dose levels of 9.8 and 19.6 mg/kg/d can induce a significant risk to the offspring following treatment of F0-mice before mating. The NOEL obtained in this study for the effects of permethrin on the development of the F1-progeny is 4.9 mg/kg/d. (c) 2006 Elsevier Inc. All rights reserved.

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Abstract: Synthetic pyrethroids are lipophilic insecticides whose biological activity seems to be

directly related to their chemical structure. In this investigation differences in cutaneous sensation were detected by human participants between synthetic pyrethroids with a cyano group in the (S)-configuration of the 3-phenoxybenzyl alcohol of their molecular structure (fenvalerate) and those that do not (permethrin). A strong relation was noted between insecticidal potency and degree of induced cutaneous sensation for the alpha-cyano and non-cyano pyrethroids, with a prominent difference between the two. No sensation was observed by any of the same participants on topical exposure to the inert ingredients of these agents. A linear correlation between concentration and degree of induced dysaethesia was observed for both pyrethroids. Regressing the cutaneous sensation on the common logarithm of concentration resulted in a regression equation of Y = 84.0 + 31.0X1 for fenvalerate and Y = 27.5 + 15.8X1 for permethrin. A highly efficacious therapeutic agent for pyrethroid exposure was noted to be dl-alpha tocopherol acetate. An impressive degree of inhibition of paraesthesia resulted from the topical application of vitamin E acetate, with a therapeutic index of almost 100%.

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Abstract: Pesticides have been considered potential chemical mutagens. In fact, some studies show that various agrochemical ingredients possess mutagenic properties inducing mutations, chromosomal alterations or DNA damage. Experimental evidence shows a marked correlation between mutagenicity and carcinogenicity and indicates that short-term mutagenicity tests are useful for predicting carcinogenicity. The present study on rat exposed to two pyrethroids, cypermethrin and permethrin, showed different lymphocyte DNA damage depending on the type of pyrethroid, the dose, and the period of treatment. Data obtained from comet assay showed that oral treatment with 150 mg/kg body weight/day of permethrin (corresponding to 1/10 of LD50) for 60 days, induced a significant increase in all comet parameters. No lymphocyte DNA damage was measured after treatment with 25 mg/kg body weight/day of cypermethrin (corresponding to 1/10 of LD50) for the same period. A higher dose of permethrin (300 mg/kg body weight/day), for a shorter period (22 days), did not induce lymphocyte DNA damage, while supplementation with 200 mg/kg of Vitamins E and C protected erythrocytes against plasma membrane lipids peroxidation. Moreover, treatment with Vitamins E and C maintained the activity of glutathione peroxidase, which was reduced in the presence of permethrin, and reduced the osmotic fragility, which had increased following permethrin treatment. (C) 2004 Elsevier Ireland Ltd. All rights reserved.

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Abstract: Many pesticides possess hormonal activity and have thus been classified as endocrine disruptors. Pyrethroids are commonly used insecticides worldwide, but little has been done to characterize their hormone agonist/antagonist potential. We tested four frequently encountered pyrethroids, fenvalerate, sumithrin, d-trans allethrin, and permethrin, for estrogen and progesterone agonist/antagonist activities using the Ishikawa Var-I human endometrial cancer cell line and the T47D human breast cancer cell line. Both cell lines produce alkaline phosphatase as an indicator of hormonal activity. Fenvalerate and sumithrin demonstrated significant estrogenicity; at concentrations of 10 &mgr;M, these compounds achieved maximal activities comparable to that of 10 nM 17alpha-ethynylestradiol in Ishikawa Var-I cells. None of the four compounds showed statistically significant estrogen antagonist activity or acted as progestins. However, fenvalerate and d-trans allethrin significantly antagonized the action of progesterone in T47D cells. Through these hormonal pathways, exposure to certain pyrethroids may contribute to reproductive dysfunction, developmental impairment, and cancer.

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Abstract: The synthetic pyrethroid derivatives permethrin and cyhalothrin are widely used insecticides that are considered to be relatively nontoxic to higher animals. However, a variety of toxic effects on mammals have been reported. We investigated the effect of these drugs on energy

coupling by mitochondria and on the activity of the individual respiratory complexes. Using isolated rat liver mitochondria, a concentration-dependent inhibition of glutamate and succinate sustained state 3 respiration was found for both compounds in the micromolar range. The effect of pyrethroids on the activities of the complexes I to V were assessed individually in submitochondrial particles (complex I) and in freeze-thawed mitochondria (complexes Il-V). Complex I (EC 1.6.5.3) was found to be the most sensitive link within the electron transport chain. Half-maximal inhibition was observed at 0.73 mu M permethrin and 0.57 mu M cyhalothrin, respectively, and exhibited sigmoidal inhibition kinetics. Complexes II, III, IV and V (EC 1.3.5.1, 1.10.2.2, 1.9.3.1, 3.6.1.34) were not significantly inhibited by up to 50 mu M of these drugs. Thus, our results reveal a mode of action of synthetic pyrethroid insecticides not previously reported.

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Abstract: The effects of permethrin on striatal dopaminergic biomarkers were assessed in this study. Retired breeder male C57 B1/6 mice were given an ip dose of permethrin (0.1-200 mg/kg) at 7-day intervals, over a 2-week period (Days 0, 7, and 14). Animals were then sacrificed 1 day (t = 1), 14 days (t = 14), or 28 days after the last treatment (t = 28). Dopamine transporter (DAT) protein as assayed by Western blotting was increased to 115% in the 0.8 mg/kg group over that of control mice at t = 1 (P < 0.05). At t = 14, this value increased to 140% of control, and declined slightly to 133% of control at t = 28. The mice given the 1.5 mg/kg dose displayed a significant increase in DAT protein only at t = 28, to 145% of controls. Thus, upregulation of the DAT at low doses of PM is variable 24 h after treatment, and seems to stabilize by t = 28. The threshold dose for increasing DAT expression in Western blots by t = 28 was 0.2 mg/kg permethrin. [H-3]GBR 12935, used to assay DAT binding, followed the same trend as that for the Western blotting data for 0.8 and 1.5 mg/kg doses of permethrin over the 4 weeks posttreatment. At 200 mg/kg permethrin, DAT protein was unchanged vs controls (t = 1), but had significantly increased by t = 14 and continued to increase at t = 28, suggesting that the reduced dopamine transport at this dose was due to nerve terminal stress and that recovery had occurred. The protein a-synuclein was also significantly induced at the 1.5 mg/kg dose at t = 1; however, unlike DAT up-regulation, this effect had declined to control values by t = 14. Maximal induction of a-synuclein protein occurred at a dose of 50 mg/kg permethrin. These data provide evidence that the pyrethroid class of insecticides can modulate the dopaminergic system at low doses, in a persistent manner, which may render neurons more vulnerable to toxicant injury. (C) 2003 Elsevier Inc. All rights reserved.

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Abstract: Estrogens, whether natural or synthetic, dearly influence reproductive development, senescence, and carcinogenesis. Pyrethroid insecticides are now the most widely used agents for indoor pest control, providing potential for human exposure. Using the MCF-7 human breast carcinoma cell line, we studied the estrogenic potential of several synthetic pyrethroid compounds in vitro using pS2 mRNA levels as the end point. We tested sumithrin, fenvalerate, d-trans allethrin, and permethrin. Nanomolar concentrations of either sumithrin or fenvalerate were sufficient to increase pS2 expression slightly above basal levels. At micromolar concentrations, these two pyrethroid compounds induced pS2 expression to levels comparable to those elicited by 10 nM 17 beta-estradiol (five-fold). The estrogenic activity of sumithrin was abolished with co-treatment with an antiestrogen (ICI 164,384), whereas estrogenic activity of fenvalerate was not significantly diminished with antiestrogen co-treatment. In addition, both sumithrin and fenvalerate were able to induce cell proliferation of MCF-7 cells in a dose-response fashion. Neither permethrin nor d-trans allethrin affected pS2 expression. Permethrin had a noticeable effect on cell proliferation at 100 mu M, whereas d-trans allethrin slightly induced MCF-7 cell proliferation at 10 mu M, but was toxic at higher concentrations. Overall, our studies imply that each pyrethroid compound is unique in its ability to influence several cellular pathways. These findings suggest that pyrethroids should be considered to be hormone disrupters, and their potential to affect endocrine function in humans and wildlife should be investigated.

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Abstract: Occupational exposures to pesticides may increase parental risk of infertility and adverse pregnancy outcomes such as spontaneous abortion, preterm delivery, and congenital anomalies. Less is known about residential use of pesticides and the risks they pose to reproduction and development. In the present study we evaluate environmentally relevant, low-dose exposures to agrochemicals and lawn-care pesticides for their direct effects on mouse preimplantation embryo development, a period corresponding to the first 5-7 days after human conception. Agents tested were those commonly used in the upper midwestern United States, including six herbicides [atrazine, dicamba, metolachlor, 2,4-dichlorophenoxyacetic acid (2,4-D)], pendimethalin, and mecoprop), three insecticides (chlorpyrifos, terbufos, and permethrin), two fungicides (chlorothalonil and mancozeb), a desiccant (diquat), and a fertilizer (ammonium nitrate). Groups of 20-25 embryos were incubated 96 hr in vitro with either individual chemicals or mixtures of chemicals simulating exposures encountered by handling pesticides, inhaling drift, or ingesting contaminated groundwater. Incubating embryos with individual pesticides increased the percentage of apoptosis (cell death) for 11 of 13 chemicals (p less than or equal to 0.05) and reduced development to blastocyst and mean cell number per embryo for 3 of 13 agents (p less than or equal to 0.05). Mixtures simulating preemergent herbicides, postemergent herbicides, and fungicides increased the percentage of apoptosis in exposed embryos (p less than or equal to 0.05). Mixtures simulating groundwater contaminants, insecticide formulation, and lawn-care herbicides reduced development to blastocyst and mean cell number per embryo (p less than or equal to 0.05). Our data demonstrate that pesticide-induced injury can occur very early in development, with a variety of agents, and at concentrations assumed to be without adverse health consequences for humans.

39. Grosman, N. and Diel, F. Influence of pyrethroids and piperonyl butoxide on the Ca (+)-ATPase activity of rat brain synaptosomes and leukocyte membranes. International Immunopharmacology. 2005; 5(2):263-270. ISSN: 1567-5769.
Abstract: Pyrethroids are widely used insecticides of low acute toxicity in mammals but the consequences of long-term exposure are of concern. Their insecticidal action is related to neurotoxicity and, in addition. there are indications of mammalian immunotoxicity. In order to clarify structure-activity relationships of the membrane interactions of pyrethroids, the present study compared the influence of selected pyrethroids, i.e. permethrin and the more water soluble esbiol (S-bioallethrin), both type I, and cyfluthrin, type II, on the Ca2+-ATPase activity of rat brain synaptosomes and peritoneal leukocyte membranes. The pyrethroids were tested alone as well as mixed with the enhancing substance piperonyl butoxide (PBO) at concentration ratios of 1:5 and 1:10. At the highest concentration tested, pennethrin (10 muM) alone inhibited the ATPase activity of leukocyte membranes by 20%, whereas the synaptosomes were affected less. Esbiol and cyfluthrin alone did not affect. either membrane preparation significantly, whereas PBO (50 muM) alone caused 10-15% inhibition. Mixtures of either pyrethroid with PBO inhibited the ATPase activity of both types of membranes (up to 40% inhibition) in a synergistic manner. which always tended to be supra-additive. With esbiol a true potentiation took place. The synergistic interaction between pyrethroid and PBO was most apparent with mixtures of a concentration ratio of 1:5. The ATPase activity of leukocyte membranes tended to be more susceptible to inhibition than that of synaptosomes. The results are in accordance with the assumption that the mammalian toxicity of pyrethroids can be ascribed to a general disturbance of cell membrane function in neuronal tissue. The results indicate that it may also be the case in the immune apparatus.

40. Hadnagy, W.; Seemayer, N. H.; Kuhn, K. H.; Leng, G., and Idel, H. Induction of mitotic cell division distrubances and mitotic arrest by pyrethroids in V79 cell cultures. Toxicology Letters. 1999; 107(1-3):81-87. ISSN: 0378-4274.
Abstract: Five pyrethroids (fenvalerate, deltamethrin, cypermethrin, permethrin, cyfluthrin) differing in their chemical purity were investigated on their cytotoxic effects, especially on their ability to induce mitotic cell division disturbances using Chinese hamster lung cells of line V79. The colony forming ability (CFA) resulted in distinct differences of the cytotoxic effect of the tested pyrethroids, whereby permethrin was found to be most toxic. With the exception of fenvalerate all tested

pyrethroids gave rise to inhibition of cell cycle progression as shown by G2/M-arrest of synchronized V79 cells by flow cytometry as well as by the increase of the mitotic index as evaluated by light microscopy. The mitotic arresting activity could be attributed to the occurrence of abnormal mitotic figures such as initial and full C-metaphases. The results however indicate, that pyrethroids per se do not contribute to the cytotoxic effects but that other factors such as chemical impurities, source as well as manufacturing process and isomer composition may be responsible for the observed cytotoxic effects.

41. Hassouna, I.; Wickert, H.; El-Elaimy, I.; Zimmermann, M., and Herdegen, T. Systemic application of pyrethroid insecticides evokes differential expression of C-FOS and C-JUN proteins in rat brain. NeuroToxicology. 1996; 17(2):415-431.
Abstract: Expression of the c-Fos and c-Jun transcription factor was investigated by immunocytochemistry in the thalamus, hypothalamus, hippocampus and cortex of adult rats following intraperitoneal application of proconvulsant doses of the pyrethroid insecticides, cypermethrin and permethrin. Pyrethroid insecticides are used world-wide and their uptake, e.g., by nutrition and inhalation evokes severe neurological symptoms in animals and humans, but their effects on neuronal gene expression has not been elucidated. Cypermethrin induced a strong expression of c-Fos and c-jun in all the thalamic nuclei, except the ventro-posterior complex and substantia nigra, and in all the hypothalamic nuclei. In general, the immunoreactivities (IR) persisted for 8 h on their maximal levels, and were still above control levels after 24 h in several thalamic and hypothalamic areas. c-Fos-IR was strongly increased in all cortical layers with a predominance in the superficial layers II-IV, whereas c-Jun-IR was only slightly increased. In the hippocampus, cypermethrin induced a weak expression of c-Fos, but not of c-Jun, in the dentate gyrus and CA-3 area. Permethrin that has a lower pharmacological potency, evoked a similar pattern of c-Fos and c-jun expression, however, intensity and persistence of the neuronal labeling were less pronounced. Our results demonstrate that the neurotoxic effects of pyrethroid insecticides comprise molecular genetic alterations in the brain such as early and lasting induction of Fos and Jun transcription factor proteins. These changes in the neuronal program are prominent in the hypothalamus and thalamus that are involved in the regulation of the autonomic and visceral nervous systems.

