

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ



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FACULTY OF HEALTH SCIENCES
DEPARTMENT OF MEDICAL LABORATORY



Assessment of Liver Enzymes Among Cigarette Male Smokers (ALT – AST) in River Nile State 2018

*Supplementary research for attainment Bakalerious and privilege
degree in medical laboratory department*

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Introduction & Literature review

1.1 Introduction:-

Cigarette smoking is a major cause of preventable morbidity and mortality. Worldwide, more than 3 million people currently die each year from cigarette smoking ⁽¹⁾.

The risk of death in the smokers measured by the number of cigarettes smoked daily, the duration of smoking, the degree of inhalation and the age of initiation ⁽²⁾.

Cigarette smoke contains over 4000 different chemicals, 400 of which are proven to be carcinogenic; it also contains various oxidants such as oxygen free radicals and volatile aldehydes which are probably the major causes of damage to biomolecules ⁽³⁾.

1.1.1 The liver:-

The liver is a vital organ of vertebrates and some other animals. In the human it is located in the upper right quadrant of the abdomen, below the diaphragm. The liver has a wide range of functions, including detoxification of various metabolites, protein synthesis, and the production of bio chemicals necessary for digestion.

The liver is a gland and plays a major role in metabolism with numerous functions in the human body, including regulation of glycogen storage.

1.1.1.1 Liver function tests:-

1-Markers for hepato cellular necrosis (ALT; most specific for hepatocyte injury, AST ;less specific than ALT significant presence in other tissues, LD least specificant significant presence in other tissues).

2-Marker that reflect cholestasis(Alkaline phosphatase ,Gamma- glutamyl transferase).

3-tests to assess liver disorders(Total bilirubin , direct bilirubin (conjugated), indirect bilirubin (unconjugated) , Albumin, Ammonia, Alph fetoprotein)⁽⁴⁾.

1.1.1.2 Liver enzymes:-

I. Aspartate transaminase Enzyme code (EC2.6.1.1) Aspartate transaminase (AST) or aspartate amino transferase, also known as AspAT/ASAT/AAT or serum glutamic oxaloacetic transaminase (SGOT).

Reference range 5 to 30 U/L (37C) ⁽⁵⁾.

II. Alanine transaminase (EC2.6.1.2), Alanine transaminase (ALT) is a transaminase enzyme. It is also called alanine amino transferase (ALAT) and was formerly called serum glutamate-pyruvate transaminase (SGPT) or serum glutamic -pyruvic transaminase (SGPT).

Reference range of ALT 6-37 U/L (37C) ⁽⁵⁾.

1.2 literature review:-

1.2.1 Physiology of smoking:-

The active substance in tobacco, especially cigarettes, is administered by burning the leaves and inhaling the vaporized gas that results. This quickly and effectively delivers substances into the blood stream by absorption through the alveoli in the lung ⁽⁶⁾.

1.2.2. Effect of smoking in health:-

Smokers are more likely than non-smokers to develop heart disease , stroke , lung cancer and cardio vascular disease .

Smoking can cause lung disease by damaging airways and the small air sacs (alveoli).

Smoking harms nearly every organ of the body and effect a person's overall health ⁽⁶⁾.

1.3 Justification:

Smoking is a major cause of death and other chronic diseases. Worldwide This study was conducted to assess the activity of AST and ALT in Sudanese smokers and to find is there relationship between duration of smoking and number of cigarette smoke per day.

1.4 Objectives:-

1.4.1 General objective:-

To assess the effect of smoking on ALT and AST.

1.4.2 Specific objectives:-

1-To compare serum ALT and AST activities in male cigarette smokers in comparison to non-smokers.

2-To correlate between serums ALT and AST activities and duration of smoking, number of cigarettes smoked per day and age of smokers.

Materials and Methods

2.1. Materials:-

2.1.1. Study design:-

This is a cross sectional study.

2.1.2. Study area:-

The study was conducted in River Nile State.

