Egypt Field Epidemiology Training Program
Case Studies in Applied Epidemiology
September 1999

Outbreak Investigation of an Unknown Illness in a Rural Village, Egypt

Participant's Guide

Learning Objectives
After completing this case study, the participant should be able to:

- ☐ Describe major issues that need to be addressed before beginning a field investigation.
- ☐ List the steps in deciding how to conduct an outbreak investigation.
- ☐ Generate and test hypotheses related to field investigations.
- ☐ Understand how to formulate a case definition.
- ☐ Discuss how to interpret epidemiologic data and assist public health decision-makers in understanding the information.

This case study is based on an outbreak investigation that began in July 1996 in an area of southern Egypt. The work described here was done by the Ministry of Health & Population's (MOH&P) Field Epidemiology Training Program (FETP) and Central Public Health Laboratories (CPHL) in collaboration with the U.S. Centers for Disease Control and Prevention (CDC, Atlanta), National Center for Environmental Health (NCEH), NHANES Laboratory, Epidemiology Program Office, the U.S. Food and Drug Administration (FDA); and the Health Office of U.S. Agency for International Development (USAID-Cairo).

In 1998 Donna Shalala, Secretary of U.S. Department of Health and Human Services presented the Bernadino Ramazzinni Award to Dr. Abdel Nasser (Director, FETP) and Dr. Magda Rakha (UnderSecretary, CPHL) for the best example worldwide of a specific type of outbreak investigation.

This case study was developed by Dr. Abdel Nasser (FETP, Egypt), Dr. Doug Hatch (CDC/EPO), Dr. Carol Pertowski (CDC/NCEH), and Dr. Steve Yoon (CDC/NCEH) in 1999.

PART I. Outbreak: Initial information and request for assistance

In July 1996, public health officials in a region of southern Egypt, approximately 1,000 kilometers from Cairo, were alerted to an outbreak of an illness of unknown cause. Clinicians working in a health facility noticed that a substantial number of persons from a rural village were seeking treatment for an unusual illness for this area. The main symptoms were severe abdominal pain, persistent vomiting and generalized weakness. Less common symptoms included leg pain, diarrhea, jaundice (i.e. scleral icterus) and fever, but because of the isolated location of the village, information was incomplete. Seventeen persons were hospitalized and one death occurred in a young child. Clinicians diagnosed illness as suspected gastroenteritis and patients were generally treated with oral rehydration solution (ORS), and when necessary, intravenous fluids, antiemetics and various antibiotics, including tetracycline and ampicillin.

The local health officials were concerned that the outbreak might be due to *Vibrio cholerae* or an other infectious agent that could be a risk to the public's health in nearby areas. Assistance from the Preventive Sector in the Ministry of Health and Population (MOHP) was officially requested.

Question 1. Is your office willing to assist in the investigation? Why?

Question 2. What additional information would be helpful in deciding whether to assist local public health officials?

Question 3. Discuss what administrative steps are usually taken to prepare for an outbreak investigation in a remote area, including issues related to logistics.

Because local health officials as well as community leaders in the area of the outbreak were concerned about the number of ill persons and the possibility of an infectious disease spreading to other areas, investigators from the Ministry of Health and Population (MOH&P) and the Field Epidemiology Training Program (FETP) agreed to join the investigation team.

A meeting was held at the MOH&P to review the preliminary findings from the initial investigations, explore possible causes of this outbreak, and advise senior officials on how to proceed. Below is a table of reported symptoms from the initial investigation.

Question 4. What infectious agent(s) could explain the illness described? How would you prepare to document these?

Question 5. What other causes or diseases should be considered?

Question 6. At this point how would you advise MOH&P officials to proceed, and what additional information would you like to have?