42. Heder, A. F. ; Hirsch-Ernst, K. I.; Bauer, D.; Kahl, G. F., and Desel, H. Induction of cytochrome P450 2B1 by pyrethroids in primary rat hepatocyte cultures. Biochemical Pharmacology. 2001; 62(1):71-79. ISSN: 0006-2952.
Abstract: Numerous xenobiotics are capable of inducing their own metabolism and by enzyme induction can also lead to enhanced biotransformation of other xenobiotics. In this project, we examined the influence of pyrethroids (permethrin, cypermethrin, and fenvalerate) on the expression and activity of the phenobarbital (PB)-inducible cytochrome P450 2B1 isoform (CYP2B1) in primary rat hepatocyte cultures. Incubation of hepatocyte cultures with pyrethroids resulted in a marked CYP2B1 induction. Among the tested pyrethroids, permethrin elicited the most pronounced induction of CYP2B1 mRNA, which exceeded maximal induction achieved by PB at concentrations approximately 10-fold higher. Furthermore, permethrin induced CYP3A1 mRNA expression, while the expression of the CYP1A1 isoform, which in vivo is not responsive to PB treatment, was not significantly affected by pyrethroids. Permethrin-dependent enhancement of CYP2B1 and CYP3A1 mRNA expression was repressed by the hepatotrophic cytokine epidermal growth factor, which is known to also inhibit PB-dependent induction of CYP2B1. Several metabolites of permethrin formed by hepatocytes (3-(2',2'-dichlorovinyl)-2,2-dimethylcyclopropanecarboxylic acid, 3-phenoxybenzyl alcohol, and 3-phenoxybenzoic acid) were ineffective in inducing CYP2B1 mRNA. Furthermore, permethrin stimulated the expression of the luciferase reporter gene under control of the CYP2B1 promoter (comprising the PB-responsive enhancer module) in transiently transfected primary hepatocyte cultures. Thus, permethrin-stimulated gene expression occurred on the transcriptional level. Taken together, these results indicate that the pyrethroid permethrin is a PB-like inducer. Due to its superior potency in induction, permethrin appears as a useful substance for mechanistic studies to elucidate the mechanism of enzyme induction by phenobarbital.

43. Hijzen, T. H. and Slangen, J. L. Effects of type I and type II pyrethroids on the startle response in rats. Toxicology Letters. 1988; 40:141-152.

Abstract: The effects of type I and type II pyrethroids on the acoustic startle response of male Wistar rats were studied in three experiments. Pyrethroids were administered p.o. in 0.2 ml corn oil. In Expt. 1, techn. cis-permethrin (0, 30, 60, 90 mg/kg) and techn. cis,trans-cypermethrin (0, 60, 90, 120 mg/kg) incresed startle in a dose dependent way, whereas overt behavior and habituation of the startle response were not affected. In Expt. 2, NAK 1901 (0, 1, 2.5, 4 mg/kg) enhanced startle amplitudes dose dependently and deltamethrin (0, 2, 4, 6 mg/kg) had no effect on startle. After both deltamethrin and NAK 1901 toxic signs increased dose-dependently but an effect on potentiation of the startle response was not found. In Expt. 3, deltamethrin (0, 2, 4, 6 mg/kg) was administered and the highest dose attenuated startle amplitudes significantly, whereas toxic signs increased dose-dependently. Deltamethrin had no effect on habituation of the startle response. It is concluded that type one pyrethroids increase the amplitude of the startle reflex; this effect does not depend on the presence of overt signs of toxicity. Type II pyrethroids have dissimilar effects on startle. Results are discussed suggesting that the effects of type II pyrethroids on startle amplitude are mediated by an indirect metabolic effect and a direct effect at the site of the muscle.

44. Hudson, P. M.; Tilson, H. A.; Chen, P. H., and Hong, J. S. Neurobehavioral effects of permethrin are associated with alterations in regional levels of biogenic amine metabolites and amino acid neurotransmitters. Neurotoxicology. 1986; 7(1):143-53. ISSN: 0161-813X.
Abstract: Oral administration of 120 or 240 mg/kg permethrin produced dose- and time-related tremor in rats with the peak effect occurring 5 hrs after dosing. Subsequent experiments done 5 hrs postdosing found that 45 to 180 mg/kg permethrin produced dose-related increases in rectal temperature and enhanced responsiveness to an acoustic stimulus. Tremor was detected at 90 and 180 mg/kg. Neurochemical analyses of regional biogenic amines and their metabolites and amino acids 5 hrs after 90 or 180 mg/kg indicated that 5-HIAA levels were increased in the hypothalamus (HYP), brain stem (BS), hippocampus (HPC), and striatum (STR); 5-HT was not affected. MHPG was increased in the HYP and BS, while NE was decreased at the high dose only. DOPAC and HVA were increased in the STR after 90 and 180 mg/kg, while DA was not affected. Aspartate levels were increased in the BS and STR; glutamate was increased in the BS. Taurine, glutamine, glycine, and GABA were not affected. A time-course analysis of neurochemical changes 2, 5, 12, and 24 hrs postdosing indicated that 5 hrs was the time of peak effect for permethrin. Permethrin-induced tremor and hyperthermia were significantly correlated with dose- and time-related changes in MHPG, 5-HIAA, and ASP.

45. Igarashi, A. ; Ohtsu, S.; Muroi, M., and Tanamoto, K. Effects of possible endocrine disrupting chemicals on bacterial component-induced activation of NF-kappaB. Biological & Pharmaceutical Bulletin. 2006; 29(10):2120-2122. ISSN: 0918-6158 (Print).
Abstract: Endocrine disrupting chemicals (EDCs) have a possibility to exacerbate infectious diseases because EDCs disturb the human immune system by interfering with endocrine balance. To assess the influence of EDCs on the innate immune function of macrophages, we investigated the effects of thirty-seven possible endocrine disruptors on lipopolysaccharide (LPS)- or bacterial lipopeptide (Pam(3)CSK(4))-induced activation of nuclear factor kappa B (NF-kappaB). Alachlor, benomyl, bisphenol A, carbaryl, kelthane, kepone, octachlorostyrene, pentachlorophenol, nonyl phenol, p-octylphenol and ziram inhibited both LPS- and Pam(3)CSK(4)-induced activation of NF-kappaB. Simazine inhibited only LPS-induced activation. A strong inhibitory effect was observed with ziram and benomyl. On the other hand, diethylhexyl adipate and 4-nitrotoluene tended to enhance the activation induced by Pam(3)CSK(4) and LPS, respectively. Aldicarb, amitrole, atrazine, benzophenone, butyl benzyl phthalate, 2,4-dichlorophenoxy acetic acid, dibutyl phthalate, 2,4-dichlorophenol, dicyclohexyl phthalate, diethylhexyl phthalate, diethyl phthalate, dihexyl phthalate, di-n-pentyl phthalate, dipropyl phthalate, malathion, methomyl, methoxychlor, metribuzin, nitrofen, permethrin, trifluralin, 2,4,5-trichlorophenoxyacetic acid and vinclozolin had no significant effects at 100 muM. These results indicate that some agrochemicals have the potential to inhibit macrophage function and suggest that endocrine disruptors may influence the development of bacterial infections.

46. Imamura, L.; Hasegawa, H.; Kurashina, K.; Hamanishi, A.; Tabuchi, A., and Tsuda, M. Repression of activity-dependent c-*fos* and brain-derived neurotrophic factor mRNA expression by pyrethroid

insecticides accompanying a decrease in Ca^{2+} influx into neurons. Journal of Pharmacology & Experimental Therapeutics. 2000; 295(3):1175-1182. ISSN: 0022-3565.
Abstract: Permethrin, a type I pyrethroid insecticide, is known to affect sodium channels of neurons and prolong sodium currents. On the other hand, the expression of brain-derived neurotrophic factor (BDNF) and c-fos genes is activated through Ca(2+) influx into neurons, in an activity- dependent manner. In this study, therefore, we investigated whether permethrin influenced the Ca(2+) signal-induced expression of these genes. In primary culture of mouse cerebellar granule cells (CGCs), stimulation with veratridine, a potent agonist for sodium channels, which causes membrane depolarization in neurons, induced c-fos and BDNF mRNA expression accompanying the Ca(2+) influx into neurons. Pretreatment with permethrin at doses nontoxic to CGCs repressed the induction of these genes dose dependently, with trans-permethrin more potent than cis-permethrin. Consistent with this, the increase in Ca(2+) influx caused by veratridine was repressed by permethrin. The membrane depolarization induced by elevating the potassium (K(+)) concentration in medium (high K(+)) caused the activation of c-fos and BDNF genes, which was also repressed by permethrin. Immunoblotting analysis of c-Fos and a gel-mobility assay of AP-1 DNA-binding activity supported the decrease in c-Fos synthesis in permethrin-treated CGCs. The type II pyrethroid cypermethrin also affected the expression of these genes but less effectively than permethrin. Thus, pyrethroids inhibit the activity-dependent gene expression in neurons.

47. Imamura, L.; Hasegawa, H.; Kurashina, K.; Matsuno, T., and Tsuda, M. Neonatal exposure of newborn mice to pyrethroid (*Permethrin)* represses activity-dependent c-*fos* mRNA expression in cerebellum. Archives of Toxicology. 2002; 76(7):392-397.
Abstract: In a previous report, we demonstrated that the exposure of cultured mouse cerebellar granule cells to permethrin, a type I pyrethroid insecticide, repressed the induction of activity-dependent c-fos and brain-derived neurotrophic factor (BDNF) gene expression, accompanying a decrease in Ca2+ influx into neurons. In addition, it has been suggested that some pyrethroids, including permethrin, are endocrine-modulating chemicals and accumulate in human breast milk. In this study, therefore, we investigated whether lactational exposure of newborn mice to permethrin influenced c-fos, BDNF and P-actin gene expression in the developing neonatal cerebellum. In the cerebella of control neonates, c-fos mRNA expression was characterized by a significant increase in postnatal weeks 2 and 3, followed by a marked decrease. In the cerebella of permethrin-treated neonates, the expression of c-fos mRNA was dose-dependently repressed by cis-permethrin more effectively than by trans-permethrin at postnatal week 3, without alterations in the body or cerebellum weights of neonates. In the fourth and fifth week, however, c-fos mRNA expression had decreased to the same level as that in the control and permethrin-treated neonates. A decrease in BDNF mRNA expression tended to be observed in the cerebella of newborn mice on exposure to permethrin. Thus, our results indicate that the activity-dependent gene expressions in cerebellar neuronal cells can be repressed by permethrin both in vitro and in vivo, and suggest that lactational exposure to pyrethroids might affect the postnatal development of the mammalian brain.

48. Imamura, L.; Kurashina, K.; Kawahira, T.; Omoteno, M., and Tsuda, M. Additional repression of activity-dependent c-*fos* and BDNF mRNA expression by lipophilic compounds accompanying a decrease in Ca^{2+} influx into neurons. Neurotoxicology. 2005; 26(1):17-25. ISSN: 0161-813X.
Abstract: Recently, it has been proposed that a variety of environmental disruptors (EDs) disturb the neonatal development of the brain in mammals because of their lipophilic characteristics. Therefore, the synergism of these lipophilic compounds is important when evaluating the risk from EDs. In mouse cerebellar granule cells (CGCs), the activity-dependent expression of the brain-derived neurotrophic factor (BDNF) gene is activated through an influx of calcium ions (Ca2+) into CGCs caused by membrane depolarization, which is involved in the activity-dependent development of not only the cerebellum but also other regions of the brain after birth. In our previous study, we reported that permethrin and some other pyrethroid insecticides, which are suspected of being EDs, repressed the induction of c-fos and BDNF mRNA expression, accompanying a reduction of Ca2+ influx at doses non-toxic to CGCs. In the present study, we investigated whether other lipophilic compounds influenced the Ca2+ signal-induced expression of both genes as permethrin did and, if so, whether these effects were synergistic or additional. Pretreatment with p,p'-DDT, diethylstilbestrol (DES) or bisphenol A dose-dependently repressed the induction of both genes as well as the increase in the

uptake of Ca2+ by CGCs. Simultaneous exposure of CGCs with permethrin, p,p'-DDT and DES, in addition, revealed an additional repression on the induction of the genes and the Ca2+ uptake. These results suggest that toxic effects of EDs might, at least additionally, occur in the brain even if the concentration of each compound is lower than the effective dose for humans.

49. Institoris, L.; Siroki, O.; Undeger, U.; Basaran, N., and Desi, I. Immunotoxicological investigation of subacute combined exposure by permethrin and the heavy metals arsenic(III) and mercury(II) in rats. Int Immunopharmacol. 2001; 1(5):925-933. ISSN: 1567-5769.
Abstract: Effects of combined 28 days of oral exposure to the insecticide Permethrin (Pe), alone or in combination with arsenic-III (As) or Hg-II (Hg), were investigated on certain toxicological (body weight, organ weights), haematological (white blood cell (WBC) and red blood cell (RBC) counts, haematocrit (Ht), mean cell volume (MCV), cell content of the femoral bone marrow) and immune function (IgM-PFC, delayed-type hypersensitivity (DTH) reaction) parameters of male Wistar rats. Immunotoxic (H = high) and NOEL (L = low) doses of the three substances were determined in preliminary experiments under identical experimental conditions. In the present study, the immunotoxic dose of Pe (126 mg/kg) was combined with the NOEL dose of As (3.33 mg/kg) or Hg (0.40 mg/kg), and the NOEL dose of Pe (12.6 mg/kg) with the immunotoxic dose of As (13.3 mg/kg) or Hg (3.20 mg/kg). A separate group of animals, treated with the appropriate high dose component only, was used as internal control. Significant interactions were observed in the liver weight of the animals treated with Pe(H)-As(L) or As(H)-Pe(L), in the cell content of the femoral bone marrow in case of Pe(H)-As(L) and Pe(H)-Hg(L) combinations, as well as in the number of PFCs formed from 10(6) spleen cells in the Pe(H)-As(L) and in the maximum of DTH reaction in the Hg(H)-Pe(L) combination. The results show that combined exposures by the investigated substances modify the toxic (including immunotoxic) effects of the single compounds. These findings rise the probability that the interactions observed can also be present in human situations altering the health hazard of this three chemicals.

