ABSTRACTS OF SUBMITTED PRESENTATIONS

S21

Title: Investigation of an Ocular-Respiratory Syndrome Associated with Influenza Immunization, Canada, 2000.

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Objectives: In late October 2000, Health Canada was informed of a cluster of individuals who developed ocular or respiratory symptoms within hours of influenza immunization. This prompted intensive public health investigations to determine the frequency and severity of this unusual vaccine-associated adverse event and its cause and mechanism.
Methods: Canadian public health authorities developed a case definition (red eyes, respiratory symptoms, facial edema or a combination of these occurring within 2 to 24 hours of receipt of the influenza vaccine, and all resolving within 48 hours of onset) and implemented enhanced surveillance of the reaction. A supplementary symptom and risk factor questionnaire was designed to accompany all reports of influenza vaccine-associated adverse events. Public health authorities and researchers met regularly to share data, determine epidemiological and clinical studies needed, agree on communication strategies, and make decisions related to influenza immunization programs.
Results: By January 24th, 2001, Health Canada received reports of 1,313 persons who met the case definition. Of these, 66% presented with red eyes with or without other symptoms. Seventy-eight percent (78%) were between 30 and 59 years of age; 76% were female. Eight cases required hospitalization, none died. Fifty-three percent (53%) of the cases had been immunized in the past, 21% of these had experienced adverse reactions. Cases were significantly less likely to have been immunized with influenza vaccine in the past as compared to individuals who did not meet the case definition (p value=0.004). Cases were not more likely to have a history of environmental allergies or asthma. Approximately 13 million doses of influenza vaccine were distributed in Canada this season; 9 million from manufacturer A and 4 million from manufacturer B. Over 96% of cases were associated with the influenza vaccine from manufacturer B. Transmission electronmicroscopy studies identified the presence of a higher level of aggregates and unsplit virions in vaccine from manufacturer B as compared to vaccine from manufactuer A and from vaccine produced by both manufacturers in the previous season.
Conclusion: The identification of this influenza vaccine-associated adverse event and the ensuing public health response has led to a review of pre-licensure requirements and guidelines for use of next year's vaccine. Because the benefits of influenza immunization greatly exceed the risks associated with this adverse event, health authorities agreed that influenza immunization programs should continue.

S22 Characterization of endogenous avian leukosis viruses in a chick fibroblast substrate used in the production of measles and mumps vaccines

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Previous findings of low-level of reverse transcriptase (RT) activity in chick-cell derived measles and mumps vaccines showed this activity to be associated with retroviral particles containing RNA of both subgroup E endogenous avian leukosis viruses (ALV-E) and endogenous avian viruses (EAV). To better characterize vaccine-associated ALV-E, we examined the endogenous ALV proviruses (*ev* loci) present in a White Leghorn CEF substrate pool by restriction fragment length polymorphism. Five *ev* loci were detected, *ev*-1, *ev*-3, *ev*-6, *ev*-18 and *ev*-19. Both *ev*-18 and *ev*-19 can express infectious ALV-E, while *ev*-1, *ev*-3, and *ev*-6 are defective. We analyzed the full-length sequence of *ev*-1 and identified an adenosine insertion within the *pol* RT-β region which results in a truncated RT-β and integrase, possibly explaining its RT(-) phenotype. Based on the *ev* loci sequences, RT-PCR assays were developed to examine expression of ALV-E particles (EV) in CEF supernatants. Both ev-1- and *ev*-3-like RNA sequences were identified as well as two other RNA sequences with intact *pol* regions, presumably of *ev*-18/-19 origin. Inoculation of susceptible quail fibroblasts with CEF culture supernatants from both 5-azacytidine-induced and non-induced CEF led to ALV infection, confirming the presence of infectious ALV-E. Our data demonstrate that both defective and nondefective *ev* loci can be present in CEF vaccine substrates and suggest that both *ev* classes may contribute to the ALV present in vaccines.

