Practical Training Exercise

ANALYZING AND MANAGING RISKS IN LIFE SCIENCES RESEARCH

Based on the article by El-Zoghby EF, et al. “Isolation of avian influenza H5N1 virus from vaccinated commercial layer flock in Egypt.” Virol J. 2012 Nov 27; 9: 294.

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ADVANCING SCIENCE, SERVING SOCIETY

This exercise was developed by Center for Science, Technology and Security Policy at the American Association for the Advancement of Science (AAAS).

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Learning Objectives

1. Develop the skills to think critically about risks and risk mitigation strategies needed in your own scientific environment;

2. Enhance your ability to identify strategies and approaches that minimize identified risk while continuing to maintain the high-quality and utility of the scientific activity; and

3. Apply the risk analysis framework to your own or your peers' scientific activities.

Participant Expectations

By the end of this exercise, you will have familiarity with:

1. The definitions of different types of risks associated with laboratory, field, and public health research.
2. The process of risk analysis—risk identification, assessment, management, and communication—including:
   - How to identify and assess risks by considering the possible likelihood and consequences of risks, and the risks versus benefits of a research activity,
   - Strategies for managing risks, and
   - Who, when, and how to communicate risks.
3. How to apply the risk analysis framework to your own scientific activities.

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Ground Rules for Participation

1. Prior to starting this exercise, participants should have read the case study article.
2. Ask the facilitator to clarify questions about the case study article.
3. Focus on understanding and analyzing the diverse risks involved in the research rather than on critiquing the methodologies or research choices of the authors.
4. Interact with one another in a way that encourages open communication and exchange of ideas. For example, listen to everyone's ideas respectfully.
5. You may want to take your own notes to enhance your ability to actively participate in the training activity.

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Biorisk Glossary

These definitions are from the WHO's *Responsible Life Science for Global Health Security: A Guidance Document*.

- Bioethics
- Biorisk
- Biorisk reduction
- Laboratory biosafety
- Laboratory biosecurity
- Dual-use life sciences research
- Research excellence

<u>Additional concepts:</u>

- Protection of human subjects
- Protection of animal subjects
- Responsible research/responsible conduct of research

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Risk Analysis Framework

Your risk review will follow these 4 Stages:

1. **Risk Identification**
2. **Risk Assessment**
3. **Risk Management**
4. **Risk Communication**

1. Risk Identification

process by which researchers consider all possible internal, external, and organizational risks.

Asks the question:

- ***What are the possible risks associated with the research?***

2. Risk Assessment

process by which researchers identify needed resources and consider biosafety/biosecurity recommendations.

Also defined as the “process of evaluating the risk(s) arising from a hazard(s), taking into account the adequacy of any existing controls and deciding whether or not the risk(s) is acceptable).” (OHSAS 18001: 2007)

Asks the questions:

- ***How likely are the risks to occur?***
- ***What are the potential consequences if the risks occur?***
- ***Do the risks outweigh the benefits?***

3. Risk Management

process by which researchers consider regulations/guidelines, training, and SOP compliance issues.

Asks the question:

- ***What risk management strategies could minimize the likelihood that the risk will occur or the consequences if the risks occurred?***

Possible strategies: physical barriers, personnel training or vetting, regulations and laws, and/or alternative experiments