PART 2. Outbreak: Case Definition

One of the first steps that needs to be taken on an outbreak investigation is to identify all people that may be ill due to the same cause. Several pieces of information are necessary, including the identification of ill people (***WHO***), place of occurrence (***WHERE***), and the time of occurrence (***WHEN***). This is the basic information needed to develop a case definition. The case definition is the foundation of an outbreak investigation. The case definition can be "broad". Broad case definitions when there is uncertainty about the extent of the outbreak and you need to identify or "capture" as many ill people as possible. The problem with a broad case definition is that it may include ill persons who are not actually ill due to the infection or exposure causing the outbreak. A "narrow" case definition, which is more specific or selective, may be useful if you suspect that there are other illnesses with similar symptoms, but are not due to the same factor or agent causing the outbreak. An example may be an outbreak of illness among persons who work in a specific occupation or live near a potential source of illness such as a chemical plant.

Question 7. Develop an initial case definition. What information do you need? Think about person, place, time.

Below is a table of cases identified by reviewing hospital charts. Cases include those individuals that were hospitalized as well as those that were treated as outpatients.

Reported Symptoms in Ill Persons & Household Members

Symptoms	Hospitalized	
	Yes (%)	No (%)
Headache	6 (35%)	6 (26%)
Leg Pain	5 (29%)	2 (11%)
Abdominal Pain	4 (24%)	5 (21%)
Fever	4 (23%)	3 (11%)
Fatigue	3 (17%)	4 (16%)
Vomiting	2 (12%)	4 (16%)
Nausea	1 (6%)	4 (16%)
Diarrhea	0 (0%)	2 (10%)
Total	17	24

Source: FETP, CPHL & NCEH Investigation, Southern Egypt--1996

Local Ministry officials inform you that the results of stool cultures were negative for *Vibrio cholerae*, Salmonella species (including typhoid), Shigella, and other enteric pathogens. Serologic testing of persons with jaundice showed no antibody documenting acute hepatitis A, B and no serologic test was positive for hepatitis C. Water samples taken also showed no coliform contamination.

One test of sera taken from an ill person, however, showed an elevated serum level for Pb (i.e. lead). The sample which showed an elevated serum Pb level was collected with regular red-top glass tube and ordinary syringe and needle.

LEAD (Pb) INFORMATION SHEET

Health effects
Exposure to lead can result in a range of health problems. In high concentrations (blood lead levels >= 70 µg/dL), lead can cause seizures, coma, and even death. Exposure to lead can also result in abdominal colic and anemia (range around 30- 40 µg/dL). Among young children, exposure to lead at lower concentrations, as low as 10 µg/dL, has been associated with lower intelligence, decreased hearing, and delayed neurologic development. The U.S. Centers for Disease Control and Prevention has set 10 µg/dL as the blood lead level of concern for young children (children < age 6 years).

Possible sources of lead exposure
Lead may be present in the household:
- improperly fired ceramics
- lead based paint
- traditional medicines
- cosmetics, e.g. kohl or surma
- canned food (if lead solder used in the can)
- water (if lead solder used on water pipes or on storage containers)

Leaded gasoline:
In countries still using lead in gasoline, combustion of leaded gasoline can result in contamination of the air and of soil and dust.

People may also be exposed to lead through industries contaminating the air and soil:
- lead smelting
- battery recycling
- industries using lead solder
- ceramics

* Parents working with lead at work may bring dust contaminated with lead home to their children.

Sample collection/analysis
Collection of blood:
Because the amount of blood lead that can cause health effects is so small, it is very important to assure that the sample is not contaminated with lead. The skin surface must be carefully cleaned to avoid contamination with lead dust. Blood collection materials should be batch tested to be certain that they are free of lead contamination. Blood should be collected in tubes with appropriate anti-coagulant, e.g. EDTA.

Analysis of blood lead:
Because the amount of lead in blood is small, it is important to use a laboratory or equipment that has a process for standardization. Lead is analyzed in whole blood, not sera.

Levels of concern for lead in environmental sources

Media	Acceptable level
Water	< 15 ppb
Soil	< 200 ppm (clean up is recommended in the US at levels > 500 ppm)
Dust	
Paint	

<u>Question 8</u>. How would you explain the meaning of the above information to senior MOH&P officials? Think about what you need to explain in terms of what was initially stated as the possible cause, and what was discovered after testing the sera of ill persons.