50. Institoris, L.; Undeger, U.; Siroki, O.; Nehez, M., and Desi, I. Comparison of detection sensitivity of immuno- and genotoxicological effects of subacute cypermethrin and permethrin exposure in rats. Toxicology. 1999; 137(1):47-55. ISSN: 0300-483X.
Abstract: Immuno- and genotoxicological effects of a 28-day oral treatment by equitoxic (1/10, 1/25, 1/50 LD50) doses of cypermethrin (55.4, 22.2, and 11.1 mg/kg) and permethrin (125.7, 50.3, and 12.6 mg/kg) were compared on male Wistar rats. Humoral and cell-mediated immunity were investigated by PFC assay and delayed type hypersensitivity (DTH) reaction (footpad swelling assay), and the genotoxic effects were studied by structural and numerical chromosome aberrations in bone marrow cells. The experimental system also involved certain general toxicological (body weight gain, organ weights) and haematological [white blood cell (WBC), red blood cell (RBC), haematocrit (Ht) and cell content of the femoral bone marrow] investigations. Among the immune function assays, only DTH reaction decreased at the two higher cypermethrin (CY) doses. These doses also increased the number of numerical chromosome aberrations of the bone marrow cells but did not change the number of structural aberrations. All CY doses decreased the mean cell volume (MCV) of RBCs and the Ht value. The two higher doses also reduced the WBC count in the peripheral blood. Permethrin (PE), in the applied dose range, had no effect on the examined immune function parameters, but all three doses increased the number of numerical chromosome aberrations. A dose-dependent increase in the liver weight, decreased MCV value, and elevated cell content of the femoral bone marrow were also observed. Under these experimental conditions, examination of chromosome aberrations proved to be less sensitive in detection of exposure by cypermethrin than applied immune function assays did. Permethrin, on the contrary, increased the number of numeric aberrations at all dose levels but had no effect on the immune function parameters examined.

51. Ishmael, J. and Lithfield, M. H. Chronic toxicity and carcinogenic evaluation of permethrin in rats and mice. Fundamental & Applied Toxicology. 1988; 11(2):308-322. ISSN: 0272-0590.
Abstract: Groups of Alpk:AP (Wistar-derived) rats were fed diets containing 0, 500, 1000 or 2500 ppm permethrin for 2 years and Swiss-derived mice were maintained for their lifetime (80% mortality) on diets containing 0, 250, 1000, or 2500 ppm permethrin. Changes of toxicological significance were confined to the top dose level of 2500 ppm permethrin in both species. Tremors

and hypersensitivity to noise were noted in rats at this dose during the first 2 weeks of study but such signs were not seen in mice. Pathological examination of the central and peripheral nervous systems did not reveal abnormalities attributable to permethrin administration. The effect on mice at 2500 ppm permethrin was shown by decreased body weight gain. Liver hypertrophy, associated with increase in liver weight, microsomal enzyme activity, and proliferation of smooth endoplasmic reticulum occurred in the rat with similar but less marked changes in the mouse. This was considered to be an adaptive response of no toxicological significance. No evidence of a carcinogenic effect was seen in the rat study. In the mouse study a slight elevation in benign lung tumor incidence in males only at 2500 ppm permethrin was observed but was not considered to represent a carcinogenic effect.

52. Kakko, I.; Toimela, T., and Tahti, H. The synaptosomal membrane bound ATPase as a target for the neurotoxic effects of pyrethroids, permethrin and cypermethrin. Chemosphere. 2003; 51(6):475-80. ISSN: 0045-6535.
Abstract: Pyrethroids are used widely as insecticides both in agriculture and in households. A cellular target of pyrethroids is the sodium channel in the membrane. In the present study, the activity of the membrane bound integral protein ATPase was studied as a biomarker for the membrane effect of the pyrethroids permethrin and cypermethrin. Male Sprague-Dawley rats were used for cerebral synaptosome preparation. The isolation of synaptosomes was performed with the Percoll gradient method. Both total ATPase and Mg(2+) activated ATPase were studied by determining inorganic phosphate liberated from the substrate ATP. One hour exposure to permethrin (Biokill) and cypermethrin (Ripcord) insecticide products affected ATPase activities. The activity of Na(+), K(+) ATPase decreased dose-dependently in 10-50 microM concentrations of permethrin, and Mg(2+) activated ATPase increased over twofold in the same concentrations of the active components. The effect of the cypermethrin compound Ripcord was not clearly dose-dependent. The activity of total ATPase was almost entirely lost in the concentrations of 100 microM of permethrin and cypermethrin. The results support the idea that membrane ATPases are one target of the neurotoxic effect of pyrethroid compounds.

53. ---. The toxicity of pyrethroid compounds in neural cell cultures studied with total ATP, mitochondrial enzyme activity and microscopic photographing. Environmental Toxicology and Pharmacology. 2004; 15(2-3):95-102. ISSN: 1382-6689.
Abstract: Pyrethroids are important insecticides used largely because of their high activity as an insecticide and their low mammalian toxicity. Some studies have demonstrated that these products, especially compounds with an alpha-cyano group, show neurotoxic effects on the mammalian central nervous system (CNS). In this study, we investigate with different methods the cell toxic effects of commercial, chemically different pyrethroid compounds on neuronal cell line SH-SY5Y Natural pyrethrin and permethrin (both with no alpha-cyano group) and cypermethrin (with an alpha-cyano group), were studied. For toxicity determinations, SH-SY5Y neuroblastoma. cells were exposed to pyrethroids at 0.1-100 muM concentrations for 1 day. The cell toxicity was evaluated by determining the total ATP with a luminescence method, the mitochondrial metabolic activity (WST-test) with a photometric method, and the morphological changes of the cell cultures with microscopic digital photographing at different dose levels of compounds. The results obtained with WST-1 method and with the measurement of total ATP were different. ATP measurement seemed to show cytotoxicity at lower concentrations than WST-1 method. There was induction of enzyme activities with WST-1 test with all pyrethroid compounds studied at low concentrations. With the ATP assay, exposure to 0.1-100 muM of natural pyrethrin, as well as of permethrin and cypermethrin showed dose-dependent cytotoxicity. The most toxic pyrethroid was cypermethrin followed by permethrin and natural pyrethrin. Our study confirms that the cell toxicity was dependent on the chemical structure of pyrethroids and pyrethroids without an alpha-cyano group show the weakest physiological effect. Microscopic photographs of exposed cell cultures correlated to the toxic effects revealed by the metabolic tests. (C) 2003 Elsevier B.V. All rights reserved.

54. Karen, D. J. ; Li, W.; Harp, P. R.; Gillette, J. S., and Bloomquis, J. R. Striatal dopaminergic pathways as a target for the insecticides permethrin and chlorpyrifos. Neurotoxicology. 2001; 22(6):811-817. ISSN: 0161-813X.

Abstract: Because insecticide exposure has been linked to both Parkinsons disease and Gulf War illness, the neurotoxic actions of pyrethroid and organophosphate insecticides on behavior and striatal dopaminergic pathways were investigated in C57BL/6 mice treated with permethrin (three i.p. doses at 0.2-200 mg/kg) or chlorpyrifos (three s.c. doses at 25-100 mg/kg) over a 2-week period. Permethrin altered maximal [3H]dopamine uptake in striatal synaptosomes from treated mice, with changes in Vmax displaying a bell-shaped curve. Uptake was increased to 134% of control at a dose of 1.5 mg/kg. At higher doses of PM (25 mg/kg), dopamine uptake declined to a level significantly below that of control (50% of control at 200 mg/kg, $P < 0.01$). We also observed a small, but statistically significant decrease in [3H]dopamine uptake by chlorpyrifos, when given at a dose of 100 mg/kg. There was no significant effect on the Km for dopamine transport. Evidence of cell stress was observed in measures of mitochondrialfunction, which were reduced in mice given high-end doses of chlorpyrifos and permethrin. Although cytotoxicity was not reflected in decreased levels of striatal dopamine in either 200 mg/kg PM or 100 mg/kg CPF treatment groups, an increase in dopamine turnover at 100 mg/kg CPF was indicated by a significant increase in titers of the dopamine metabolite, 3,4-dihydroxyphenylacetic acid. Both permethrin and chlorpyrifos caused a decrease in open field behavior at the highest doses tested. Although frank Parkinsonism was not observed, these findings confirm that dopaminergic neurotransmission is affected by exposure to pyrethroid and organophosphorus insecticides, and may contribute to the overall spectrum of neurotoxicity caused by these compounds.

55. Kasat, K.; Go, V., and Pogo, B. G. Effects of pyrethroid insecticides and estrogen on WNT10B proto-oncogene expression. Environment International. 2002; 28(5):429-432. ISSN: 0160-4120.
Abstract: Breast cancer is a serious illness affecting approximately one in nine women in the United States. Although an actual cause for breast cancer is unknown, genetic and environmental factors have been associated with its onset. Elevated levels of estrogen and heightened expression of the WNT10B proto-oncogene have been implicated in the development of human malignant breast tumors because they enhance the proliferation of mammary tissue. Two pyrethroid insecticides, sumithrin and fenvalerate, have been shown to mimic estrogenic activity in MCF-7 human breast carcinoma cells by inducing pS2 expression whereas two other pyrethroids, permethrin and d-trans allethrin do not have the same capability. To investigate if estrogen and these four pyrethroid insecticides could affect the expression of a gene related to mammary gland development, WNT10B expression in pyrethroid-treated MCF-7 cells was examined. MCF-7 cells under normal growth conditions do not express WNT10B. Reverse-transcriptase polymerase chain reaction (RT-PCR), nested PCR and Southern hybridization were employed to detect WNT10B expression. As controls, cells were treated with either ethanol, corn oil, or Vista LPA solvent. When compared to the solvent-treated controls, sumithrin, fenvalerate and estrogen treated MCF-7 cells all had increased levels of WNT10B expression. The non-estrogenic pyrethroids, d-trans allethrin and permethrin, demonstrated a similar elevation of WNT10B expression at a lower concentration, but not at the higher concentration. The results suggest that pyrethroid insecticides and estrogen can enhance the expression of the WNT10B proto-oncogene. However, since both the estrogenic and non-estrogenic substances amplified Wnt10B expression, the mechanism likely involves multiple distinct pathways.

56. Kennedy, D.; Hurst, V.; Konradsdottir, E., and Einarson, A. Pregnancy outcome following exposure to permethrin and use of teratogen information. American Journal of Perinatology. 2005; 22(2):87-90. ISSN: 0735-1631.
Abstract: NIX is a 1% permethrin creme rinse used for the treatment of head lice. There are no studies regarding human exposure during pregnancy. The primary objective of this study was to examine the safety of permethrin exposure during pregnancy. The secondary objective was to examine how teratogen information is perceived and used by women who requested information regarding this product. Women who had called the Motherisk and MotherSafe Programs to inquire about exposure to permethrin during pregnancy were followed-up to ascertain the outcome of their pregnancies. These women were compared with another group who had not been exposed to any known teratogenic drugs. Women who decided not to use permethrin were administered an additional questionnaire. We enrolled 147 women and completed outcomes on 113 pregnancies of women who had used permethrin some time during their pregnancy. There were 106 live births, six spontaneous abortions, one therapeutic abortion, and one major malformation in the women who

used permethrin in the first trimester. The mean birthweight was 3540 +/- 492 g and the mean gestational age was 40 +/- 1 weeks. There were no statistically significant differences between the exposed and comparison groups in any of the pregnancy outcomes. Of the 34 women who chose not to use permethrin and who completed the additional questionaire, 18 (52%) did not use permethrin because they did not feel the information was sufficiently reassuring. The results of this study suggest that the use of permethrin products during pregnancy appears to be relatively safe because there was no increase in the rates of major malformations. We also found that some women will not use a product during pregnancy unless they can receive a 100% guarantee that it will not harm their baby.

57. Kim, I. Y.; Han, S. Y.; Kang, T. S.; Lee, B. M.; Choi, K. S.; Moon, H. J.; Kim, T. S.; Kang, I. H.; Kwack, S. J.; Moon, A.; Ahn, M. Y., and Kim, H. S. Pyrethroid insecticides, fenvalerate and permethrin, inhibit progesterone-induced alkaline phosphatase activity in T47D human breast cancer cells. Journal of Toxicology & Environmental Health-Part a-Current Issues. 2005; 68(23-24):2175-2186. ISSN: 1528-7394.
Abstract: Pyrethroid insecticides exhibited a weak estrogenic activity by stimulation of MCF-7 cell proliferation and induction of alkaline phosphatase (AlkP) enzyme activity in cultured Ishikawa cells. Previously it was reported that fenvalerate and permethrin significantly inhibited the 17 beta-estradiol-induced MCF-7 BUS cell proliferation. Although certain pyrethroid insecticides exert estrogenic or antiestrogenic activities, it is not clear whether pyrethroid insecticides act as progesterone agonists or antagonists. Therefore, the aim of this study was to evaluate the effects of fenvalerate and permethrin on AlkP activity as a progesterone-specific response in T47D cells. In the present study, the stimulation of AlkP activity was concentration dependent with addition of progesterone, and maximum activity was observed at concentration of 1 x 10(-8) M. Both fenvalerate (1 x 10(-6) M) and permethrin (1 x 10(-6) M) did not stimulate the AlkP activity, but progesterone (1 x 10(-8) M)-induced AlkP activity was significantly inhibited at 1 x 10(-6) M concentration of fenvalerate and permethrin, respectively. Progesterone receptor (PR) levels in cytosolic protein of T47D cells were studied to determine the relationship between cellular PR expression and AlkP activity. Similar to AlkP activity, progesterone (1 x 10(-8) M) significantly increased PR protein levels compared to control. However, PR protein levels were not affected in T47D cells cultured with fenvalerate and permethrin alone, whereas fenvalerate and permethrin significantly decreased progesterone-induced PR protein levels. Our data indicate that fenvalerate and permethrin exhibit antiprogestagenic activity in T47D human breast cancer cells.