2.1.3. Study population: -

Thirty smokers were enrolled as test group and thirty nonsmokers as control group were enrolled in this study according to inclusion and exclusion criteria.

2.1.3.1 Inclusion Criteria:-

Healthy looking cigarettes male Smokers in River Nile State.

2.1.3.2 Exclusion Criteria:-

Cigarettes male Smokers who have liver disease, alcohol abuse, bone disease, cardiac disease were excluded.

2.1.4 Samples:-

Samples were collected under aseptic conditions and placed in sterile plain containers, and after clotting centrifuged for 3 minutes at 3000 RPM to obtain serum, then obtained serum were kept at -20c till the time of analysis.

800 μ/l of reagent and 200 μ/l of buffer were taken with 10 μ/l sample mixed and incubate for one minute read in spectrophotometer.

2.1.5 Ethical consideration:-

Smokers who voluntarily accepted to participate in the study were included.

2.1.6. Equipments:-

Mindary instrument, Centrifuge, Sterile plain containers, Disposable syringes, 70% alcohol, Tourniquets, Cotton,

Micropipettes (automatic pipettes) and Graduated pipettes.

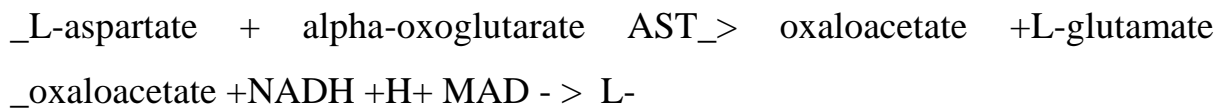
2.1.7. Data analysis: -

Data was analyzed using SPSS computer program.

2.2. Methods:

2.2.1. Estimation of serum aspartate aminotransferase:-

(appendix II) Principle of method:-



In the assay reactions, the AST catalyze the reversible transamination of L-aspartate and alpha-

oxoglutarate to oxaloacetate and L-

glutamate. The oxaloacetate is then reduced to malate in the presence of malate dehydrogenase with NADH

the rate of photometrically determined NADH decrease directly proportional to the rate of formation of oxaloacetate and thus the AST activity.

Calculation:

The analyzer calculates the activity of each sample automatically with specified valid calibration factor from calibration process Conversion factor of traditional unit (U/L) into SI-units (M Kat/L): $1 \text{ U/L} = 16.67 \times 10^{-3} \text{ M kat/L}$

$$1 \text{ M Kat /L} = 60 \text{ U/L}$$

Reference values:

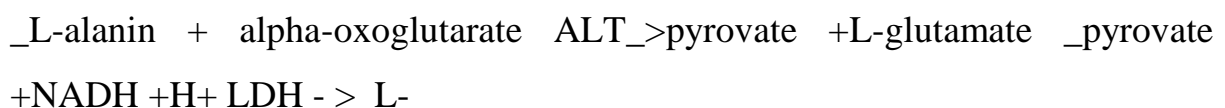
The reference intervals measured at 37 C

MALE : <35 U/L , < 0.58 M kat /L .

FEMALE: <31 U/L , < 0.52 M kat/L .

2.2.2. Estimation of serum alanin aminotransferase:

Principle of method:-



In the assay reactions, the ALT catalyze the reversible transamination of L-alanin and alpha-oxoglutarate to pyrovat and L-glutamate -

The pyrovate is then redused to lactate in the presence of lactate dehydrogenase

with the concurrent oxidation of reduced B-nicotinamid adenine dinucleotide (NADH) to B-nicotin amid adenine dinucleotide (NAD) - This change in absorbance is directly proportional to the activity of ALT in the sample.