<u>Question 9</u>. What would you ask from the local officials about the possible source of lead exposure of this individual?

According to the local officials, the village is a rural farming village with unpainted mud-brick housing, few cars or trucks and minimal gasoline use. There was no known use of Pb-containing pesticides. The village is not located near any industry, such as battery recycling or smelting operations using Pb or other metals.

FETP was assigned to lead the outbreak investigation by the MOH&P. The National Center of Environmental Health (NCEH) at the U.S. Centers for Disease Control and Prevention (CDC) was contacted to help with the investigation.

Question 10. What type of expert assistance would be necessary for the investigation? Who would make up the investigation team? Why?

PART 3. Field Investigation and Laboratory Results

The team of epidemiologists, environmental scientists and laboratorians visited the involved village. Ill persons hospitalized were interviewed using a standardized questionnaire, and a sample of blood was collected after obtaining informed consent. Blood samples were sent to the NCEH laboratory at the U.S. CDC for testing for Pb concentration.

The blood lead level measurements collected from ill villagers and their family members are shown below. There are two BLL measurements, the first was done in July 1996 and the second was done in September 1996.

Age and blood lead levels of ill villagers and family members, Egypt 1996

ID	Age‡	BLL†	ID	Age‡	BLL†
1	3	68.8	22	33	103.1
2	4	45.3	23	33	46.2
3	6	49.1	24	35	78.2
4	7	83.9	25	35	50.3
5	9	48.3	26	36	64.1
6	10	58.5	27	36	67.0
7	11	16.8	28	38	78.8
8	12	76.1	29	40	58.0
9	13	61.4	30	45	86.5
10	14	78.5	31	47	75.7
11	15	47.5	32	49	58.0
12	15	57.1	33	56	.
13	16	67.6	34	60	26.1
14	16	.	35	65	103.8
15	17	26.5	36	65	38.6
16	19	78.2	37	65	35.0
17	19	56.5	38	70	72.4
18	20	54.0	39	70	57.4
19	22	72.6	40	76	38.4
20	26	73.9	41	78	43.5
21	27	62.6			

‡ Age in years
† Blood lead levels measured September, in µg/dL
. Missing value

Question 11. Plot the data from above table on the attached graph paper. Remember to label the axis and title the graph.

Question 12. What does the graph of age and BLL tell you about the possible sources of exposure? Why?

Following are the formulas for calculating the mean and the median.

$$Mean = \frac{\sum_{1}^{n} x}{n}$$

$$Median: \begin{cases} even\ number = \frac{n}{2} \\ odd\ number = \frac{n+1}{2} \end{cases} \quad where\ \ n = number\ of\ elements$$

Question 13. Calculate the mean, median, minimum, and maximum age and blood lead levels.

The relationship between blood lead levels and age can be examined by using a statistical method called regression. The theoretical basis of regression is beyond the scope of this case study. However, we have provided a print-out from an EpiInfo session below. In this example, the effect of age (indepdent or predictor variable) on the blood lead level (dependent variable) of ill individuals (original cases) is evaluated.

The value of interest is the correlation coefficient (r^2), which gives an indication of how closely one variable can predict the value of the other variable. Correlation coefficient (r^2) can range from 0 (no correlation) to 1 (perfect correlation). In the example below, r^2 is 0, suggesting that there is no relationship between age and blood lead level.

```
=========> REGRESS PBCDC AGE
Correlation coefficient: r   = -0.06   r^2 = 0.00
95% confidence limits:   -0.56 < r^2 < 0.56

Source             df   Sum of Squares   Mean Square   F-statistic
Regression          1          49.2344       49.2344          0.13
Residuals          37       14510.1748      392.1669
Total              38       14559.4092

ß Coefficients

                         ß            95% confidence                  Partial
Variable     Mean    coefficient     Lower        Upper    Std Error    F-test
AGE        32.3333   -0.0509198   -0.342110   0.240271    0.143710    0.1255
Y-Intercept           62.2694853
```

Question 14. What does the correlation coefficient tell you about the relationship between the two variables?