58. Kim, I. Y.; Shin, J. H.; Kim, H. S.; Lee, S. J.; Kang, I. H.; Kim, T. S.; Moon, H. J.; Choi, K. S.; Moon, A., and Han, S. Y. Assessing estrogenic activity of pyrethroid insecticides using in vitro combination assays. Journal of Reproduction & Development. 2004; 50(2):245-255. ISSN: 0916-8818.
Abstract: Pyrethroid insecticides are among the most commonly used classes of insecticides worldwide, but their endocrine disrupting activities remain unclear. Therefore, in the present study, we examined the estrogenic activities of pyrethroid insecticides in E-screen and competition binding assays. In addition, we measured estrogen receptor (ER) protein and pS2 mRNA levels in human breast cancer cells (MCF-7 BUS) to clarify the mechanism of their estrogenicity. Seven pyrethroid insecticides (bioallethrine, cypermethrin, deltamethrin, fenvalerate, permethrin, sumithrin, and tetramethrin) were tested because of their worldwide usage. In addition, 17beta-estradiol was tested as a positive control. As expected, 17beta-estradiol significantly increased MCF-7 BUS cell proliferation at concentrations of 10(-11) M and above. Of the pyrethroid insecticides tested, only sumithrin increased MCF-7 BUS cell proliferation in a dose-dependent manner; the maximum induction of cell proliferation was observed at a dose of 10(-5) M. In the anti-estrogenic activity test, bioallethrin, fenvalerate, and permethrin significantly inhibited 17beta-estradiol-induced MCF-7 BUS cell proliferation at 10(-6) M, a concentration comparable to the effective dose (10(-9) M) of ICI 182,780, a pure ER antagonist. However, none of the pyrethroid insecticides competitively inhibited the binding of [H-3]estradiol to rat uterus ERs in competition binding assays. Both 17beta-estradiol (10(-10) M) and sumithrin (10(-5) M) decreased the levels of cytosolic ERalpha and ERbeta protein expression significantly as compared with the vehicle control. In addition, 17beta-estradiol (10(-10) M) increased pS2 mRNA expression markedly, and sumithrin significantly increased pS2 mRNA levels in a dose-dependent manner. The other six compounds tested in the

present study did not affect ER protein levels or pS2 mRNA levels. These results suggest that certain pyrethroid insecticides may be considered to be estrogen-like chemicals that act through pathways other than direct ER binding, and may function as endocrine modulators in both wildlife and humans.

59. Kim, S. S.; Lee, R. D.; Lim, K. J.; Kwack, S. J.; Rhee, G. S.; Seok, J. H.; Lee, G. S.; An, B. S.; Jeung, E. B., and Park, K. L. Potential estrogenic and antiandrogenic effects of permethrin in rats. Journal of Reproduction & Development. 2005; 51(2):201-210. ISSN: 0916-8818.
Abstract: Many environmental chemicals including pesticides have been reported to possess hormonal activities, and thus are classified as endocrine disruptors. Permethrin, a synthetic pyrethroid insecticide, is used worldwide, which provides potential environmental exposure. However, relatively few studies have reported on hormonal activities, particularly estrogenic and androgenic activities of permethrin, and the results of these studies are in some respects contradictory. Therefore, this study investigated the potential estrogenic and androgenic activities of permethrin in vitro and in vivo. We conducted an uterine Calbindin-D-9k (CaBP-9k) gene expression assay and an uterotrophic assay for estrogenic activity, and a Hershberger assay for androgenic activity. The CaBP9k gene, one of the intracellular calcium binding proteins, is estrogen-responsive in the uterus. The rat uterotrophic and Hershberger assays are generally used as in vivo short-term screening assays for detecting the estrogenic and androgenic activities of chemicals, although these assays are still being validated by the Organization for Economic Cooperation and Development (OECD). Northern blot analysis showed the induction of uterine CaBP-9k mRNA level in response to permethrin as well as co-administration of permethrin with E2. In the uterotrophic assay using 18-day-old female rats, subcutaneous treatments with permethrin (10 to 800 mg/kg) for three days increased relative uterine wet weights, and E2-induced uterine weights. These effects were statistically significant at 800 and 200 mg/kg, respectively. Moreover, permethrin-induced uterine weights were inhibited by the coadministration of ICI 182,780, an antiestrogen. In the Hershberger assay, the administration of permethrin orally to testosterone propionate-treated castrated male rats led to statistically significant reductions in androgen-dependent sex accessory tissue (ventral prostate, seminal vesicles, levator ani and bulbocavernosus muscles, Cowper's gland and glans penis) weights at all doses tested (10, 50 and 100 mg/kg). These results suggest that permethrin might have estrogen-like effects on female rats, but antiandrogen-like effects on males.

60. Kojima, M.; Fukunaga, K.; Sasaki, M.; Nakamura, M.; Tsuji, M., and Nishiyama, T. Evaluation of estrogenic activities of pesticides using an in vitro reporter gene assay. International Journal of Environmental Health Research. 2005; 15(4):271-280. ISSN: 0960-3123.
Abstract: The estrogenic activities of 32 pesticides in agricultural products were evaluated using the E-CALUX assay system developed by Xenobiotic Detection Systems Inc (North Carolina, USA). This system utilizes human ovarian carcinoma cells (BG1) stably transfected with an estrogen-responsive luciferase reporter gene plasmid. It was found that tolclofos-methyl, prothiofos, diazinon, Thiabenclazole (TBZ) and pyriproxyfen had estrogenic activity. Several pesticides are often present in agricultural products. Therefore the estrogenicity of the mixtures of two kinds of pesticides was evaluated. The activity of diazinon/tolclofos-methyl, pyriproxyfen/prothiofos and TBZ/o-phenylphenol (OPP) was increased up to 1.2-5.3 fold. On the other hand, chlorfluazuron, imazalil and chlorfenapyr had anti-estrogenic activity. Further, to evaluate the change in the estrogenic activity of pesticide metabolites, an experimental system was established using a rat S9 mixture. Metabolites of permethrin and OPP had no estrogenic activity, but they had weak activity after the metabolism. On the other hand, the metabolites of TBZ exhibited less estrogenic activity than the original compounds.

61. Kostka, G.; Palut, D.; Kopec-Szlezak, J., and Ludwicki, J. K. Early hepatic changes in rats induced by permethrin in comparison with DDT. Toxicology. 2000; 142(2):135-143.
Abstract: In this study permethrin [(3-phenoxyphenyl)-methyl-3-(2,2-dichloroethenyl)-2,2-dimethylcyclopropane carboxylate] and DDT [1,1-(2,2,2 trichloroethylidene)-bis-(4-chlorobenzene)] were compared in rats for their effects on early hepatic changes, proposed in the literature to be useful endpoints in screening for non-genotoxic hepatocarcinogenesis and/or liver tumour promotion. We compared the effects of both insecticides on the following endpoints: hepatomegaly,

mitogenesis (DNA synthesis, mitotic activity, percentage of binuclear cells) and liver pathology. Male Wistar rats received permethrin (PERM) or DDT in one, three, five and 14 daily oral doses (at 24-h intervals) equivalent to 1/10 LD50. Distinct differences in early liver response between PERM and DDT were observed. DDT stimulated the early effect consisting of hepatomegaly accompanied by an increase in hepatocellular proliferation with signs of cell necrosis. Thus, it might be concluded, that the mitogenic effect of DDT was at least partly related to a regenerative liver response. Although PERM significantly affected DNA synthesis and increased binuclear hepatocytes, this compound did not increase the number of mitotic figures. These results suggest that PERM may inhibit of phase G(2) in the cell cycle and consequently it may suppress the cell entering into the stage of mitosis (M-phase). In addition, the present findings provide evidence for the occurrence of abnormal mitoses in the hepatocytes of rats treated with DDT.

62. Kou, J. H.; Gillette, J. S., and Bloomquist, J. R. Neurotoxicity in murine striatal dopaminergic pathways following co-application of permethrin, chlorpyrifos, and MPTP. Pesticide Biochemistry & Physiology. 2006; 85(2):68-75. ISSN: 0048-3575.
Abstract: The neurotoxic action of permethrin and chlorpyrifos on striatal dopaminergic pathways was investigated in C57BL/6 mice. Technical permethrin (50/50 ratio of cis and trans isomers, 200 mg/kg) and/or chlorpyrifos (75 mg/kg) were administered three times over a two-week period, with 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP, 20 mg/kg) given on day one. Alterations in expression of a-synuclein, dopamine transporter (DAT), and tyrosine hydroxylase (TH) were analyzed at 1 or 28 days post-treatment. MPTP alone produced a long-lasting lesion in striatal dopaminergic pathways, with a depression of TH and DAT protein at both post-treatment times. Chlorpyrifos or permethrin alone had no effect on TH or DAT expression levels. No greater effect on protein expression was observed in mice treated with both MPTP and insecticides at I day post-treatment. However, by day 28 a significant reduction ($p < 0.05$) of TH and DAT was observed in the mice treated with MPTP, permethrin, and chlorpyrifos, compared with the mice given MPTP alone. Significant alteration ($p < 0.05$) of alpha-synuclein expression by MPTP (45% decrease) and permethrin (20% increase) occurred at I day post-treatment, but reverted to control levels by day 28. Parallel experiments with pure cis or trans isomers of permethrin (100 mg/kg), showed that each isomer caused about half the up-regulation of alpha-synuclein. These findings demonstrate that the co-application of pyrethroid or organophosphorus insecticides enhance the neurotoxicity of MPTP in C57BL/6 mice, and that a slowly developing neurotoxicity may occur after termination of high-dose exposure.

63. Kunimatsu, T.; Yamada, T.; Ose, K.; Sunami, O.; Kamita, Y.; Okuno, Y.; Seki, T., and Nakatsuka, I. Lack of (anti-) androgenic or estrogenic effects of three pyrethroids (esfenvalerate, fenvalerate, and permethrin) in the hershberger and uterotrophic assays . Regulatory Toxicology & Pharmacology. 2002; 35(2 Pt 1):227-237. ISSN: 0273-2300.
Abstract: Synthetic pyrethroids are among the most common pesticides and insecticides currently in use worldwide. Recently, chemicals classified as synthetic pyrethroids are suspected as being endocrine disrupting chemicals. However, no study has been conducted to assess their potential hormonal activities using in vivo test specifically focused on endocrine disruption. In the present study, we evaluated the interaction of three pyrethroids (esfenvalerate, fenvalerate, and permethrin) with androgen receptor (AR)- and estrogen receptor (ER)- mediated mechanisms using in vivo short-term assays. While internationally standardized protocols for the Hershberger and uterotrophic assays have not yet been fully developed, both are widely used and are being considered by OECD as short-term screening assays for hormonal activity. A 5-day Hershberger assay using castrated male rats measures agonistic and androgenic ability of the test chemicals to AR of several accessory glands/tissues (the ventral prostate, dorsolateral prostate, seminal vesicles with coagulating glands, and levator ani plus bulbocavernosus muscles). Esfenvalerate (5, 10, or 20 mg/kg/day), fenvalerate (20, 40, or 80 mg/kg/day), or permethrin (25, 50, or 75 mg/kg/day) was administered by oral gavage for 5 days to castrated male Crj:CD(SD)IGS rats (1 week after the castration, 11 weeks of age) with or without coadministration of 0.25 mg/kg/day testosterone propionate (subcutaneous injection on the dorsal surface). The highest dose levels tested for each chemical were considered the maximum level that could be used without causing excessive systemic toxicity. None of esfenvalerate, fenvalerate, and permethrin showed any androgenic or antiandrogenic effects. Reference control of

p,p'-DDE and methyltestosterone (100 mg/kg/day) provided significant effects in this assay protocol. Potential effects of these pyrethroids mediated through the ER were evaluated by means of 3-day uterotrophic assay using ovariectomized Crj:CD(SD)IGS rats (2 weeks after the ovariectomy, 8 weeks of age). No increase in weight of uterus (wet or blotted) was observed following oral exposure to esfenvalerate (5, 10, or 20 mg/kg/day), fenvalerate (20, 40, or 80 mg/kg/day), or permethrin (37.5, 75, or 150 mg/kg/day), respectively. Again, the highest dose levels tested for each chemical were considered the maximum level that could be used without causing excessive systemic toxicity. Reference controls consisting of ethynyl estradiol (0.03 mg/kg/day) and methoxychlor (125 mg/kg/day) both showed a significant effect in this assay protocol. It is concluded that, based on the results of these two reliable in vivo assays, none of esfenvalerate, fenvalerate, or permethrin exhibit any potential to cause adverse (anti-) androgenic or estrogenic effects at dose levels below that of those causing excessive systemic toxicity.

64. Lees, G. Effects of pyrethroid molecules on rat nerves in vitro: potential to reverse temperature-sensitive conduction block of demyelinated peripheral axons. British Journal of Pharmacology. 1998; 123(3):487-496. ISSN: 0007-1188.
Abstract: 1. Prolongation of action potentials by cooling or pharmacological treatment can restore conduction in demyelinated axons. We have assessed the ability of pyrethroids (in vitro) to modify action potential kinetics and to reverse conduction block in lesioned peripheral nerve. 2. Fast Na+ currents were isolated in mammalian neuroblastoma (NIE115). Pyrethroids (4 microM) concurrently slowed inactivation and produced a spectrum of pronounced tail currents: s-bioallethrin (duration 12.2+/-7 ms), permethrin (24.2+/-3 ms) and deltamethrin (2230+/-100 ms). 3. Deltamethrin (5 microM) effected a slowly developing depression of compound action potential (CAP) amplitude in peroneal nerve trunks ($P<0.05$). Permethrin produced no net effect on CAP amplitude, area or repolarization time. 4. s-Bioallethrin (5 microM) enhanced CAP area, time for 90% repolarization and induced regenerative activity in a subpopulation of axons. 5. Tibial nerve trunks were demyelinated by lysolecithin (2 micro1) injection: 6-14 days later, slowly-conducting axons in the CAP (and peri-axonal microelectrode recordings) were selectively blocked by warming to 37 degrees C. 6. At 37 degrees C, s-bioallethrin (45 min, 5 microM) produced much greater after-potentials in lesioned nerves than in uninjected controls: area ($P< 0.05$) and relative amplitude ratios ($P< 0.0001$) were significantly altered. 7. In 3 of 4 cells (single-unit recording), s-bioallethrin restored conduction through axons exhibiting temperature-dependent block by raising blocking temperature (by 1.5 to > 3 degrees C) and reducing refractory period. 8. s-Bioallethrin induced temperature-dependent regenerative activity only in a sub-population of axons even after prolonged superfusion (> 1 h). 9. It was concluded that pyrethroids differentially alter Na+ current kinetics and action potential kinetics. The effects of s-bioallethrin are consistent with reversal of conduction block by demyelinated axons but regenerative/ectopic firing even in normal cells is likely to underpin its acknowledged neurotoxic actions and severely limit the clinical potential of this and related molecules.