Results

Table (3.1) Distribution of study group according to age

Age	Frequency	Percent
25 – 35 years	7	23.3%
36 – 45 years	9	30.0%
46 – 55 years	8	26.7%
> 55 years	6	20.0%
Total	30	100%

Mean	3.43
Std. Error of Mean	0.196
Std. Deviation	1.073

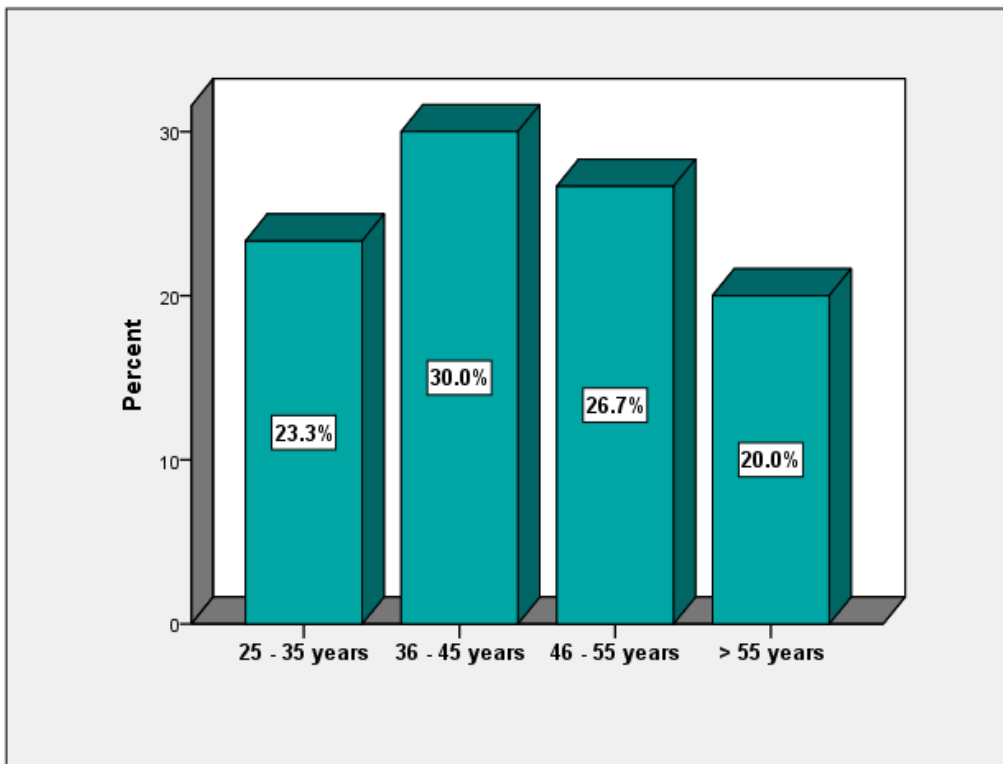


Figure (3.1) Distribution of study group according to age

Table (3.2) Distribution of study group according to duration of smoking

Duration of smoking	Frequency	Percent
< 10 years	14	46.7%
10 - 20 years	9	30.0%
21 - 30 years	4	13.3%
> 30 years	3	10.0%
Total	30	100%