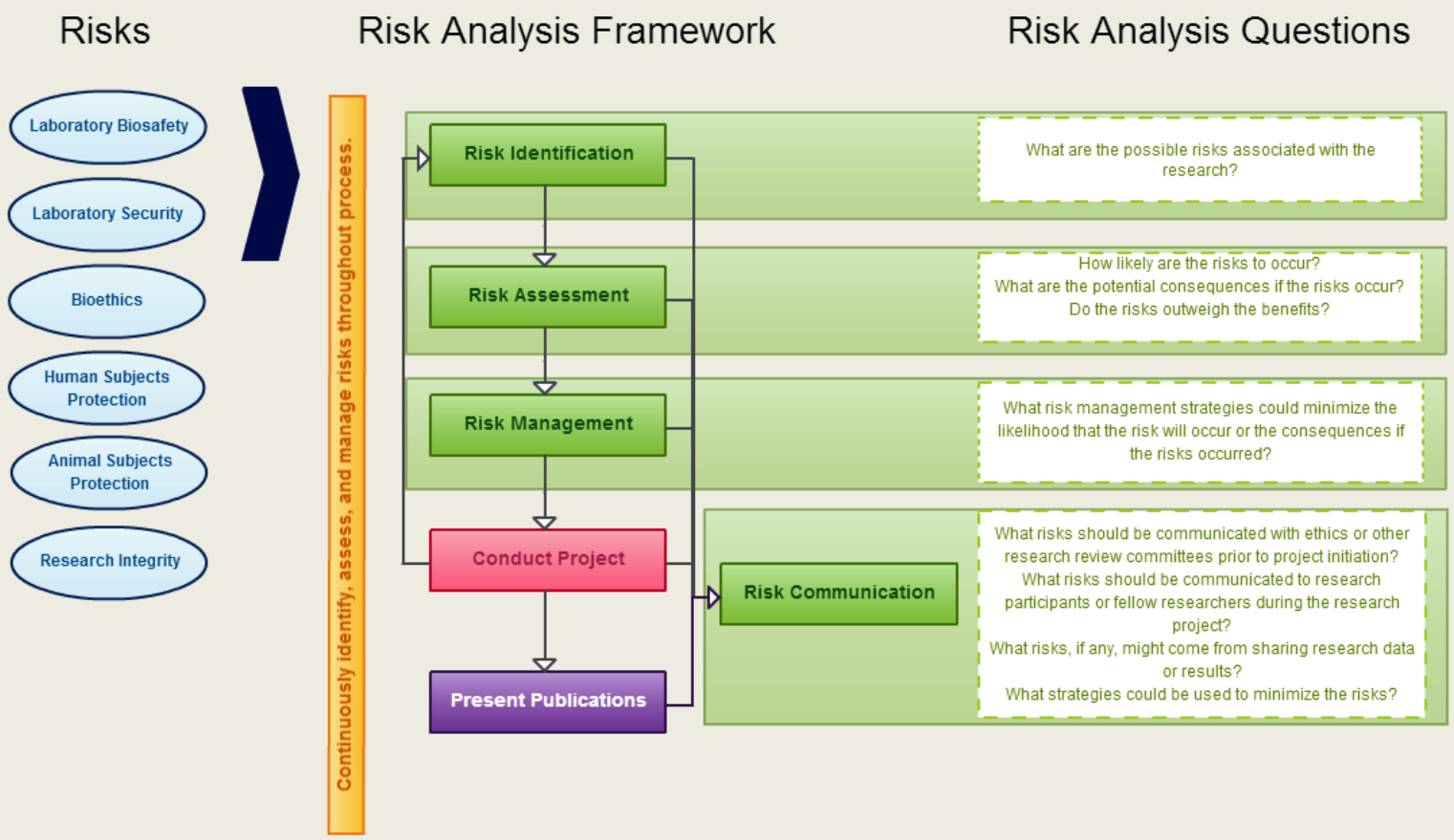
4. Risk Communication

process by which researchers consider communication strategies, non-compliance issues and approval/modification processes.

Asks the questions:

- ***What risks should be communicated with ethics or other research review committees prior to project initiation?***
- ***What risks should be communicated to research participants or fellow researchers during the research project?***
- ***What risks, if any, might come from sharing research data or results?***
- ***What strategies could be used to minimize the risks?***

Simplified Risk Analysis Chart



CASE STUDY

Isolation of avian influenza H5N1 virus from vaccinated commercial layer flock in Egypt

El-Zoghby EF, et al. “Isolation of avian influenza H5N1 virus from vaccinated commercial layer flock in Egypt.” Virol J. 2012 Nov 27; 9: 294.