65. Lein, P. J. and Fryer, A. D. Organophosphorus insecticides induce airway hyperreactivity by decreasing neuronal M2 muscarinic receptor function independent of acetylcholinesterase inhibition. Toxicological Sciences. 2005; 83(1):166-176. ISSN: 1096-6080.
Abstract: We previously demonstrated that the organophosphorus (OP) insecticide chlorpyrifos potentiates vagally induced bronchoconstriction independent of acetylcholinesterase (AChE) inhibition by decreasing the function of neuronal M2 muscarinic receptors that normally inhibit acetylcholine release from parasympathetic nerves supplying airway smooth muscle. However, it has been reported that different OPs may not affect muscarinic receptors equally. To determine if the effects of chlorpyrifos on airway hyperreactivity can be generalized to other OPs, we tested whether parathion and diazinon also inhibit neuronal M2 receptor function resulting in airway hyperreactivity. In control animals, the M2 agonist pilocarpine inhibits vagally induced bronchoconstriction in a dose-related manner. Treatment of guinea pigs with either parathion (1-10 mg/kg, sc) or diazinon (0.75-75 mg/kg, sc) shifted pilocarpine dose-response curves significantly to the right, indicating loss of neuronal M2 receptor function. These OP treatments also significantly potentiated vagally induced bronchoconstriction. Treatments that did not decrease M2 receptor function (parathion at 0.1 mg/kg, sc, or the non-OP insecticide permethrin at 150 mg/kg, sc) also did

not cause airway hyperreactivity. None of the OP treatments altered bronchoconstriction induced by iv acetylcholine or methacholine in vagotomized guinea pigs, suggesting that OP-induced airway hyperreactivity is not due to altered function of muscarinic receptors on airway smooth muscle or to AChE inhibition. AChE assays of lung, blood, and brain confirmed that parathion and diazinon decreased M2 function at concentrations that did not inhibit AChE. These data suggest that multiple diethyl phosphorothionate OPs cause airway hyperreactivity via a common mechanism of M2 receptor dysfunction independent of AChE inhibition.

66. Li, A. A.; Mink, P. J.; Mcintosh, L. J.; Teta, M. J., and Finley, B. Evaluation of epidemiologic and animal data associating pesticides with Parkinson's disease. Journal of Occupational & Environmental Medicine. 2005; 47(10):1059-1087. ISSN: 1076-2752.
Abstract: Exposure to pesticides may be a risk factor for developing Parkinson's disease (PD). To evaluate the evidence regarding this association in the scientific literature, we examined both analytic epidemiologic studies Of PD cases in which exposure to pesticides was queried directly and whole-animal studies for PD-like effects after systemic pesticide exposure. Epidemiologic studies were considered according to study quality parameters, and results were found to be mixed and without consistent exposure-response or Pesticide-specific patterns. These epidemiologic studies were limited by a lack of detailed and validated pesticide exposure assessment. In animal studies, no pesticide has yet demonstrated the selective set of clinical and pathologic signs that characterize human PD, particularly at levels relevant to human populations. We conclude that the animal and epidemiologic data reviewed do not provide sufficient evidence to support a causal association between pesticide exposure and PD.

67. McCain, W. C.; Lee, R.; Johnson, M. S.; Whaley, J. E.; Ferguson, J. W.; Beall, P., and Leach, G. Acute oral toxicity study of pyridostigmine bromide, permethrin, and DEET in the laboratory rat. Journal of Toxicology & Environmental Health. 1997; 50(2):113-124.
Abstract: This study investigated the lethal interaction of pyridostigmine bromide (PB), permethrin, and DEET when given to adult male rats by gavage and was separated into two phases. Phase I determined the acute oral lethal dose-response relationship of each compound with the vehicle, propylene glycol. Phase II was divided into two portions: a dose-response study using probit units obtained from phase I [lethal dose (LD) 16, 30, 50, 70, and 84], and an interaction study that contained low levels (calculated LD16, additive LD32) of the two compounds while the concentration of the third compound was varied. Rats were fasted overnight, dosed, and observed for 14 d. A significant increase in lethality occurred when PB, permethrin, and DEET were given concurrently when compared to expected additive values. Furthermore, solutions containing PB and permethrin or PB and DEET also caused a significant increase in lethality when compared to expected additive valuer. This information suggests that lethality in this study was more than an additive effect.

68. McKillop, C. M.; Brock, J. A.; Oliver, G. J., and Rhodes, C. A quantitative assessment of pyrethroid-induced paraesthesia in the guinea-pig flank model. Toxicology Letters. 1987; 36 (1):1-7. ISSN: 0378-4274.
Abstract: Pyrethroids are known to induce cutaneous effects in man which are distinct from the classical irritation and vascular responses. These effects are characterised by transient facial burning and tingling sensations. The aetiology of this cutaneous effect is related to the ability of pyrethroids to produce trains of nerve impulses in afferent nerves by prolonging the opening of the neuronal sodium channel. The veratrum alkaloids which are structurally dissimilar to the pyrethroids are known to affect the sodium channel in a similar manner. Using the guinea-pig flank model which has been developed to study this cutaneous phenomenon we have constructed dose-response curves to three structurally related pyrethroids (permethrin, cypermethrin and deltamethrin) and to a mixture of veratrum alkaloids (veratrine). In addition we have examined the time course over which these chemicals elicit a response.

69. Meyer, D. A. and Shafer, T. J. Permethrin, but not deltamethrin, increases spontaneous glutamate release from hippocampal neurons in culture. Neurotoxicology. 2006; 27(4):594-603. ISSN: 0161-813X.
Abstract: Pyrethroid insecticide modulation of the voltage-gated sodium channel (VGSC) is proposed to underlie their effects on neuronal excitability. However, some in vitro evidence

indicates that target sites other than VGSCs could contribute to pyrethroid disruption of neuronal activity. VGSC-independent, pyrethroid-induced changes in neurotransmitter release were examined to investigate the possibility that target sites other than VGSCs contribute to pyrethroid effects. Using whole-cell patch clamp recordings, deltamethrin and permethrin effects on glutamate-mediated miniature excitatory postsynaptic currents (mEPSCs) from pyramidal neurons in mixed hippocampal cultures were examined. In the presence of the VGSC antagonist tetrodotoxin, the type I pyrethroid permethrin (10 mu M) increased the average frequency of mEPSCs from a basal level of 1.0 +/- 0.4 to 3.5 +/- 0.6 Hz, with peak frequency of 9.9 +/- 1.5 Hz (n = 6). Permethrin did not affect the distribution of current amplitudes, indicating that permethrin increased the probability of glutamate release at the presynaptic terminal without effects on postsynaptic responses. Removal of calcium from the extracellular solution following the induction of the permethrin-mediated effect decreased mEPSC frequency (6.8 +/- 1.8 Hz, n = 3) to near control levels (1.9 +/- 0.8 Hz for control versus 2.5 +/- 0.6 Hz for permethrin minus Ca2+, respectively). However, the N- and P/Q-type voltage-gated calcium channel antagonist omega-conotoxin MVIIC had no effect on the permethrin-dependent increase in mEPSC frequency. In contrast to permethrin, the type 11 pyrethroid deltamethrin (10 mu M) failed to affect mEPSC frequency. These results indicate that permethrin causes a calcium-dependent increase in glutamate release from hippocampal neurons that is independent of effects on voltage-gated sodium or N- or P/Q-type voltage-gated calcium channels. The data indicate that permethrin increases mEPSC frequency via an alteration in intracellular calcium dynamics at the presynaptic terminal. (c) 2006 Elsevier Inc. All rights reserved.

70. Meyer, K. J. ; Reif, J. S.; Veeramachaneni, D. N. R.; Luben, T. J.; Mosley, B. S., and Nuckols, J. R. Agricultural pesticide use and hypospadias in eastern Arkansas. Environmental Health Perspectives. 2006; 114(10):1589-1595. ISSN: 0091-6765.
Abstract: INTRODUCTION: We assessed the relationship between hypospadias and proximity to agricultural pesticide applications using a GIS-based exposure method. METHODS: We obtained information for 354 cases of hypospadias born between 1998 and 2002 in eastern Arkansas; 727 controls were selected from birth certificates. We classified exposure on pounds of pesticides (estimated by crop type) applied or persisting within 500 to of each subject's home during gestational weeks 6 to 16. We restricted our analyses to 38 pesticides with some evidence of reproductive, developmental, estrogenic, and/or antiandrogenic effects. We estimated timing of pesticide applications using crop phenology and published records. RESULTS: Gestational age at birth [odds ratio (OR) = 0.91; 95% confidence interval (CI), 0.83-0.99], parity (OR = 0.79; 95% CI, 0.65-0.95), and delaying prenatal care until the third trimester (OR = 4.04; 95% CI, 1.46-11.23) were significantly associated with hypospadias. Risk of hypospadias increased by 8% for every 0.05-pound increase in estimated exposure to diclofiop-methyl use (OR = 1.08; 95% CI, 1.01-1-15). Pesticide applications in aggregate (OR = 0.82; 95% CI, 0.70-0.96) and applications of alachlor (OR = 0.56; 95% CI, 0.35-0.89) and permethrin (OR = 0.37; 95% CI, 0.16-0.86) were negatively associated with hypospadias. CONCLUSIONS: Except for diclofop-methyl, we did not find evidence that estimated exposure to pesticides known to have reproductive, developmental, or endocrine-disrupting effects increases risk of hypospadias. Further research on the potential effects of exposure to diclofop-methyl is recommended.

[pesticides applied in the study area: (from Table 1):
pyrethroids: bifenthrin, permethrin, cypermethrin, deltamethrin, esfenvalerate, zeta-cypermethrin;
carbamates: aldicarb, carbaryl, methyl parathion;
organophosphates: chlorpyrifos, malathion, dimethoate;
herbicides: bromoxynil, dicamba, diuron, cyanazine, simazine, alachlor, atrazine, trifluralin, 2,4-D, acetochlor, metribuzin, molinate;

diclofop-methyl, fenoxaprop, prometryn, propiconazole, quizalofop-ethyl, ciflubenzuron, iprodione, fipronil, pentachloronitrobenzene, pendimethalin,
trifluralin, metolachlor, (s)-metolachlor, carboxin, fenoxaprop, prometryn]

71. Mitchell, J. A.; Kallman, M. J., and Wilson, M. C. Behavioral responses to dermally applied pyrethroids. Toxicologist. 1986; 6(1):219.

Abstract: Locomotor-activity male Swiss mice were used to assess the behavioral effects of the pyrethroids fenvalerate and permethrin. Both significantly reduced the % of saccharine intake without affecting total fluid intake. Locomotor activity was increased by both as well.

72. Mitchell, J. A.; Wilson, M. C., and Kallman, M. J. Behavioral effects of Pydrin and Ambush in male mice. Neurotoxicology & Teratology. 1988; 10(2):113-119. ISSN: 0892-0362.
Abstract: Male Swiss mice, 20-25 g, were utilized to assess the effects of dermal and oral administration of the pyrethroid insecticide formulations Pydrin (30% fenvalerate) and Ambush (25.6% permethrin). Animals were subjected to a conditioned taste aversion procedure using a normally preferred 0.3% saccharin solution. Subjects were allowed 30 min access to a drinking syringe containing the saccharin solution, followed immediately by the administration of the pyrethroid or control solution. Pydrin (0.3, 3.0, or 30 mg/kg orally; 60, 600, or 1800 mg/kg dermally) and Ambush (0.5, 5.0, or 50 mg/kg orally; 30, or 300 mg/kg dermally) produced significant (p less than 0.05) reductions in the percent saccharin consumed. Total fluid intake, however, was not significantly altered by any of the treatments. The effect of the insecticides on both grouped and individual activity was also assessed in 20-25 g male Swiss mice. Activity measurements were taken over the 4-hr time period immediately following the administration of the pyrethroid or control solution. Pydrin (30 mg/kg orally; 600 and 1800 mg/kg dermally) and Ambush (50 mg/kg orally; 300 mg/kg dermally) significantly (p less than 0.05) increased activity in both grouped and individually tested mice. When subjects were individually tested, significant increases were seen in non-ambulatory, but not in ambulatory activity. The results of this work indicate that administration of the commercially available preparations of Pydrin and Ambush in mice at doses that do not induce the tremor and choreoathetosis-salivation syndromes usually associated with pyrethroid insecticides may result in behavioral changes.

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Abstract: Gulf War personnel were given pyridostigmine bromide (PB) as a prophylactic treatment against organophosphate nerve agent exposure, and were exposed to the insecticide permethrin and the insect repellent N,N-diethyl-m-toluamide (DEET). The purpose of this study was to assess the effects of PB to modulate release of inflammatory biomarkers after topical chemical exposure to chemical mixtures containing permethrin and DEET applied in ethanol or water vehicles. Treatments were topically applied to isolated perfused porcine skin flaps (IPPSFs). Concentrations of interleukin-8 (IL-8), tumor necrosis factor-alpha (TNF-alpha) and prostaglandin E-2 (PGE(2)) were assayed in perfusate to probe for potential inflammatory effects after complex mixture application. IPPSFs (n = 4/treatment) were topically dosed with mixtures of permethrin, DEET, and permethrin/DEET, in ethanol. Each treatment was repeated with perfusate spiked with 50 ng/ ml of PB. Perfusate was also spiked with 30 ng/ml diisopropylfluorophosphate to simulate low level organophosphate nerve agent exposure. Timed IPPSF venous effluent samples (0.5,1,2,4, and 8 h) were assayed by ELISA for IL-8 and TNF-alpha and by EIA for PGE(2). Overall, PB infusion caused a decrease or IL-8 and PGE(2) release. Effects on TNF-alpha were vehicle dependent. To probe the potential mechanism of this PB effect, human epidermal keratinocyte HEK cell cultures were exposed to permethrin DEET permethrin/DEET, with and without PB in DMSO. IL-8 was assayed at 1, 2, 4, 8, 12 and 24 h. PB suppressed IL-8 in permethrin and ethanol treatment from 4 to 24 h confirming the IPPSF results. In conclusion, these studies suggest that systemic exposure to PB suppressed IL-8 release at multiple time points in two skin model systems. This interaction merits further study.

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Abstract: Since pyrethroids are involved in reactive oxygen species production and no investigations have yet been performed on smooth muscle cell integrity, we studied the influence of permethrin- and cypermethrin-treatment on rabbit aorta using confocal laser scanning fluorescence microscopy, which allows cell viability to be assessed within the wall of living rabbit aorta. The data obtained show that the pyrethroid-treatment (10-100 mu M) impairs the smooth muscle cell viability. A

double-labeling protocol allowed us to distinguish cytotoxic effects of permethrin- and cypermethrin-treatment in aortic rings. In conclusion, permethrin seems to induce more oxidative stress on the aorta wall than that cypermethrin does.