<u>Question 15</u>. Given what you know so far, what would be your next step in the investigation? What are other risk factors for elevated blood lead level? Think about exposures within the household, and outside the household.

PART 4. Result of Environmental Analysis

Below are tables from environmental analysis. Samples were taken from the homes of ill individuals. Blood lead levels presented are mean values.

Potential Sources of Lead Exposure
FETP, CPHL & NCEH investigation, Aswan, Egypt, 1996 (N = 41)

Risk Factors

Source	Exposure	BLL (µg/dL)
Medicine (Traditional)	Yes	67.3
	No	80.5
Kohl (Cosmetic)	Yes	69.0
	No	79.3
Paint (Interior of houses)	Yes	64.8
	No	80.5
Bread baked at home	Yes	78.8
	No	No exposed persons
Lamseh added	Yes	69.0
	No	79.3
Mill used to grind flour	Mill #1	81.9
	Mill #2	No exposed persons

Pb Concentrations in Environmental Samples

	% sample Pb detectable	Mean Pb level
Soil	29	57 ppm
Dust	22	43.5 µg/dL
Paint	0	Not applicable

Pb Concentrations — Household Items

	% sample Pb detectable	Mean Pb level
Tap water	80	8.9 ppb
Zeer (ceramic)	60	4.3 ppb
Container	0	Not applicable
Ceramic plates	0	Not applicable
Cooking pots	0	Not applicable

Question 16. What do the tables above tell you about the roles of traditional medicine, kohl, paint and flour? Examine the BLLs between exposed and unexposed groups.

Question 17. The Minister asked you to determine the extent of the lead problem. What would be your next step in the investigation?

PART 5. Determining the Extent of the Problem

In order to obtain accurate information about a population, one can either examine every member of the population, or examine selected members of the group and make estimates about the entire population. The process of selecting members from the population is called sampling.

There are several ways to sample. Random sampling is one of the ways. In order to carry out random sampling, a list of every eligible member of the population, called sampling frame, is required. Samples then can be selected from the sampling frame by using a random number table. Simple random sampling is not done very often because of the cost involved in obtaining a complete sampling frame. Also, with simple random sample, the sampled individuals may be spread out across a large area making it time consuming and expensive to visit.

There are other sampling methods, including cluster sampling, stratified sampling, and WHO EPI cluster sampling. Some of these sampling methods are used when simple random sampling process not possible for one of several reasons mentioned previously. In terms of simplicity, WHO EPI cluster sampling may be easiest to carry out, because unlike any other sample design, except for convenience sample, a sampling frame is not required.

All sampling methods, if done properly, should provide an accurate estimates of the entire population that the samples are selected from.

To determine whether a similar source of lead existed for other families in the village, a survey of the village was conducted. It was determined that a random sample survey of individuals living in the village would be used to assess the extent of lead poisoning in the village and risk factors for lead poisoning.