Outline of Case Study

Part 1: Research Question/Hypothesis

Part 2: Background Information Overview

Part 3: Methodology

Part 4: Risk Analysis in the Research Article

Part 5: Research Results and Conclusions

Research Question/Hypothesis

Research Statement:

All human cases of H5N1 infection in Egypt, except for three cases, were linked to direct contact with infected birds in backyards and/or live bird markets.

The currently circulating HPAI H5N1 virus in Egypt (from the 22.1/C clade) continues to circulate despite large scale vaccination of commercial and backyard birds.

The authors propose to isolate and characterize, at the molecular level, “an HPAI H5N1 virus isolated from twenty weeks old chicken layer flock, which was vaccinated three times with a commercial inactivated H5N1 vaccine.”

Background Information Overview

Background Avian Influenza (H5N1)

- The highly pathogenic H5N1 avian influenza virus first infected humans in Hong Kong in 1997.
- In 2003-2004, HPAI H5N1 re-emerged and subsequently spread throughout the world.
- Human infections with H5N1 are rare and often occurs when people come in close contact with H5N1.
- H5N1 influenza infections in humans is not well understood.
- Human-to-human transmission is rare and usually within close contact with each other.
- In 2006, H5N1 first emerged in Egypt and became endemic in the avian population in 2008.

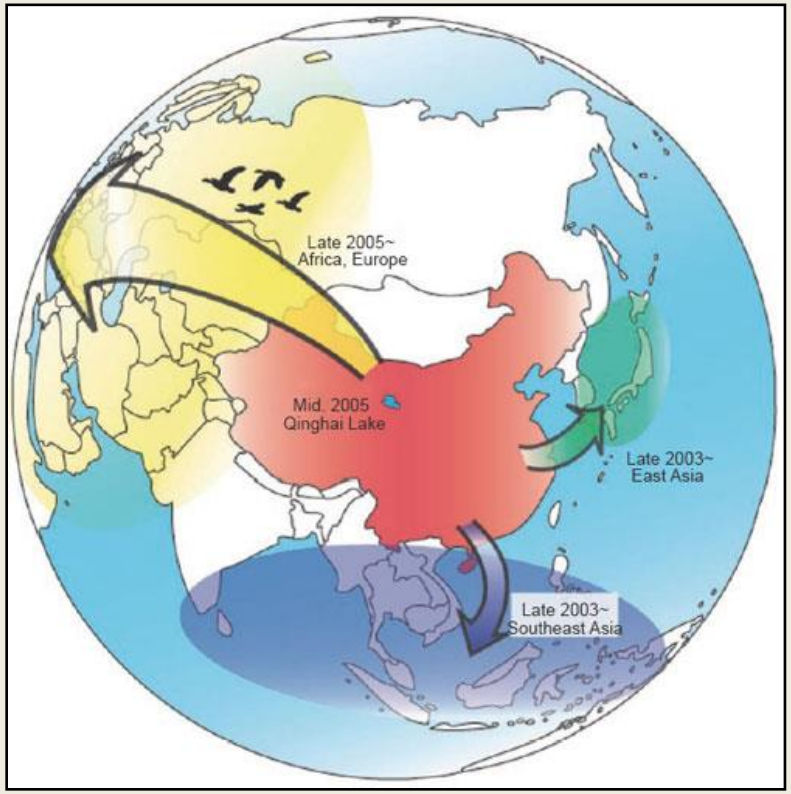


Photo Credit: Cell Research, 2009

Background Information Overview

Photo Credit: Rimmele, 2013

H5N1 in Egypt

H5N1 was first identified in Egypt in 2006.

Infection of H5N1 in several types of birds, donkeys, and pigs have been reported.