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Abstract: Pyrethroids are divided into two groups according to their chemical structures: type I pyrethroids are devoid of a cyano moiety at the alpha-position (i.e. permethrin, PERM), while type II pyrethroids have an alpha-cyano moiety (i.e. cypermethrin, CY). Type I pyrethroids cause a type I poisoning syndrome or "T syndrome", whereas type II pyrethroids induce a type II choreoathetosis syndrome, known as "CS syndrome". The aim of the present work is to compare the effect of PERM and CY on erythrocyte plasma membrane fluidity of rats, treated orally for 60 days with low and high doses of these insecticides. The different modifications induced by pyrethroids on lipid peroxidation, osmotic fragility and the antioxidant enzymes (superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx)) were measured. The data obtained show that PERM, which proved more permeable than CY, produced an increase of fluidity and polarity in the hydrophilic-hydrophobic region of the erythrocyte bilayer even at low doses. Also at high doses, filtering through the membrane more easily, the PERM influenced more markedly the intracellular enzymatic activity, compared with CY, reducing GPx activity and increasing SOD activity. Because of its hydrophilic character, CY limits oxidative damage in the erythrocyte cytosol at high doses. (C) 2003 Elsevier Ireland Ltd. All rights reserved.

76. Nasuti, C.; Gabbianelli, R.; Falcioni, M. L.; Di Stefano, A.; Sozio, P., and Cantalamessa, F. Dopaminergic system modulation, behavioral changes, and oxidative stress after neonatal administration of pyrethroids. Toxicology . 2007; 229(3):194-205. ISSN: 0300-483X.
Abstract: Pyrethroids are a class of insecticides involved in different neurological disorders. They cross the blood-brain barrier and exert their effect on dopaminergic system, contributing to the burden of oxidative stress in Parkinson's disease through several pathways. The aim of the present study was to evaluate the effect of neonatal exposition to permethrin and cypermethrin (1/10 of DL50) in rats from the eighth to the fifteenth day of life. Open-field studies showed increased spontaneous locomotor activity in the groups treated with permethrin and the one treated with cypermethrin, while a higher number of center entries and time spent in the center was observed for the cypermethrin-treated group. Lower dopamine and higher homovanillic acid levels were measured in the striatum from both treated groups. A reduction of blood glutathione peroxidase content was measured, while no change in blood superoxide dismutase was observed. Carbonyl group formation increased in striatum, but not in erythrocytes. Lipid peroxidation occurred in erythrocytes, but not in striatum. No changes in fluidity at different depths of plasma membrane were measured in striatum or erythrocytes. The activation of monocyte NADPH oxidase by phorbol esters (PMA) shows that superoxide anion production was reduced in the pyrethroid-treated groups compared to the control group. Our studies suggest that neonatal exposition to permethrin or cypermethrin induces long-lasting effects after developmental exposure giving changes in open-field behaviors, striatal monoamine level, and increased oxidative stress. Although the action of pyrethroids on various target cells is different, a preferential interaction with the extracellular side of plasma membrane proteins can be observed. (c) 2006 Elsevier Ireland Ltd. All rights reserved.

77. Nation, P. N. and Roth, S. H. Complex effects of the insecticide permethrin on an isolated sensory neuron. Proc West Pharmacol Soc. 1987; 30:343-347. ISSN: 0083-8969.

78. Olgun, S.; Gogal, R. M.; Adeshina, F.; Choudhury, H., and Misra, H. P. Pesticide mixtures potentiate the toxicity in murine thymocytes. Toxicology. 2004; 196(3):181-195. ISSN: 0300-483X.
Abstract: The immunotoxic risks of multiple pesticide exposure were evaluated. C57BL/6 mouse thymocytes were exposed to lindane, malathion, and permethrin, either separately or in mixtures of two pesticides, in vitro. These pesticide exposures caused both apoptotic and necrotic cell death in thymocytes as evaluated by flow cytometric analysis in combination with 7-aminoactinomycin-D (7-AAD), Annexin-V/propidium iodide (PI) staining assays and lactate dehydrogenase release assays.

When cells exposed to mixtures of two pesticides, a significantly greater than additive interaction was observed in both apoptotic and necrotic populations of cells. The gel electrophoresis of DNA of cells showed DNA ladder formation with limited genomic DNA and increased laddering in mixture exposures. Based on these findings, it is suggested that these pesticides are potent immunotoxicants, in vitro, and that the mechanism of cytotoxicity observed upon exposure to these pesticides may, at least in part, be due to induction of apoptosis. We also provided evidence that induction of drug metabolizing mixed function oxidase system with lindane may, in part, be responsible for the potentiation of cytotoxicity in the combined exposures. As more information is obtained on the potential immunotoxic effects of pesticides, further insights will be gained for the risk assessment of these environmental pollutants.

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Abstract: The role of oxidative stress in immune cell toxicity caused by the pesticides lindane, malathion and permethrin was investigated in thymic cells from C57BL/6 mice. Thymocytes treated with any of these pesticides (concentrations ranging between 50-150 mu M) were found to generate both superoxide (O-center dot(2)-) and H2O2. The production of center dot O-2(-) was detected with hydroethidine-ethidium bromide assay. H2O2 production was monitored with a flow cytometric fluorescent (DCFH-DA) assay. All three pesticides stimulated center dot O-2(-) release after 5 min exposure. Lindane and permethrin, but not malathion, continued to have significant ($p <= 0.05$) effects on center dot O-2(-) generation following 15 min of exposure. The lindane + malathion mixture was found to cause more-than-additive increase in center dot O-2(-) production compared to individual pesticide treatments (at both 5 and 15 min). However, the effect of the lindane + permethrin mixture was not significantly different than individual components of this mixture. The effects of these pesticides on levels of antioxidant enzymes were also investigated, and only mixtures were found to have significant ($p <= 0.05$) effects. Thus, lindane + malathion and lindane + permethrin mixtures increased total superoxide dismutase (SOD) specific activity, had no effect on catalase levels and inhibited GSH-peroxidase and GSH-reductase specific activities. Although the results of these studies do not explain the mechanism of action of these pesticides on the generation of center dot O-2(-) and H2O2, it is worthy of note that mixtures of these chemicals have oxidative responses greater than those of single chemicals.

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Abstract: The study examined the interactions between a commercial formulation of methyl parathion (CF-MP) and a commercially formulated product of permethrin (CF-PMT) in male rats. The acute toxicity (LD50 values) and brain cholinesterase activity were investigated as toxicological endpoints. Results indicated that CF-MP modified the acute toxicity of CF-PMT. When animals were treated with a mixture, the addition of 380 mg/kg of CF-MP reduced the LD50 of CF-PMT by only 9.0%; however, when rats received CF-MP at 464 mg/kg, the LD50 of CF-PMT was reduced by 37% ($P < 0.001$). Also, CF-PMT decreased the CF-MP-induced inhibition of cholinesterase activity by 50% ($P < 0.05$). It was interesting to observe that xylene, which is the most abundant component in the vehicle of both formulations, had no effect on the CF-MP-induced inhibition of the cholinesterase activity. There was no relation between lethality and the inhibition of the brain cholinesterase activity in rats treated with mixtures containing CF-MP+CF-PMT or with either commercially formulated product alone. Considering the increased toxicity observed in rats treated with CF-PMT+CF-MP, it would be advisable to investigate further the interactions between both pesticides.

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relationships. Neurotoxicology & Teratology. 1987; 9:387-394.
Abstract: Pyrethroid insecticides have been divided into Types I and II based on behavioral profiles of toxicity produced by life-threatening dosages. In order to assess potential alterations in acquired (operant) behavior, acute dosage-effect and time-course determinations for permethrin (Type I) and cypermethrin (Type II) were made. Long-Evans rats responded for food according to a multiple schedule consicting of four different variable-interval schedules. Permethrin (100-400 mg/kg) and cypermethrin (7.5-60 mg/kg) were administered PO 1.5 hr pre-session and their effects on response rates and between-component response patterning were determined. Permethrin reduced responding in a manner which was independent of the baseline response rate, while the rate reductions following cypermethrin administration showed a dependence on the baseline levels of responding, with low response rates showing differential sensitivity to disruption. When select dosages of each compound were delivered at various presession times, onset of and recovery from the rate-decreasing effects were more rapid with cypermethrin, with rates returning to baseline levels by 12 hr post-dosing. Responding was maximally suppressed 24 hr after administration of permethrin and returned to baseline levels 48 hr after administration. The disruption of response patterning following cypermethrin was maximal at 1.5 hr after administration, with complete recovery 12 hr post-dosing. Differential effects on response patterning, in potency, and in the time-course of effects of permethrin and cypermethrin suggest a type-specificity for pyrethroid effects on schedule-controlled behavior at dosages far below those producing lethality in rats.

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Abstract: Epidemiological studies have linked insecticide exposure and Parkinson's disease. In addition, some insecticides produce damage or physiological disruption within the dopaminergic nigrostriatal pathway of non-humans. This study employed immunohistochemical analysis in striatum of the C57BL/6 mouse to clarify tissue changes suggested by previous pharmacological studies of the pyrethroid insecticide permethrin. Dopamine transporter, tyrosine hydroxylase, and glial fibrillary acidic protein immunoreactivities were examined in caudate-putamen to distinguish changes in amount of dopamine transporter immunoreactive protein from degeneration or other damage to dopaminergic neuropil. Weight-matched pairs of pesticide-treated and vehicle-control mice were dosed and sacrificed on the same days. Permethrin at 0.8, 1.5 and 3.0 mg/kg were the low doses and at 200 mg/kg the high dose. Brains from matched pairs of mice were processed on the same slides using the avidin-biotin technique. Four fields were morphometrically located in each of the serial sections of caudate-putamen, digitally photographed, and immunopositive image pixels were counted and compared between members of matched pairs of permethrin-treated and vehicle-control mice. For lowdoses, only 3.0 mg/kg produced a significant decrease in dopamine transporter immunostaining. The high dose of permethrin did not produce a significant change in dopamine transporter or tyrosine hydroxylase immunostaining, but resulted in a significant increase in glial fibrillary acidic protein immunostaining. These data suggest that a low dose of permethrin can reduce the amount of dopamine transporter immunoreactive protein in the caudate-putamen. They also suggest that previously reported reductions in dopamine uptake of striatal synaptosomes of high-dose mice may be due to nondegenerative tissue damage within this region as opposed to reductions of dopamine transporter protein or death of nigrostriatal terminals. These data provide further evidence that insecticides can affect the primary neurodegenerative substrate of Parkinson's disease.

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Abstract: Cutaneous exposure to the pyrethroid insecticide permethrin significantly suppresses contact hypersensitivity (CH) response to oxazolone in C57BL/6N mice. Additionally, cis-urocanic acid (cUCA), an endogenous cutaneous chromophore isomerized to its active form following exposure to ultraviolet radiation, modulates cell-mediated cutaneous immune responses. This study describes cutaneous immune alterations following combined topical permethrin and intradermal cUCA exposure. Female C57BL/6N mice were administered 5, 50 or 100 mug cUCA daily for 5

consecutive days. CH was then evaluated by the mouse ear swelling test (MEST) response to oxazolone. Decreased responses of 52.3%, 76.3% and 76.3%, respectively, as compared to controls were observed. Then, mice were co-exposed to 5 mug cUCA daily for 5 days and 1.5, 5, 15, or 25 muL permethrin, on either day 1, 3 or 5 of the cUCA treatment to evaluate combined immunomodulatory effects of the two chemicals, or cUCA daily for 5 days followed by permethrin on day 3, 5, or 7 after the last cUCA injection to demonstrate prolonged immunosuppressive effects. Two days after final treatment, mice were sensitized with oxazolone and MEST was performed. Mice receiving five cUCA injections and permethrin topically on cUCA injection day 1 showed up to 93.3% suppression of MEST compared to vehicle control. CH was suppressed by 87.5%, 86.6% and 74.2% in mice treated with 25 muL permethrin on days 3, 5 and 7 after cUCA, respectively, compared to vehicle control. Taken together, these data indicate co-exposure to cUCA and permethrin profoundly suppresses cell-mediated cutaneous immunity.

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Abstract: Background/Purpose: Cutaneous cis-urocanic acid (cUCA) or ultraviolet B exposure has been shown to cause diminished cutaneous contact hypersensitivity (CH) and to induce systemic tolerance (increased regulatory T lymphocytes) in mice. Permethrin is also a known CH inhibitor, but the molecular mechanisms are currently poorly understood. In this study, CH was evaluated in four strains of mice: an immunosensitive strain (C57BL/6N), an immunoresistant strain (SvImJ), a strain developed from C57BL/6N mice but genetically altered at both the tumor necrosis factor-alpha receptors (TNFalphap55R and p75R), and a strain developed from C57BL/6N but genetically deleted at the interferon-gamma (IFNgamma) locus. Methods: CH was evaluated in each group via oxazolone challenge following a 5-day exposure to intradermal (ID) cUCA or a single exposure to topical permethrin, or co-exposure to both chemicals in 5-week-old female C57BL/6N, SvImJ, and C57BL/6N mice genetically altered at the TNFalpha or IFNgamma locus. Results: A 5-day exposure to ID cUCA or a single exposure to topical permethrin resulted in diminished CH response in C57BL/6N mice, and this effect was exacerbated with concurrent exposure to both chemicals. CH in SvImJ was both cUCA- and permethrin-resistant relative to C57BL/6N mice, as 5-day cUCA or a single exposure to permethrin did not diminish CH, nor did concurrent exposure to cUCA and permethrin. Mice deleted at both TNFalphaR loci displayed similar but somewhat blunted diminished CH responses to cUCA or permethrin. This trend became significant with combined chemical exposure. IFNgamma knockout mice displayed similar diminished CH responses to cUCA or permethrin alone. Unlike C57BL/6N mice, the IFNgamma knockout mice did not show a further reduction in CH with combined chemical exposure. Conclusions: These results suggest the following: (1) Mouse strains show variable susceptibility to permethrin- and cUCA-induced immunomodulation. (2) TNFalpha may be involved in the immunomodulatory effects of cUCA and permethrin. (3) IFNgamma may be required for the more than additive depression of CH caused by cUCA+permethrin.