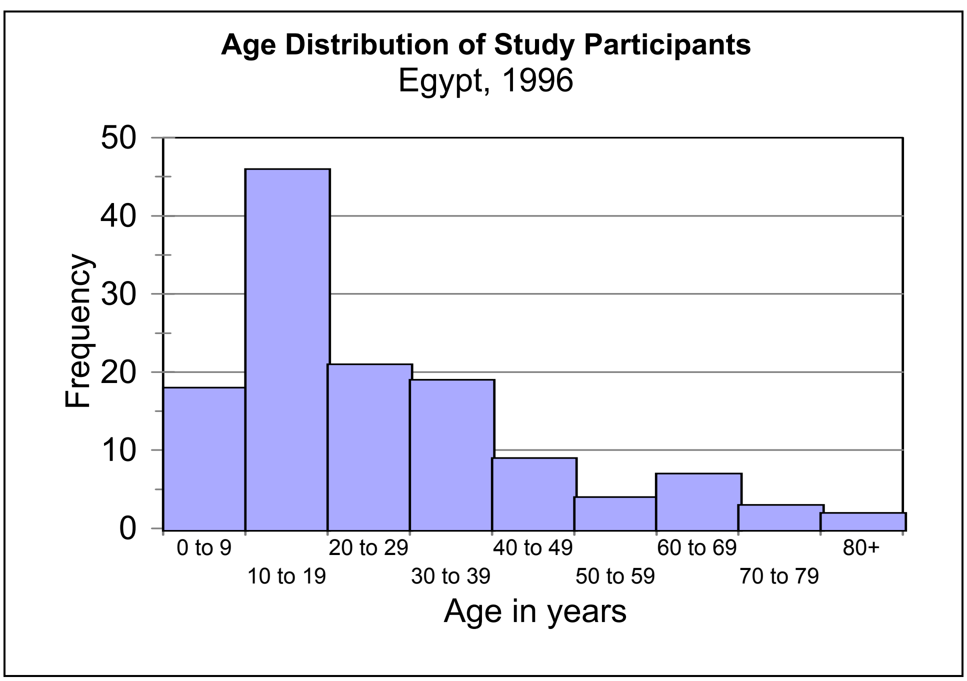
Question 18. In order to randomly select individuals for participation in the sample survey what information do you need? How would you obtain such information?

Question 19. The village had approximately 6500 residents older than 2 years of age. You decide on a 2% sample. How many people do you need to sample? How would you go about sampling these people?

Following are data from the village survey. One hundred twenty nine individuals living in the village were selected at random. Informed consents were obtained and blood samples were taken, along with environmental samples and behavioral information of the study participants.

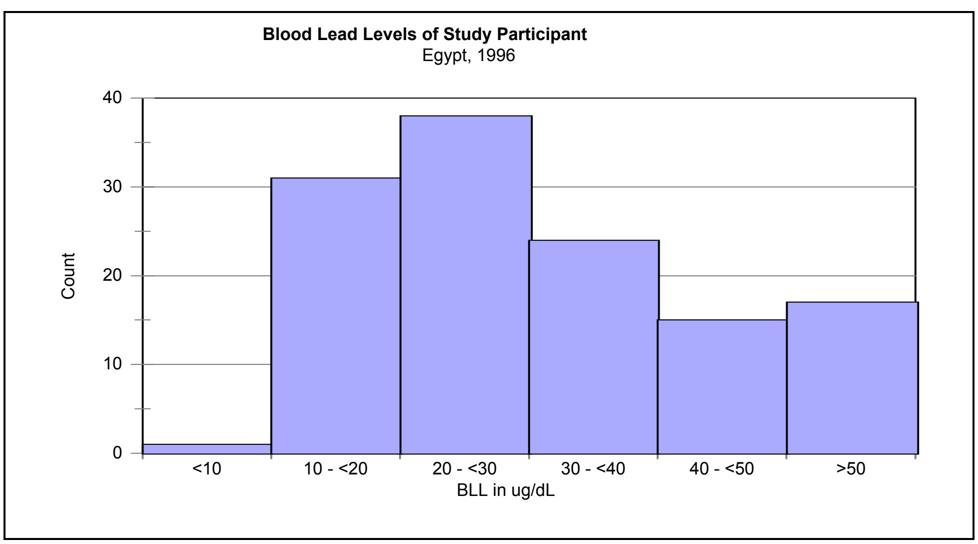
Age Distribution of Affected Persons, Egypt, 1996

Age	Count	Age	Count	Age	Count	Age	Count
3	1	17	3	32	1	59	1
4	3	18	5	33	2	60	2
5	5	19	3	34	1	65	5
6	2	20	3	35	7	70	2
7	1	21	2	37	2	76	1
8	1	22	3	38	2	80	2
9	5	23	1	39	1		
10	9	24	3	40	3		
11	6	25	2	41	1		
12	2	26	1	43	1		
13	6	27	3	45	2		
14	5	28	2	46	1		
15	4	29	1	48	1		
16	3	30	3	50	3		