- Until July, 2012, 168 humans were infected in Egypt, 60 of which were fatal.
- Human infections with HPAI H5N1 were caused by direct exposure to infected birds. (Kandeel, 2010)

Background Information Overview

How Influenza Mutates

- Has 2 surface glycoprotein genes
  - HA – hemagglutinin
  - NA – neuraminidase
- Virus undergoes natural mutations
  - Antigenic drift: mutations within individual genome
  - Antigenic shift: exchanging genes between 2 viruses during co-infection (genetic recombination)

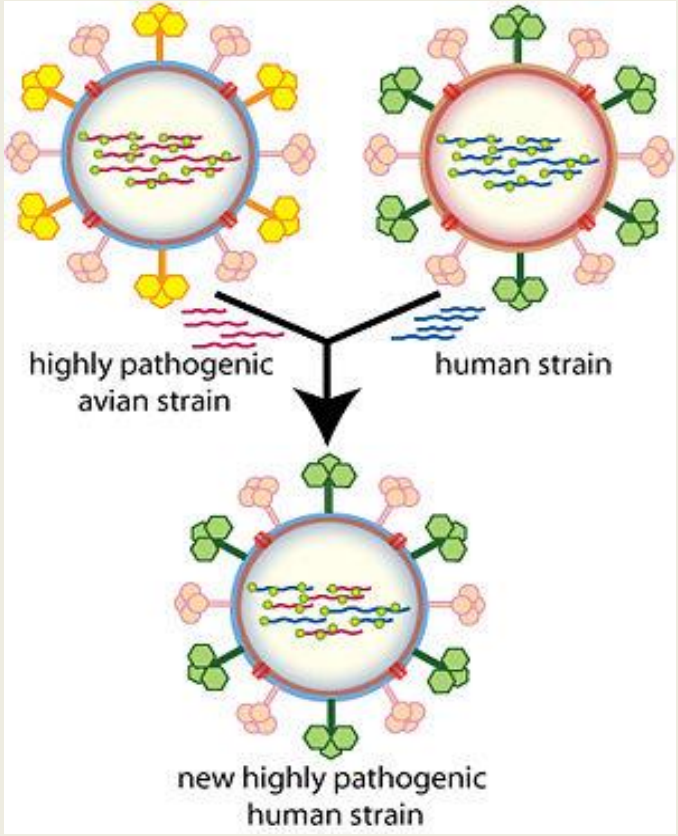


Photo Credit: Wikipedia, 2012

Background Information Overview

Influenza in Egypt Today

Photo Credit: Reuters, 2012

- “Despite control efforts, the virus became endemic in poultry in Egypt by 2008.” Over 70% of the Egyptian poultry production is marketed through live bird markets. (El-Zoghby, 2012)
- Egypt developed inactivated H5N1 and H5N2 vaccines to minimize the spread of H5N1 in poultry.
- Mutations in the 22.1/C clade of H5N1 have mutation in their HA and NA proteins, which might suggest decreasing virulence in mammals and adaptation to humans.

Research Methodology

- **Sample population.** The sample population was commercial chickens that received the standard commercial H5N1 inactivated vaccine.
- **Sample symptomatic chickens.** Throat swabs and serum samples were collected from chickens showing clinical symptoms of H5N1 infection.
- **Virus isolation.** Viruses were isolated in specific pathogen free fertilized chicken eggs (embryos) after inoculation with the samples. Virus titers were estimated by egg-infective dose.
- **H5-specific antibodies detection.** Hemagglutination inhibition tests were used to identify H5-specific antibodies in the serum samples.
- **Viral RNA extraction.** Viral RNA was extracted from pooled swab samples using a commercially available kit. Real-time reverse transcription polymerase chain reaction (RT-qPCR) was used to amplify viral RNA.
- **Sequence analysis of HA and NA genes.** The HA and NA gene were amplified and sequenced using a commercially available kit. The online BLASTN algorithm was used to deduce the amino acid sequences from the viral gene sequences.
- **Phylogenetic analysis.** The H5 and N1 sequences from the sample population was compared to existing H5N1 sequences, which were obtained from the online GenBank database, using standard bioinformatic analysis.
- **Protein structure modelling**. The tertiary structures of the H5 and N1 proteins were created using commercially available software. The locations of the substitutions in the HA and NA protein sequences were mapped onto the structures using commercially available software.