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Abstract: Immunomodulatory effects of a single topical permethrin exposure, 5-day exposure to cis -urocanic acid (cUCA), or a combination of the two chemicals were evaluated in 4- to 5-week-old female C57BL/6N mice. Permethrin alone decreased thymic weight and cellularity. Although cUCA alone did not affect thymic end points, coexposure to topical permethrin and cUCA exacerbated the thymolytic effects of permethrin. The single topical dose of permethrin also depressed several immune responses in isolated splenic leukocytes. This included splenic T-cell proliferative response to mitogen, splenic macrophage hydrogen peroxide production, and splenic B lymphocyte-specific antibody production. Unlike the effect of coexposure to these agents on thymic end points, cUCA did not exacerbate permethrin's adverse effect on any of the splenic end points examined. These results appear to suggest divergent mechanisms by which these compounds affect precursor and functionally mature T cells. At the doses used in this study, permethrin caused neurotoxic effects, including lethality, in a portion of the mice. For undetermined reasons, cUCA significantly increased

the rate of lethality caused by permethrin. Although the permethrin doses used in this study exceed that typically used in human medicine, these results raise some concerns about the possibility that sunlight, via cUCA, may increase the risk of adverse central nervous system and immune effects caused by permethrin alone.

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Abstract: Immunomodulatory effects of single topical exposure to permethrin were evaluated in 5-week-old female C57BL/6N mice. Mice exposed to 5-25 microl permethrin (equivalent to 220-1100 mg/kg body weight) on shaved interscapular skin were evaluated for altered body weight; splenic and thymic organ weight and cellularity; thymocyte cell surface expression, cellular apoptosis; splenic macrophage phagocytosis and hydrogen peroxide production; splenic B cell antibody production and T cell cytolytic activity; and mitogen-induced proliferation of splenocytes and thymocytes after in vivo or in vitro permethrin exposure. Topical permethrin application (25 microl) caused 32% inhibition of splenic T cell proliferation; in vitro exposure to permethrin also diminished splenocyte proliferation by 72% at 25 microM and 86% at 100 microM. permethrin did not appear to affect other leukocyte functional assays. Dose-related decreases in thymic cellularity of 52 and 80% were seen in mice exposed to 15 and 25 microl permethrin, respectively. Apoptosis was significantly increased in CD4(-)8(-) and CD4(-)8(+) thymocytes, and the CD4(+)CD8(+) thymocyte subpopulation was most severely diminished, suggesting possible chemical-induced apoptotic mechanism of thymic atrophy. Permethrin also caused splenic hypocellularity by 31% at 15 microl, and by 50% at 25 microl, an effect that may relate to inhibited proliferation or reduced seeding from the hypocellular thymus.

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Abstract: Permethrin was applied to the shaved dorsal interscapular region of C57Bl/6N mice at doses of 0.5, 1.5 or 5.0 mul/day. These doses corresponded to approximately 22-220 mg/kg/day topical insecticide. Mice were exposed to permethrin in this manner daily for 10 or 30 consecutive days, or every other day for 7 or 14 exposures. The splenic macrophage chemiluminescent response was depressed in a dose-dependent manner at 2 and 10 days post-exposure to permethrin. Phagocytic ability of macrophages was not inhibited. Antibody production as shown by plaque-forming cell (PFC) assay decreased significantly after 10 consecutive days of exposure to permethrin, These data indicate that topical permethrin exposure may produce systemic immune effects.

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Abstract: The modulation of acetylcholinesterase (AChE) of rat brain by two pyrethroids--permethrin (PM) and cypermethrin (CPM)--was studied both in vivo and in vitro. PM inhibited AChE activity in all regions of the rat brain (cerebral cortex, cerebellum, corpora striata, brain-stem, hippocampus, and hypothalamus) at 4, 8, and 12 h after gastric intubation, whereas CPM elevated the enzyme activity in vivo. Substrate-dependent enzyme kinetic studies have shown that PM and CPM behave as mixed-type inhibitors, as evidenced by alterations in both Michaelis-Menten constant (Km) and maximal velocity (Vmax) values. This indicates that both PM and CPM and substrate acetylcholine interact at hydrophobic subsites and may be able to bind simultaneously to the enzyme.

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Abstract: To understand the significance of the inhibitory action of 1,1,1-trichloro-2,2-bis(p-chlorophenyl)ethane (DDT) and pyrethroid insecticides on calmodulin, a universal Ca2+ binding protein, a bovine heart phosphodiesterase-calmodulin system was studied. It was found that, at

concentrations of less than 10(-5) M, the inhibitory action of DDT of the phosphodiesterase was due entirely to its action on calmodulin alone. Cypermethrin was less potent than DDT, but it also affected only calmodulin. Permethrin was the most potent inhibitor affecting calmodulin and, to a lesser extent, phosphodiesterase. The inhibitory action of these insecticides on calmodulin raises a possibility that many unsuspected Ca2+-related systems are being affected by these insecticidal chemicals, as calmodulin is known to play vital roles in many biological reactions dependent upon Ca2+. These include modulation of phosphodiesterase, neurotransmitter release, adenylate cyclase, Ca2+-dependent protein kinase, myosine light chain kinase and various membrane phosphorylation systems.

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Abstract: Topical exposure to permethrin has often been implicated as a mitigating factor in the illnesses reported in Gulf War veterans. These studies were designed to assess the effect of co-exposure to low level sulfur mustard. JP-8 jet fuel, N,N-diethyl-m-toluamide (DEFT) and fabric occlusion on the percutaneous absorption and skin disposition of topically applied C-14-permethrin (40 mug/cm(2)) in the isolated perfused porcine skin flap (IPPSF) model. Extent of dermal absorption in vehicle controls in the IPPSF was comparable to literature values for humans. These studies demonstrated a two-fold increased C-14-permethrin percutaneous absorption and almost three-fold increased penetration when JP-8 was present, compared to a one-third decreased permethrin flux in the presence of sulfur mustard. Complete occlusion slightly increased C-14-permethrin absorption. while occlusion with fabric showed no significant effect. A previously noted effect of DEFT to inhibit permethrin absorption was still seen in the presence of sulfur mustard exposure. These studies suggest that co-exposure to JP-8 or sulfur mustard may modulate transdermal flux of C-14-permethrin. However. the JP-8 increase in absorption and penetration was less than the five-fold increase previously seen with arterial infusion of pyridostigmine bromide and diisopropylfluorophosphate in the IPPSF. The toxicologic significance of this moderate increase in permethrin absorption remains unclear.

92. Rose, G. P. and Dewar, A. J. Intoxication with four synthetic pyrethroids fails to show any correlation between neuromuscular dysfunction and neurobiochemical abnormalities in rats. Archives of Toxicology. 1983; 53:297-316.
Abstract: The neurological effects of four synthetic pyrethroids resmethrin, permethrin, cypermethrin, and deltamethrin have been investigated in the rat to establish whether there is a correlation between the clinical-functional status of the animal and peripheral nerve damage as measured biochemically. Neuromuscular dysfunction was assessed by means of the inclined plane test and peripheral nerve damage by reference to beta-glucuronidase and beta-galactosidase activity increases in nerve tissue homogenates from treated and control animals. A transient functional impairment was found in animals treated with any one of the four pyrethroids tested and in all cases this was maximal at the end of the seven day subacute dosing regimen. Significant increases in beta-glucuronidase and beta-galactosidase were found 3-4 weeks after the start of dosing in the distal portion of the sciatic/posterior tibial nerves from permethrin, cypermethrin, and deltamethrin treated animal; but no changes were found in remesthrin-dosed animals. It is concluded therefore, that there is no direct correlation between the time-course of the neuromuscular dysfunction and the neurobiochemical changes. This suggests that these pyrethroids have at least two distinct actions - a short term pharmacological effect and at near-lethal dose levels a more chronic neurotoxic effect that results in sparse axonal nerve damage.

93. Sahib, I. K. ; Prasada Rao, K. S., and Desaiah, D. Pyrethroid inhibition of basal and calmodulin stimulated Ca2+ ATPase and adenylate cyclase in rat brain. Journal of Applied Toxicology. 1987; 7(2):75-80. ISSN: 0260-437X.
Abstract: Effects of two classes of pyrethroids, permethrin and resmethrin (type I), cypermethrin and deltamethrin (type II), on basal (calmodulin-deficient) and calmodulin stimulated activities of Ca2+ ATPase and adenylate cyclase from rat brain were studied in vitro. None of the pyrethroids inhibited synaptosomal basal Ca2+ ATPase, but permethrin and deltamethrin inhibited basal adenylate cyclase in the nuclear fraction of a brain homogenate. Both groups of pyrethroids decreased the calmodulin

activated Ca2+ ATPase and adenylate cyclase from brain synaptosomes and nuclear fraction. The results indicate that calmodulin-stimulated Ca2+ ATPase is more sensitive to type II pyrethroids and pyrethroids are more effective on calmodulin stimulated enzymes than basal enzyme activities. Since calmodulin, adenylate cyclase and Ca2+ ATPase are known to participate in various brain processes, it is possible that pyrethroids alter neural transmission, however, additional in vivo work would be needed to confirm this possibility.

94. Saito, K.; Tomigahara, Y.; Ohe, N.; Isobe, N.; Nakatsuka, I., and Kaneko, H. Lack of significant estrogenic or antiestrogenic activity of pyrethroid insecticides in three in vitro assays based on classic estrogen receptor alpha-mediated mechanisms. Toxicological Sciences. 2000; 57(1):54-60. ISSN: 1096-6080.
Abstract: Estrogenic and antiestrogenic activity of pyrethroid insecticides (d-trans-allethrin, cypermethrin, empenthrin, fenvalerate, imiprothrin, permethrin, d-phenothrin and prallethrin) was evaluated using a suite of three in vitro assays based on classic human estrogen receptor alpha (hER alpha)-mediated mechanisms. A mammalian cell-based luciferase reporter gene assay was developed for examining effects on hER alpha-mediated gene activation. hER alpha-independent effects on the gene activation were examined using control cells with constitutive luciferase activation by a herpes simplex virus thymidine kinase (HSV-TK) promoter for determining appropriate dose levels of test chemicals. Moreover, the test chemical-dependent interaction between hER alpha and a coactivator (transcriptional intermediary factor 2: TIF2) was analyzed by a yeast two-hybrid method, competitive binding to hER alpha being assayed by a fluorescence polarization method. Significant ($p < 0.05$) positive effects of estrogenic substances (E2/estradiol, diethylstilbestrol, and p-nonylphenol) were detected in all assays. An antiestrogen, 4-hydroxytamoxifen, significantly inhibited E2-mediated transactivation and interaction between hER alpha and TIF2 through hER alpha binding ($p < 0.05$). However, none of the pyrethroids tested showed significant ($p < 0.05$) estrogenic or antiestrogenic effects (100 nM-10 microM), indicating that they do not impact on the classic hER alpha-mediated activation pathway in vitro.

95. Shafer, T. J.; Meyer, D. A., and Crofton, K. M. Developmental neurotoxicity of pyrethroid insecticides: Critical review and future research needs. Environmental Health Perspectives. 2005; 113(2):123-136. ISSN: 0091-6765.
Abstract: Pyrethroid insecticides have been used for more than 40 years and account for 25% of the worldwide insecticide market. Although their acute neurotoxicity to adults has been well characterized, information regarding the potential developmental neurotoxicity of this class of compounds is limited. There is a large age dependence to the acute toxicity of pyrethroids in which neonatal rats are at least an order of magnitude more sensitive than adults to two pyrethroids. There is no information on age-dependent toxicity for most pyrethroids. In the present review we examine the scientific data related to potential for age-dependent and developmental neurotoxicity of pyrethroids. As a basis for understanding this neurotoxicity, we discuss the heterogeneity and ontogeny of voltage-sensitive sodium channels, a primary neuronal target of pyrethroids. We also summarize 22 studies of the developmental neurotoxicity of pyrethroids and review the strengths and limitations of these studies. These studies examined numerous end points, with changes in motor activity and muscarinic acetylcholine receptor density the most common. Many of the developmental neurotoxicity studies suffer from inadequate study design, problematic statistical analyses, use of formulated products, and/or inadequate controls. These factors confound interpretation of results. To better understand the potential for developmental exposure to pyrethroids to cause neurotoxicity, additional, well-designed and well-executed developmental neurotoxicity studies are needed. These studies should employ state-of-the-science methods to promote a greater understanding of the mode of action of pyrethroids in the developing nervous system.

96. Spencer, F. and Berhane, Z. Uterine and fetal characteristics in rats following a post-implanational exposure to permethrin. Bulletin of Environmental Contamination & Toxicology. 1982; 29:84-88.
Abstract: The natural pyrethrins constitute a group of potent contact insecticides which are extracted from dried chrysanthemum flowers. Permethrin, one of the newly synthesized pyrethroid and an ester of chrysanthemic acid, is presently being widely used as a component in household and farm sprays. This extensive usage is attributable to its high insecticidal activity and low mammalian

toxicity. From a reproductive standpoint, pyrethrum displayed a strong teratogenicity and produced sterility when administered to chickens. The purpose of the present study was: 1. to determine the effects of the post-implantational exposure to permethrine on mammalian reproduction using certain aspects of placental biochemistry and intra-uterine development as endpoints; and 2. to assess the scope of permethrin's possible reproductive toxicity with rats as the mammalian model.

97. Stein, E. A. ; Washburn, M.; Walczak, C., and Bloom, A. S. Effects of pyrethroid insecticides on operant responding maintained by food. Neurotoxicology & Teratology. 1987; 9(1):27-31. ISSN: 0892-0362.
Abstract: The pyrethroids are potent insecticides with low concomitant mammalian lethality when compared with other major insecticides. While high doses can lead to hyperactivity, tremors, convulsion and death, low doses have not been as well studied. Since operant behavior can be a sensitive measure of CNS function, male Holtzman rats were trained on a VR25 schedule maintained by 45 mg food pellets. Rats were injected IP with one of four different technical grade pyrethroids: permethrin, allethrin, deltamethrin and fenvalerate. All agents were effective in reducing operant responding and did so in a dose-dependent manner at levels 10 to 100 times below their LD50 values. Time course studies indicated a relatively short duration of action for the Type I agents of less than 60 min for permethrin and 15 min for allethrin. Type II agents were generally effective for greater than 60 min. Results of these studies indicate that operant responding maintained by food is a sensitive measure of the behaviorally disruptive effects of subconvulsive doses of pyrethroids.

