CHAPTER V

DATA ANALYSIS

In this study, the incidence and factors affecting tissue necrosis from *N. kaouthia* or *C. rhodostoma* bites will be evaluated. The result may be used in planning and evaluating strategies to prevent this condition and hopefully as a future guide to the management of patient in whom necrosis has already developed.

1. Questions and design study

Question 1 : What is the incidence and severity of tissue necrosis in victims after *N. kaouthia* or *C. rhodostoma* bites in Thailand ?

Design study : The question is for estimating the incidence of tissue necrosis frequency resulting from cuprit snakes, the assessment will be as follows,

No effect: No symptom/sign

Minimal: Pain, swelling, or ecchymosis within 5-7.5 cm of bite site

Moderate: Pain, swelling, or ecchymosis involving less than half the extremity (7.5-50 cm from the bite)

Severe : Pain, swelling, or ecchymosis extending beyond affected extremity (>100 cm from bite site)

Also, a correlation between the degree of snake clinical envenomation and tissue necrosis will be determined.

- The snakebite severity score (SSS) range of severity is 0 to 3 or 0 to 4 for each area of evaluation (pulmonary system, cardiovascular system, gastrointestinal system , central nervous system, hematological symptoms and local wound). A higher score indicates more severe effects. The score total ranges from 0-20 points. Each area will be added together to give accumulative data and justified the level of severity. The ratio scales should be following;

  No effect : Score 0-2

  Minimal severity : Score 3-5

  Moderate severity : Score 6-8

  Strong severity : Score 9-20

Question 2 : What are the factors effecting tissue necrosis and clinical consequences in patients after *N. kaouthia* or *C. rhodostoma* bites in Thailand both in prospective and retrospective studies ?

Design study : *The questionnaire* will consider tissue necrosis outcomes in relation to a population at risk from *N. kaouthia* or *C. rhodostoma* bites. The associated factors included demographic data, geographic data, factors related to snakebites and factor related to treatment. Analysis of data will be carried out using multiple logistic regression for prediction of more than one independent variable.

Question 3 : What is the difference of incidence, associated factors affecting tissue necrosis and clinical consequences in patient after culprit snakes bites both in prospective and retrospective studies ?

Design study : This questionnaire will consider incidence of tissue necrosis and associated factors in relation to clinical consequences at risk from *N. kaouthia* or *C. rhodostoma* bites comparing between prospective and retrospective studies.

Question 4 : Can enzyme inhibitors prevent or reduce tissue necrosis after *N. kaouthia* or *C. rhodostoma* bites in victims ?

Design study : This protocol will use intervention in an animal model resulting to testing chemical substances. The experiment is composed of *in vitro* and *in vivo* study to evaluate the advantages of chemical reagents. Each result will be determined to be continuous data and analyzed by student t-test comparison between control and experimental groups.

2. Missing data and Outliers

2.1. Missing data. Values can be missing because they were not measured, not answered, were unknown or were lost. Data mining methods vary in the way they treat missing values. Typically, they ignore the missing values, or omit any records containing missing values, or replace missing values with the mode or mean, or infer missing values from existing values (Miller, Gray and Woodward, 1993).

2.2. Outlier, a data item whose value falls outside the bounds enclosing most of the other corresponding values in the sample. It may indicate anomalous data and

should be examined carefully since it may carry important information. Outliers might indicate erroneous data collection, or might come from different parts of the population, of examples, indicating a new phenomenon.

In this study, after blinding two investigators, the contact person of each hospital will check every detail of the questionnaire to prevent data mistake. If some data are not completed, especially the snakebite severity score (SSS), they will be obtained through chart abstraction by the responsible nurses. Each item of the SSS part is very important for determinating snake envenomations and the severity of tissue necrosis.

3. Data summary

In continuous data, the most commonly used measure of central tendency is the mean. To compute the mean, all the numbers are added up and divide by how many numbers there are. It's not the average nor a halfway point, but a kind of center that balances high numbers with low numbers (Campbell and Machin, 1993).

The mode is the most frequently occurring number in a list of numbers. It is the closest thing to what people mean when they say something is average or typical. It will be a category when the data are nominal or qualitative. The mode represents frequency by using percentage or proportion.

In data analysis, the purpose of statistically computing a measure of dispersion is to discover the extent to which scores differ, cluster, or spread from around a measure of central tendency. The most commonly used measure of dispersion is the standard deviation. This variance is calculated by subtracting the mean from each number,

squaring it, and dividing the grand total (Sum of squares) by how many numbers there

are. The square root of the variance is the standard deviation.