Risk Analysis in this Research Article

While risk analysis is an important part of science, few scientific publications include in-depth descriptions of how the authors assessed and managed risk.

Today your task is to perform a risk analysis based on this research article.

To begin, answer the following question:

Based on your current knowledge of the experimental procedures or research purpose, what risks might be important to consider in designing, carrying out, or communicating this research?

Risk Identification

Isolation of avian influenza H5N1 virus from vaccinated commercial layer flock in Egypt

Questions
What, if any, are the potential biosafety risks to researcher and staff during sample collection, transport, and handling?
What, if any, are the potential biosafety and biosecurity risks associated with the laboratory identification and analysis of the H5N1 viruses?
Does this study pose any potential risks to human health in Egypt?
Does this study have any potential to be used to cause harm?
What, if any, are the animal treatment and welfare risks to the commercial chickens in this study? Because this study positively identifies some flocks as containing H5N1 carriers, are there additional risks of spreading of H5N1?

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Risk Assessment

Isolation of avian influenza H5N1 virus from vaccinated commercial layer flock in Egypt

Question
What steps in this study present the greatest biosafety threats to the research team? How does working with a pathogen of global health concern, such as H5N1, change this assessment?
What is the minimum biosafety level at which this study should be conducted? Could you carry out this study at your own institution? Why?
What are the resources, expertise, training, and tools that could be useful in assessing the risks identified for this research project?

Risk Management

Isolation of avian influenza H5N1 virus from vaccinated commercial layer flock in Egypt

Question
What international, domestic, and institutional regulations, laws, and best practice help to mitigate the identified risks?
To minimize biosecurity hazards from this study, what standard operating procedures (SOPs) in laboratory analysis and field collection should be employed?
What, if any, protective measures (safety and/or security) should be in place before the start of the study?
What, if any, are the specialized competencies, skills, and training needed to carry out this research activity, including safely and successfully collecting, transporting, and analyzing samples?
Other than physical barriers and training, what scientific approaches could be used to minimize the risk of accidental exposure to the researcher, theft of the samples, or misuse of the study results?

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Research Results and Conclusions

Results

- Several birds that were vaccinated with a commercially inactivated H5N1 vaccine had respiratory distress and other clinical symptoms of infection with HPAI H5N1 and approximately 27% died.
- All swab samples contained H5N1 virus.
- The isolated viruses were closely related to the 22.1/C group of H5N1 viruses isolated from live bird markets, backyards, and humans.
- Mutations in the HA and N1 genes were observed.

Conclusions

- Updated H5N1 vaccines for commercial poultry could be necessary in Egypt because of continued infection of birds despite vaccination.
- “Birds in backyards and LBM remain the main potential source of H5N1 infection to both commercial poultry and humans in Egypt”
- The authors advocate for targeted surveillance to better understand the spread of HPAI H5N1 in commercial poultry.
- The authors state that rigorous control measures, including “enforcement of biosecurity, culling infected birds, and constant updates of [the] vaccine virus strains” are needed to prevent transmission between backyard birds, commercial poultry, birds in live bird markets, and humans.

Risk Communication

Isolation of avian influenza H5N1 virus from vaccinated commercial layer flock in Egypt

Question
What are the risks that should be communicated during this research? To whom?
How would you communicate the risks and risk management steps to an institutional ethics/experiment review board or committee?
What, if any, risks are associated with making the sequences of the HA and N1 genes publically available?
Should the government play a role in limiting what types of information researchers share and the venues for information sharing when working with potentially dangerous pathogens?
Are there any circumstances under which researchers should not share all or some of their findings with the public (e.g., specific mutations of important viral genes)? What resources, if any, are available to assist researchers in considering the benefits and risks of communicating research results?