Mean	1.87
Std. Error of Mean	0.184
Std. Deviation	1.008

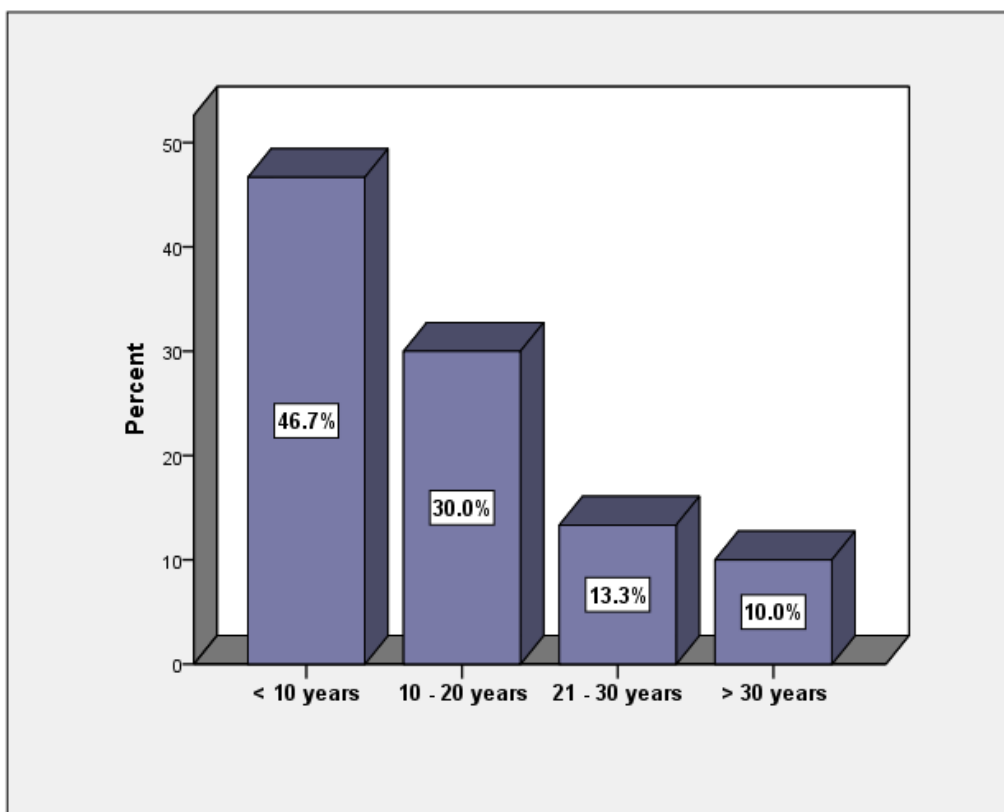


Figure (3.2) Distribution of study group according to duration of smoking

Table (3.3) Distribution of study group according to number of cigarette smoke per day

Number of cigarette/ day	Frequency	Percent
< 5 cigarettes	1	3.3%
5 - 10 cigarettes	20	66.7%
11 - 15 cigarettes	3	10.0%
16 - 20 cigarettes	6	20.0%
Total	30	100%