Frequency of Blood Lead Levels Awsan Survey, Egypt, 1996

BLL	Count	BLL	Count	BLL	Count	BLL	Count
9.0	1	20.7	1	29.4	1	43.4	1
11.0	1	21.5	1	29.8	1	44.4	1
11.2	1	21.6	1	29.9	2	44.8	1
11.8	1	21.8	1	30.3	1	45.0	1
11.9	1	21.9	1	30.6	1	46.0	1
13.3	1	22.0	1	31.1	2	46.6	1
13.9	2	22.1	1	31.4	1	46.7	1
14.2	1	22.3	1	31.5	2	47.0	1
14.3	2	22.4	1	32.4	1	47.1	1
15.2	1	22.5	1	32.6	2	47.8	1
15.4	1	22.6	1	33.3	2	50.9	1
15.7	2	22.8	1	33.5	1	51.9	1
16.0	2	23.1	2	33.8	1	52.9	1
16.1	1	23.2	1	34.3	1	55.6	1
16.5	2	24.0	1	35.5	1	56.2	1
17.3	1	24.1	1	35.7	1	57.1	1
17.9	1	24.3	2	36.2	1	60.0	1
18.2	1	24.4	1	36.7	1	62.6	1
18.3	1	24.5	1	38.0	1	65.8	1
18.6	1	25.0	2	38.5	1	72.5	1
19.2	2	25.3	1	39.0	1	72.8	1
19.4	1	25.5	1	39.3	1	76.3	1
19.5	2	25.9	1	39.6	1	85.6	1
19.7	1	26.0	1	42.0	1	93.6	1
19.9	1	27.0	1	42.2	1	109.8	1
20.1	1	28.7	1	42.5	1	122.8	1
20.2	1	28.8	1	42.6	1	172.6	1
20.4	1	29.1	1	42.8	1		



Question 20. What do you notice about the shape of the age distribution curve? About the shape of the BLL curve?

The following output was obtained from Epi Info after running the *FREQUENCY* and *REGRESS* commands on Age and Blood lead level variables.

Age:

Total	Sum	Mean	Variance	Std Dev	Std Err
129	3325	25.775	342.113	18.496	1.629

Minimum	25%ile	Median	75%ile	Maximum	Mode
3.000	11.000	20.000	35.000	80.000	10.000

BLL:

Total	Sum	Mean	Variance	Std Dev	Std Err
126	4246	33.694	523.108	22.872	2.038

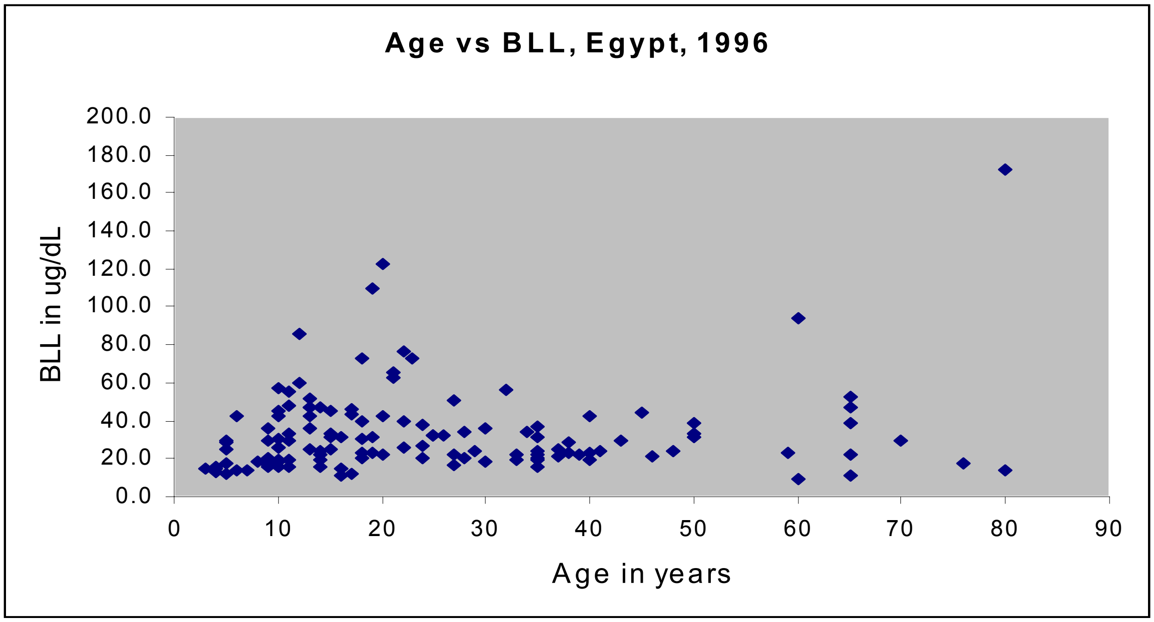
Minimum	25%ile	Median	75%ile	Maximum	Mode
9.000	19.900	27.850	42.000	172.600	13.900

```
=========> regress age bll
Correlation coefficient: r   = 0.14   r^2 = 0.02
95% confidence limits:   -0.16 < R < 0.19
```