In this protocol, the analysis for each of the three questions are summarized

below.

Question 1 ; What is the incidence and severity of tissue necrosis in victims after *N.*

kaouthia or *C. rhodostoma* bites in Thailand ?

Variables		Data analysis		
		Type of data	*Central tendency*	*Deviation*
The clinical manifestations	Pulmonary system	Ratio scale	Mean	SD /√N, 95 % CI
	Cardiovascular system	Ratio scale	Mean	SD /√N, 95 % CI
	Central nervous system	Ratio scale	Mean	SD / √N, 95 % CI
	Hematological system	Ratio scale	Mean	SD /√N, 95 % CI
	Gastrointestinal system	Ratio scale	Mean	SD /√N, 95 % CI
	Local wound	Ratio scale	Mean	SD /√N, 95 % CI
	Total score	Ratio scale	Mean	SD /√N, 95 % CI

P : Proportion ; N : Number of patients ; CI : 95 %Confidential interval ; SD : Standard

deviation

Also, the percentage of each clinical manifestation in an organ system (e.g., pulmonary, cardiovascular, central nervous, hematological and gastrointestinal system) will be tabulated.

Tissue necrosis : Local wound clinical manifestations will be used to detemined the incidence and severity of tissue necrosis. All cases of tissue necrosis will be considered, if the victims as minimal, moderate or severe of tissue necrosis.

Question 2. What are the factors effecting tissue necrosis in patients after *N. kaouthia or*

C. rhodostoma bites in Thailand ?

Variables		Data analysis		
		Type of data	*Central tendency*	*Deviation*
Demographic data	Gender	Categorical	Percentage, Proportion	$P(1-P)/\sqrt{N}$, 95 % CI
	Age	Continuos	Mean	$SD/\sqrt{N}$, 95 % CI
	Education	Categorical	Percentage, Proportion	$P(1-P)/\sqrt{N}$, 95 % CI
	Salary	Continuous	Mean	$SD/\sqrt{N}$, 95 % CI
	Occupation	Categorical	Percentage, Proportion	$P(1-P)/\sqrt{N}$, 95 % CI
Geographic data	The position of place (province , urban , rural)	Categorical	Percentage, Proportion	$P(1-P)/\sqrt{N}$, 95 % CI
	Place of biting (indoor, outdoor	Categorical	Percentage, Proportion	$P(1-P)/\sqrt{N}$, 95 % CI
Factors related to snakebites	Season	Categorical	Percentage, Proportion	$P(1-P)/\sqrt{N}$, 95 % CI

Variables		Data Analysis		
		Type of data	*Central tendency*	*Deviation*
	Predisposing factors (barefoot, darkness)	Categorical	Percentage, Proportion	SD/√N, 95 % CI
	Site of biting	Categorical	Percentage Proportion	P(1- P)/√N, 95 % CI
	Time of biting	Continuous	Mean	SD/√N, 95 % CI
	The distance between fang marks	Continuous	Mean	SD/√N, 95 % CI
Factors related to treatment	Duration between bite and seeking medical advice	Continuous	Mean	SD/√N, 95 % CI
	Duration of hospitalization	Continuos	Mean	SD/√N, 95 % CI
	Prognosis (dressing, debridgement, amputation)	Categorical	Percentage Proportion	P(1- P)/√N, 95 % CI
	First aid treatment (bandage, no bandage)	Categorical	Percentage Proportion	P(1- P)/√N, 95 % CI
	The number of antivenom used	Continuos	Mean	SD/√N, 95 % CI

Question 3 :   What is the difference of incidence, associated factors affecting tissue necrosis and clinical consequences in patient after culprit snake bites both in prospective and retrospective study ?