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Final Exercise: Risk in Your Own Research

Perform a risk analysis of your own research. Choose one past, ongoing, or future research project to analyze:

1. Identification: What are the primary risks you face in your research? Think about the risks to you and other researchers and technicians in the field, clinic, and/or lab, the general public, the environment and economy, your institution, and human and animal subjects.
2. Assessment: What are the consequences of the identified risks if they occur? How likely are they to occur? Based on your assessment of the potential consequences, are there are there any risks that could harm people, animals, crops, or the economy? What resources, capabilities, and skills are needed to mitigate these risks?
3. Management: What strategies could you use or resources you could refer to minimize or mitigate these risks? (These strategies should not decrease the quality of the research.) For ideas of possible strategies and resources, consider those discussed in this practical exercise and from your own experiences. Are there any risks associated with your research that cannot be adequately mitigated?
4. Communication: What risks, if any, are associated with communicating your research during the design or conduct of the research? What risks, if any, are associated with communicating the research results at scientific conferences and in publications? What strategies could you use to mitigate the risks? Are there any stakeholders with whom you must share or should share the risks of your research? Your findings?

Example Risk Analysis Strategy

Communicate

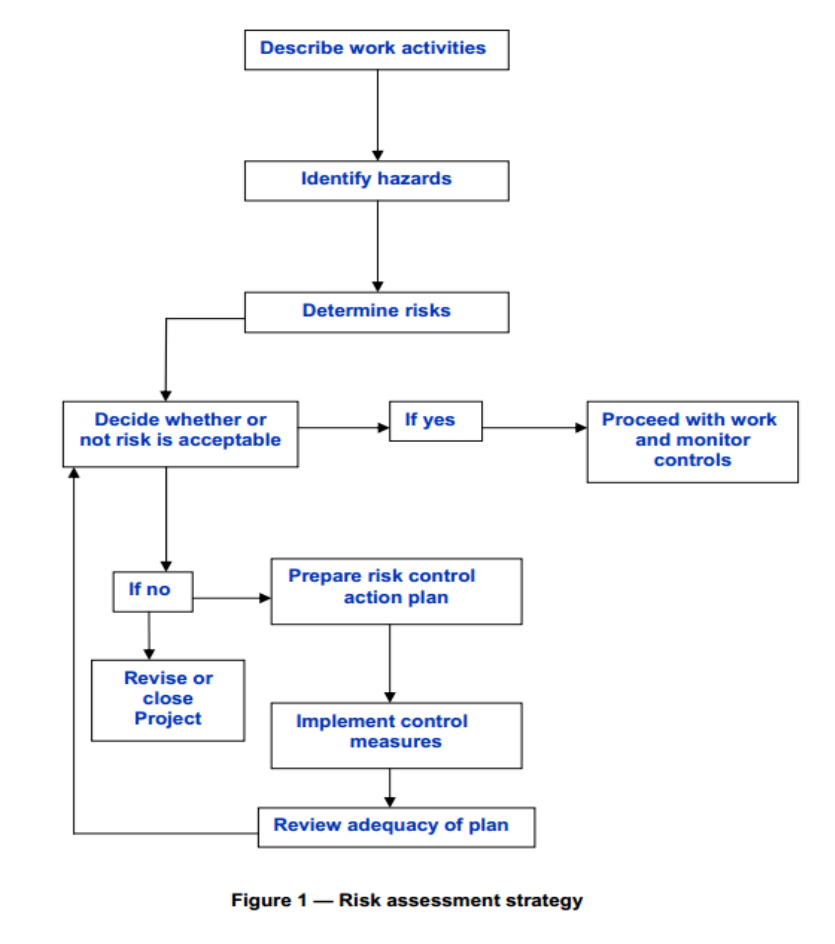

Figure 1 — Risk assessment strategy

"Laboratory risk management." CWA 15793: 2011

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Diagrams and Images

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