

CALIFORNIA STATE SCIENCE FAIR 2010 PROJECT SUMMARY

Name(s)

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Project Number

S0405

Project Title

Swine Flu Pandemic: Was the Fear Real? Development of a Real-Time PCR Assay to Detect the 2009 Influenza A (H1N1) Virus

Abstract

Objectives/Goals The experiment is designed to screen individuals exhibiting flu-like symptoms in the community using a molecular diagnostic assay for the 2009 Influenza A (H1N1) virus to determine its prevalence.

Methods/Materials

Nasal swabs were collected from consenting adults exhibiting flu-like symptoms by a trained healthcare professional. Any potential virus in the samples was inactivated on-site and the samples brought to the lab to isolate viral RNA. Sequence alignments of the various influenza A virus genes (hemagglutinin (HA), neuraminidase (NA), and matrix (M)) were performed using the Influenza Virus Resource website. Two real-time PCR primer/probe sets were designed to specifically detect the 2009 H1N1 HA and NA genes as well as another primer/probe set for the universal detection of all Type A Influenza viruses. Following isolation of viral RNA, reverse transcription-polymerase chain reactions were performed and each patient sample was scored as positive or negative for each of the three assays (HA, NA, and M) and its infection status identified.

Results

Of the 27 nasopharyngeal samples evaluated by the real-time RT-PCR assays, five were positive for the 2009 H1N1 virus. 4 were strong to moderately positive in all three assays and 1 revealed to be weakly positive. With 5 positive out of 27, this represents about 18% of the individuals being infected with the 2009 H1N1 virus. Only one was positive for only the M gene suggesting that this individual was infected with one of the more common seasonal flu viruses.

Conclusions/Discussion

Though the number of individuals screened is limited, my findings reveal that less than 20% were positive for 2009 H1N1. Since nearly all of the tested individuals were treated with Tamiflu for presumed 2009 H1N1 infection, if this rate of positivity is extrapolated to the larger population, my data suggests that more individuals were treated with antiviral drugs than was necessary. Wider implementation of molecular assays like the one developed here is likely to impose more discretionary administration of antiviral therapy and reduce overtreatment. It would also avoid development of resistant virus strains in the community. Furthermore, confirmed test results reduce anxiety for patients and close contacts at work and home, as well as for individuals at greater risk of contracting severe illness, such as pregnant women and people with immunocompromised conditions.

Summary Statement

Development of a real-time PCR assay to determine the prevalence of 2009 Influenza A (H1N1) virus infection during the recent flu pandemic.

Help Received

Dr. Borsada and her staff for sample collection; My father helped me purchase the Viral RNA kit, RT-PCR kit, as well the primers and probes and provided supervision during the RNA isolation procedure and PCR reaction set-up.