S23 Safety and Tolerability of Influenza Virus Vaccine, ~~Trivalent~~, Types A&B, Live, Cold-Adapted (CAIV-T) in Children and Adolescents with Moderate to Severe Asthma

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Background: Influenza infections can cause severe respiratory disease in high risk persons such as those with asthma, but immunization rates for high risk groups remain suboptimal. CAIV-T administered by intranasal spray was previously shown to be effective in healthy adults and healthy children.
Objective: To determine the safety and tolerability of CAIV-T in children and adolescents with moderate to severe asthma.
Methods: A randomized (1:1), double-blind, placebo-controlled study in subjects 9 to 17 years old with moderate to severe asthma. Spirometry was performed twice before vaccination to establish a baseline and once 2 to 5 days following. Peak flow, clinical asthma symptom scores, and nighttime awakening scores were reported daily from 7 days pre to 28 days post vaccination. The primary outcome was percent change in predicted FEV_1 (forced expiratory volume in 1 second) before and after vaccination.
Results: 48 subjects were enrolled. Percent change in FEV_1 from baseline to post vaccination was not different between the two groups (0.2% vs. 0.4% for the CAIV-T and placebo groups, respectively; p=0.78). Secondary outcomes did not differ between the two groups; these included the number of subjects with a decrease in FEV_1 ≥15% from baseline, reductions in peak flow ≥15%, ≥30%, or ≥2 standard deviations below baseline, use of beta adrenergic rescue medications, and clinical asthma symptom scores before and after vaccination. Asthma exacerbations, controlled with outpatient therapy, occurred in 2 of 24 CAIV-T recipients vs. 0 of 24 placebo recipients (p=0.49). The same proportion of subjects in each group experienced post vaccination symptoms within 10 days (92% and 91%, respectively; p=1.0). No serious adverse event occurred.
Conclusion: CAIV-T was generally safe and well-tolerated in children and adolescents with moderate to severe asthma.

S24 Reactogenicity and passage into the brains of enterotoxins and cpg-oligonucleotides administered intranasally to mice.

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Objectives : *Escherichia coli* heat-labile toxin (LT) and various LT derivatives are widely tested as mucosal adjuvants. The mucosal adjuvanticity of CpG oligonucleotides (CpG-ODNs) has been recently described. We have here evaluated the reactogenicity of these adjuvants in mice upon intranasal delivery.
Methods : Reactogenicity was assessed in the nasal cavities and in the brain of mice by histopathological examination of tissue sections after intranasal drop administration. The passage of the enterotoxin or the CpG-ODNs into the brain was analysed in situ by different techniques.
Results : While confirming the adjuvanticity of the different enterotoxin molecules, we noted that they all induced severe reactogenicity involving the nasal respiratory and olfactory mucosae. Moreover, there was penetration of the enterotoxins into the olfactory bulbs via the olfactory nerve that was associated with severe inflammation in the meninges, the olfactory nerve and glomerular layers of the olfactory bulbs. The enterotoxin could be detected within the brain 1 hour after its intranasal administration, and its presence could still be observed one month later. Similar results were obtained with the native LT or CT, and with LT mutants. CpG-ODNs induced a slight local inflammation in both the nasal and olfactory mucosae of the nose. The passage of the CpG-ODNs into the brain was demonstrated by immunohistochemistry and in situ hybridization. The presence of the CPG-ODNs was detected in the different layers of the olfactory bulbs, suggesting a trans-neuronal transport. The CpG-ODNs could be detected within the brain 30 min after its intranasal administration, and its presence still observed two days later.
Conclusions : These results suggest that possible severe side effects could be associated with the use of enterotoxin or CpG-ODNs as intranasal adjuvants in humans. One major safety issue is their possible passage within the brain, that was clearly observed in mice. In view of these results, we would advise that similar experiments be performed with all such potentiel mucosal adjuvants before clinical research programmes are initiated.