Variables		Data analysis		
		Type of data	*Central tendency*	*Deviation*
Demographic data	Gender	Categorical	Percentage, Proportion	P(1- P)/√N, 95 % CI
	Age	Continuous	Mean	SD/√N, 95 % CI
	Education	Categorical	Percentage, Proportion	P(1- P)/√N, 95 % CI
	Salary	Continuous	Mean	SD/√N, 95 % CI
	Occupation	Categorical	Percentage, Proportion	P(1- P)/√N, 95 % CI
Geographic data	The position of place (province , urban , rural)	Categorical	Percentage, Proportion	P(1- P)/√N, 95 % CI
	Place of biting (indoor, outdoor	Categorical	Percentage, Proportion	P(1- P)/√N, 95 % CI
Factors related to snakebites	Season	Categorical	Percentage, Proportion	P(1- P)/√N, 95 % CI

Variables		Data Analysis		
		Type of data	*Central tendency*	*Deviation*
	Predisposing factors (barefoot, darkness)	Categorical	Percentage, Proportion	SD/√N, 95 % CI
	Site of biting	Categorical	Percentage Proportion	P(1- P)/√N, 95 % CI
	Time of biting	Continuous	Mean	SD/√N, 95 % CI
	The distance between fang marks	Continuous	Mean	SD/√N, 95 % CI
Factors related to treatment	Duration between bite and seeking medical advice	Continuous	Mean	SD/√N, 95 % CI
	Duration of hospitalization	Continuos	Mean	SD/√N, 95 % CI
	Prognosis (dressing, debridgement, amputation)	Categorical	Percentage Proportion	P(1- P)/√N, 95 % CI
	First aid treatment (bandage , no bandage)	Categorical	Percentage Proportion	P(1- P)/√N, 95 % CI
	The number of antivenom used	Continuos	Mean	SD/√N, 95 % CI

Variables		Data analysis		
		Type of data	*Central tendency*	*Deviation*
The clinical manifestations	Pulmonary system	Ratio scale	Mean	SD /√N, 95 % CI
	Cardiovascular system	Ratio scale	Mean	SD /√N, 95 % CI
	Central nervous system	Ratio scale	Mean	SD / √N, 95 % CI
	Hematological system	Ratio scale	Mean	SD /√N, 95 % CI
	Gastrointestinal system	Ratio scale	Mean	SD /√N, 95 % CI
	Local wound	Ratio scale	Mean	SD /√N, 95 % CI
	Total score	Ratio scale	Mean	SD /√N, 95 % CI

Question 4 : Can enzyme inhibitors prevent or reduce tissue necrosis after

N. kaouthia or *C. rhodostoma* bites?

Variables		Data analysis		
		Type of data	*Central tendency*	*Deviation*
In vivtro experiments	Protease activity (% Inhibition)	Continuous	Mean	SD/√N, 95 % CI
	Phospholipase A$_2$ activity (% Inhibition)	Continuous	Mean	SD/√N, 95 % CI
In vivo experiments	Hemorrhage activity (diameter spot)	Continuous	Mean	SD/√N, 95 % CI
	Edema(weight)	Continuous	Mean	SD/√N, 95 % CI
	Myonecrosis (creatine phosphokinase activity)	Continuous	Mean	SD/√N, 95 % CI
	Histological study	Description		

4. Statistical test

In the first question : the incidence of tissue necrosis among victims bitten by *C. rhodostoma or N. kaouthia*, will be summarized using percent + SEM.

In the second question : the associated factors related to tissue necrosis after snakebites, multiple logistic regression statistic will be used. This is the most versatile of statistical methods and can be used in many situations. Multiple logistic regression is used to account for (predict) more than one independent variables affecting outcome.

Final question, the effectiveness of chemical substances or drugs will be examined in animal model experiment. In animal experiments, treatment values of all inhibitors will be compared to values from the group of animals injected with venom alone, using student t-test for statistic comparison. Student t-test is typically used to compare the means of two populations, is useful for deciding if some treatments worked when compared to the control group.

5. Interpretation

Calculation of the snakebite severity score (SSS) involves the evaluation of six areas the experienced investigators used in the evaluation of snakebite. The score total ranges from 0 to 20 points. The sensitivity and specificity of a change in the SSS in the detection of clinical significance worsening were a change of 1 point (Dart et al.,1996).

Statistical significance is an expression of the results which did not occur by luck or chance. Most scientists would not label their results statistically significant at anything lower than 95 percent. The "95" of a 95% confidence interval (CI) means that the estimation procedure has a 0.95 probability of producing an interval containing the true population value if the study it repeated numerous times.

6. Problem cases

In some cases , envenomation by *N. kaouthia* or *C. rhodostoma* may result in death before patient reaches hospitals or health care centers. These cases will be excluded because tissue necrosis usually is evident only 24 hours following bites and therefore the incidence and the severity of tissue necrosis cannot be determined. However, the number of deaths and their characteristics will be documented and compared to those included in the study.