Source	df	Sum of Squares	Mean Square	F-statistic
Regression	1	756.5499	756.5499	2.31
Residuals	124	40654.3072	327.8573	
Total	125	41410.8571		

ß Coefficients

Variable	Mean	ß coefficient	95% confidence Lower	Upper	Std Error	Partial F-test
BLL	33.6944	0.1075643	-0.032590	0.247719	0.070810	2.3076
Y-Intercept		21.6137759				



Question 21. If there was a relationship between Age and BLL, what would the statistics from EpiInfo show? What may be possible values of r^2? What would the graph show?

The investigation of the village had results similar to those of the outbreak investigation. Environmental samples, including dust, soil, water and household items (except for flour) showed no lead contamination. It was apparent that the lead contamination of flour was prevalent in this village.

The investigation of one of two flour mills in the village showed that lead solder ocassionally was used to repair the grinding stones. As grain was being ground up, lead could be ground up and contaminate the flour.

Question 22. What would your next steps be?

PART 6. Conclusion

Source of Pb exposure in the affected village

The mill implicated in the outbreak was visited. Upon arrival at the mill, the investigators noted a Pb smelting pot in the corner of the mill. Pb was used by the miller to attach the crosspiece to the grinding stone. (See attached diagram). Occasionally, the Pb would break off and contaminate the flour. The miller reportedly used about 2 kilograms of Pb per year. Analysis of grain from the mill showed no Pb; however, Pb was found in flour from the surface of the mill stone and in samples of flour after grinding was complete. Of note, the first cases of illness were noted in early July. The wheat harvest was at the end of May. In June, families had fresh grain as a source of flour, in addition to flour bought at the market.

The implicated mill was closed. A design not using Pb was developed and used to anchor the crosspiece to the grinding stone and the mill re-opened.

Use of Pb in flour mills

A mill wright (expert in milling) was brought to Egypt to review the milling process and to make recommendations. The mill wright noted that grinding stones are assembled from smaller pieces of stone and bound together to form the large grinding stones. Pb was used the fill in cracks between the smaller pieces of stone and to help balance the large grinding stone. Pb used to fill cracks can be ground along with the grain and contaminate the flour. As noted above, Pb was also used to anchor the cross piece. This Pb can break off at different times and be ground into the flour.

There are between 8,000 - 10,000 privately owned flour mills in Egypt that use the grinding technology of the mill implicated in the outbreak. Use of Pb is not recommended; however, because Pb was used in one mill, it was possible that Pb was also used in other mills. A survey of flour mills throughout Egypt was done to determine the extent of the contamination of flour by lead.

The Egyptian government, after reviewing the results of the investigation of the outbreak, banned the use of Pb in privately owned flour mills.

Extent of the Pb toxicity in the region

Because one flour mill in the region used Pb improperly, there was concern that residents of nearby villages may have also been exposed to Pb. A second, nearby village was selected and a survey of blood lead levels conducted in the village. Among residents of that village, the blood lead level was 14.7 µg/dL (range 3.4 - 26.7 µg/dL).