98. Stelzer, K. J. and Gordon, M. A. Effects of pyrethroids on lymphocyte mitogenic responsiveness. Research Communications in Chemical Pathology & Pharmacology. 1984; 46(1).
Abstract: The in vitro effects pyrethroids on the mitogenic responsiveness of murine splenic lymphocytes to concanavalin A (Con A) and lipopolysaccharide (LPS) have been determined. Mitogenic stimulation was measured by uptake of [6-3H]-thymidine. Permethrin and cypermethrin inhibited the mitogenic response to Con A over a concentration range of 1x10-5 M to 5x10-5 M. Relative to these pyrethroids, slightly lower concentrations of fenpropathrin were required to inhibit mitogenic responsiveness to Con A. Fluvalinate was least effective of the pyrethroids tested, and caused mitogenic inhibitions at concentrations of 1.5x10-5 M to 5x10-5 M. Unlike the other pyrethroids, concentrations of fluvalinate as high as 1x10-4 M did not result in complete inhibition of the mitogenic response to Con A. Allethrin, was the most potent inhibitor, with effective concentrations in the range of 1x10-6 M to 1.5x10-5 M. Permethrin (3x10-5 M) inhibited the mitogenic response at each of several concentrations of Con A tested, and did not cause a shift in the lymphocyte mitogenic dose-response to Con A. The mitogenic responsiveness to LPS was found to be inhibited by allethrin and permethrin at the same concentrations which inhibited the mitogenesis induced by Con A. The results support the possibility of immune suppression by pyrethroid exposure.

99. Sumida, K.; Saito, K.; Ooe, N.; Isobe, N.; Kaneko, H., and Nakatsuka, I. Evaluation of *in vitro* methods for detecting the effects of various chemicals on the human progesterone receptor, with a focus on pyrethroid insecticides. Toxicology Letters. 2001; 118(3):147-155.
Abstract: The progesterone receptor (PR) is associated with physiological events such as implantation and the maintenance of pregnancy. Recently, it has become a social concern that chemicals may exert agonistic or antagonistic effects on hormone receptors. Therefore, we examined the effects of various chemicals on the human PR, with a focus on pyrethroid insecticides, using three in vitro methods. Eight pyrethroid insecticides (fenvalerate, d-allethrin, d-phenothrin, prallethrin, empenthrin, permethrin, cypermethrin and imiprothrin), examples of environmental pollutants and positive control chemicals were subjected to a reporter gene assay (luciferase assay) using human breast cancer T-47D cells, a two-hybrid assay and a binding assay using the same whole cells or receptors (cell-free). In none of these did the eight pyrethroid insecticides show any binding to the PR, agonistic or antagonistic effects.

100. Tessier, D. M. and Matsumura, F. Increased ErbB-2 tyrosine kinase activity, MAPK phosphorylation, and cell proliferation in the prostate cancer cell line LNCaP following treatment by select pesticides. Toxicological Sciences. 2001; 60(1):38-43. ISSN: 1096-6080.

Abstract: The oncogene erbB-2 codes for a receptor tyrosine kinase that functions as a key mitotic signal in a variety of cell types. Amplification or overexpression of erbB-2 occurs in many forms of cancer, such as of the breast, colon, and prostate, and is an indicator of poor prognosis in those diseases. In the human prostate cancer cell lines LNCaP and PC-3, erbB-2 kinase was activated by pesticides of different chemical classes: (1) the organochlorine insecticides beta-hexachlorocyclohexane (beta-HCH), o,p'-dichlorodiphenyltrichloroethane (o,p'-DDT), and heptachlor epoxide; (2) the pyrethroid insecticide trans-permethrin, and (3) the fungicide chlorothalonil. o,p'-DDT also causes phosphorylation of mitogen-activated protein kinase (MAPK) and cellular proliferation of the androgen-dependent LNCaP line. However, no proliferative effect was observed in the androgen-independent PC-3 line. The proliferative effect of o,p'-DDT in LNCaP could not be blocked by the androgen receptor antagonist p,p'- dichlorodiphenyldichloroethene (p,p'-DDE), indicating that this effect of o,p'-DDT does not occur through direct interaction with the androgen receptor. Together these data demonstrate a putative mechanism for the action of certain pesticides in hormonal carcinogenesis.

101. Tisch, M.; Schmezer, P.; Faulde, M.; Groh, A., and Maier, H. Genotoxicity studies on permethrin, DEET and diazinon in primary human nasal mucosal cells. Eur Arch Otorhinolaryngol . 2002; 259(3):150-153. ISSN: 0937-4477.
Abstract: Possible genotoxic effects exerted by three widely used pesticides, permethrin, N,N-diethyl-m-toluamide (DEET) and diazinon, in primary human nasal mucosal cells were investigated. Primary nasal mucosa cells were prepared from tissue biopsies taken from 21 patients who underwent nasal surgery. Cells were exposed to 0.5-1.0 mM concentrations of permethrin, DEET and diazinon for 60 min. Genotoxic effects were detected by the alkaline microgel electrophoresis assay ("comet assay"). Within the concentration range, no significant cytotoxic effects were observed, but all three tested pesticides showed a significant genotoxic response that was concentration dependent. More pronounced genotoxic effects were observed in mucosal cells from the middle turbinate than in the inferior turbinate.The results provide some evidence for the potential carcinogenicity of these agents to human nasal mucosal cells. This should be further investigated.

102. Tyler, C. R.; Beresford, N.; van der Woning, M.; Sumpter, J. P., and Thorpe, K. Metabolism and environmental degradation of pyrethroid insecticides produce compounds with endocrine activities. Environmental Toxicology & Chemistry. 2000; 19(4 Part 1):801-809.
Abstract: Pyrethroids are semisynthetic derivatives of the chrysanthemumic acids that have been developed as insecticides, and they are in widespread use. Considerable information is available regarding the toxicity, metabolism, and environmental degradation of pyrethroids, but almost nothing is known about their interactions with hormone receptors. In this study, seven commercial pyrethroids as well as products of metabolism and environmental degradation of permethrin were tested for steroid activity (both as agonist and as antagonist) in recombinant yeasts expressing the human estrogen and human androgen receptors. Pyrethroid insecticides had steroid receptor-binding activity. Fenpropathrin and permethrin both acted as weak estrogen agonists. Allethrin, bioallethrin, and expermethrin had antiestrogenic activity with potencies between 1,000-fold (bioallethrin) and 10,000-fold (allethrin) less than the established antiestrogen 4-OH-tamoxifen. Six of the seven pyrethroids tested had antiandrogenic activity (the most active, bioallethrin, was 70-fold less potent than flutamide). These activities, however, are believed to result either from contaminants/degradation products in the parent compounds or from metabolism of the parent compounds into active metabolites by the yeast. Three derivatives of permethrin all interacted with sex steroid hormone receptors, Three-phenoxybenzyl alcohol had both estrogenic and antiandrogenic activity, with potencies more than 100-fold greater than that of the parent compound, permethrin. Three-phenoxybenzoic acid and the cyclopropane acid derivative both had antiestrogenic activity, with approximately 100-fold and 1,000-fold lower potencies than 4-OH-tamoxifen, respectively. The data presented here highlight that an understanding of the metabolism and environmental degradation of chemicals is essential for assessing the potential of chemicals to have endocrine-modulating effects.

103. Undeger, U. and Basaran, N. Effects of pesticides on human peripheral lymphocytes in vitro: induction of DNA damage. Archives of Toxicology. 2005; 79(3):169-176. ISSN: 0340-5761.

Abstract: Because of the widespread use of pesticides for domestic and industrial applications the evaluation of their genotoxic effects is of major concern to public health. Although various experimental data have provided evidence that pesticides can possess genotoxic properties in animals and in in vitro test systems after acute and chronic exposure, the information on the genotoxic effects of some of pesticides is limited and inconsistent. In the present study, the genotoxic potential of commonly used pesticides (i.e., dimethoate and methyl parathion from the organophosphate class, propoxur and pirimicarb from carbamates, and cypermethrin and permethrin from pyrethroids) have been evaluated. The genotoxic effects of these substances were examined using the single cell gel electrophoresis (comet) assay in freshly isolated human peripheral lymphocytes. The cells were incubated with 10, 50, 100 and 200 mu g/ml concentrations of the test substances for 0.5h at 37 degrees C and DNA damage was compared with that obtained in lymphocytes from the same donor not treated with substances. Hydrogen peroxide, 100 mu M, was used as a positive control. Within the concentration ranges studied, no significant cytotoxic effects were observed. Dimethoate and methyl parathion at 100 and 200 mu g/ml; propoxur at 50, 100 and 200 mu g/ml, and pirimicarb, cypermethrin and permethrin at 200 mu g/ml significantly increased DNA damage in human lymphocytes.

104. US Army. Health effects of permethrin-impregnated army battle-dress uniforms. Ms. 1994:137 pp.
Abstract: US Army report on the effects of permethrin toxicity from military uniforms.

105. Vaccari, A. ; Ruiu, S.; Mocci, I., and Saba, P. Selected pyrethroid insecticides stimulate glutamate uptake in brain synaptic vesicles. Neuroreport. 1998; 9(15):3519-3523. ISSN: 0959-4965.
Abstract: We aimed to ascertain whether pyrethroid insecticides could influence the vesicular transport of the excitatory amino acid glutamate. The incubation of rat cortical synaptic vesicles with resmethrin and permethrin, consistently stimulated both ATP-dependent and -independent uptake of [3H]glutamate, while not evoking depletion of its vesicular content. Both processes were counteracted by valinomycin, a dissipator of the transmembrane potential gradient (deltapsi(sv)). Meanwhile, the vesicular influx of 36Cl- anions was impaired by pyrethroid concentrations which did not affect the ATP-dependent uptake of [14C]methylamine, as a marker for the proton gradient (deltapH). Thus, the stimulation of glutamate transport appeared to involve mainly the deltapsi(sv). A self-attenuating effect of selected pyrethroids on putatively enhanced excitatory transmission in severe intoxication is suggested.

106. van Haaren, F.; Haworth, S. C.; Bennett, S. M.; Cody, B. A.; Hoy, J. B.; Karlix, J. L., and Tebbett, I. R. The effects of pyridostigmine bromide, permethrin and deet alone, or in combination, on fixed-ratio and fixed-interval behavior in male and female rats. Pharmacology, Biochemistry & Behavior. 2001; 69(1-2):23-33. ISSN: 0091-3057.
Abstract: Concurrent exposure to pyridostigmine bromide (PB), permethrin (PERM) and/or N,N-diethyl-m-toluamide (DEET) may have contributed to the development of a syndrome that appears to have afflicted military personnel who served during the Gulf War. The present experiment sought to evaluate the behavioral effects of these compounds alone, or in various combinations, in male and female rats. Subjects were exposed to a multiple fixed-ratio (FR) 50, fixed-interval (FI) 2-min schedule of reinforcement. PB dose-dependently decreased FR and FI response rates. FR responding was disrupted by lower doses and there were no differences between the sexes. PERM vehicle administration decreased response rates maintained by both schedules of reinforcement; this was offset by an increase in response rate after the administration of the intermediate dose of PERM. The highest dose of PERM decreased both FR and FI response rates. FR rates in male rats were more disrupted than those in female rats. Only the highest dose of DEET decreased FR and FI response rates in male and female rats. FR rates were more disrupted in female rats than in male rats. Synergistic effects were only observed when FI response rates decreased in male rats upon exposure to half the low dose of PB with half the low dose of PERM or half the low dose of PB with half the low dose of DEET. The results of this experiment thus show that small doses of PB, PERM and DEET disrupt well-established, schedule-controlled behavior in male and female rats in a schedule- and gender-dependent manner; schedule-dependent and gender-dependent synergistic effects were also observed. The mechanism by which the compounds exert these behavioral effects remains to be determined.

107. WHO, IARC Working Group on the Evaluation of Carcinogenic Risks to Humans. Permethrin. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans. 1991; 53 (Occupational Exposures in Insecticide Application, and Some Pesticides. October 1990, Lyon, France)329-348; ISSN: 1017-1606.

108. Zhang, S. Y.; Ito, Y.; Yamanoshita, O.; Yanagiba, Y.; Kobayashi, M.; Taya, K.; Li, C. M.; Okamura, A.; Miyata, M.; Ueyama, J.; Lee, C. H.; Kamijima, M., and Nakajima, T. Permethrin may disrupt testosterone biosynthesis via mitochondrial membrane damage of leydig cells in adult male mouse. Endocrinology. 2007; 148(8):3941-3949. ISSN: 0013-7227 (Print).
Keywords: pyrethroids/ permethrin/ ppm/ mice/ male/ adverse effects/ reproductive toxicity/ mitochondria/ gene expression/ membrane damage
Notes: The widely-used synthetic insecticide permethrin dramatically reduces testosterone levels and sperm counts in adult male mice exposed for six weeks. Permethrin causes reproductive damage by altering the beginning steps of testosterone synthesis in the testes, lowering testosterone production. Permethrin is used in homes and agriculture and it can be found in dust and food. Doses used in the experiment were higher than those people would encounter regularly, but effects were seen at both doses tested.-ATF
Abstract: Permethrin, a popular synthetic pyrethroid insecticide used to control noxious insects in agriculture, forestry, households, horticulture and public health throughout the world, poses risks of environmental exposure. Here we evaluate the reproductive toxicity of cis-permethrin in adult male ICR mice that were orally administered cis-permethrin (0, 35 or 70 mg/kg/day) for 6 wk. Caudal epididymal sperm count and sperm motility in the treated groups were statistically reduced in a dose-dependent manner. Testicular testosterone production and plasma testosterone concentration were decreased with an increase in luteinizing hormone (LH) significantly and dose-dependently and a significant regression was observed between testosterone levels and cis-permethrin residues in individual mice testes after exposure. However, no significant changes were observed in body, reproductive organ absolute and relative weights, sperm morphology, and plasma follicle stimulating hormone (FSH) concentration after cis-permethrin treatment. Moreover, cis-permethrin exposure significantly diminished the testicular mitochondrial mRNA expression levels of peripheral benzodiazepine receptor (PBR), steroidogenic acute regulatory protein (StAR), and cytochrome P450 side-chain cleavage (P450scc) enzyme and protein expression levels of StAR and P450scc. At the electron microscopic level, mitochondrial membrane damage was found in Leydig cells of the exposed mouse testis. Our results suggest that insecticide permethrin may cause the mitochondrial membrane impairment in Leydig cells and disrupt testosterone biosynthesis by diminishing the delivery of cholesterol into the mitochondria and decreasing the conversion of cholesterol to pregnenolone in the cells, thus reducing subsequent testosterone production.