Mean	2.47
Std. Error of Mean	0.157
Std. Deviation	0.860

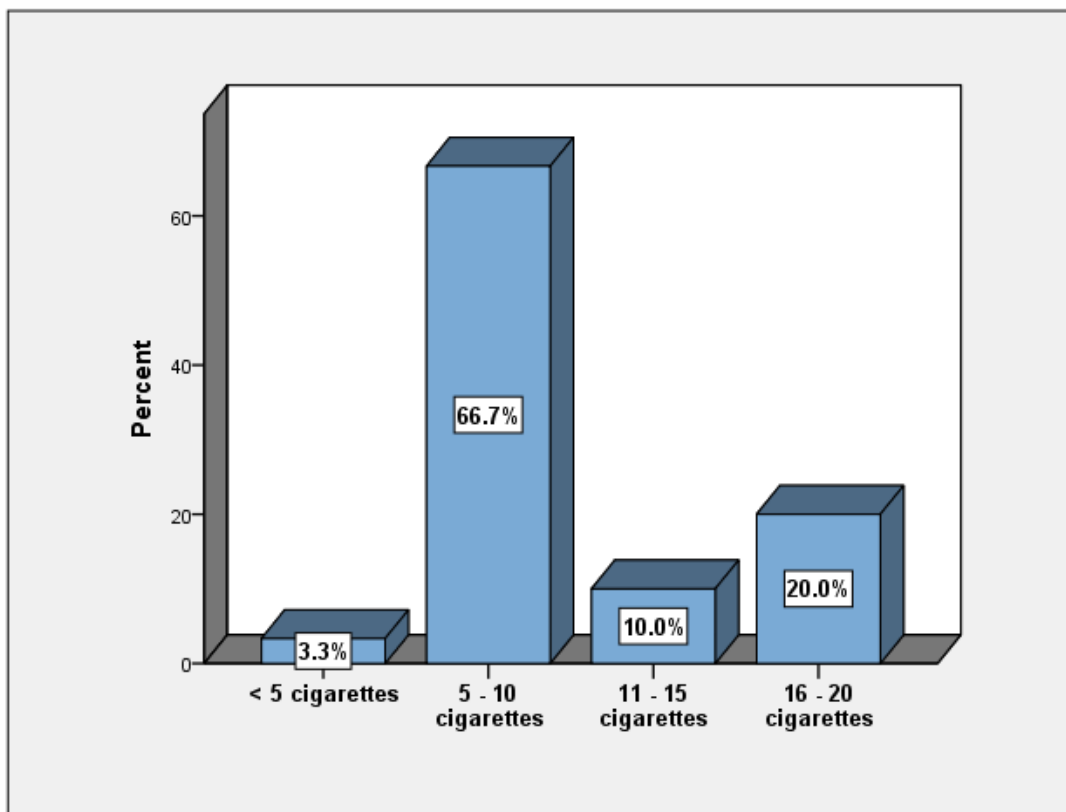


Figure (3.3) Distribution of study group according to number of cigarette smoke per day

Table (3.4) Distribution of study group according to type of smoking

Type of smoking	Frequency	Percent
Cigarette	30	100%
Others	0	0%
Total	30	100%

Mean	1.00
Std. Error of Mean	0.000
Std. Deviation	0.000

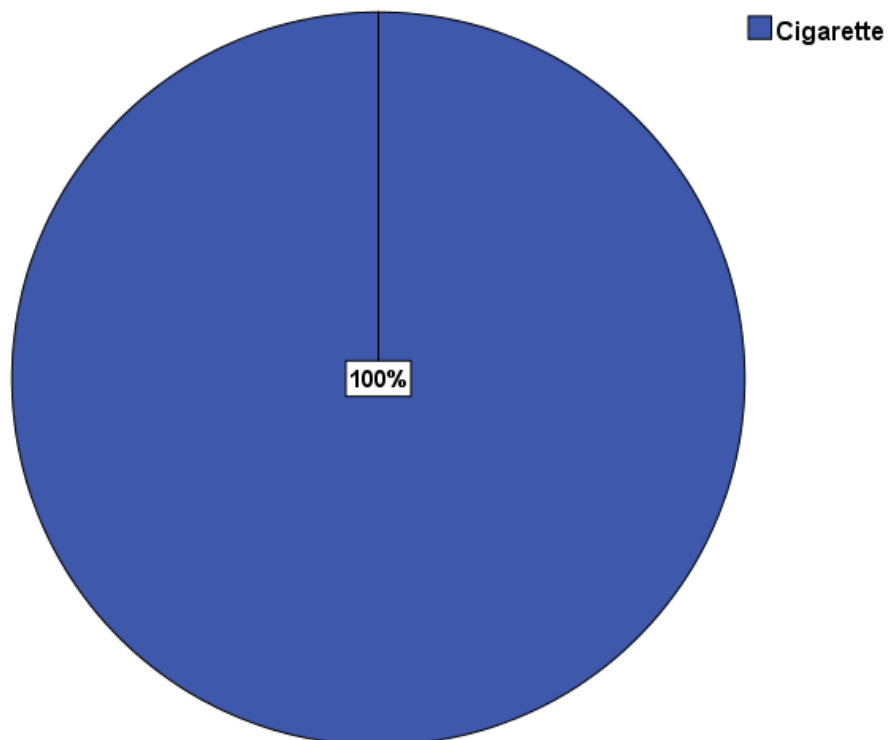


Figure (3.4) Distribution of study group according to type of smoking

Table (3.5) Distribution of study group according to other hereditary disease among study group

Other disease	Frequency	Percent
Yes	0	0%
No	30	100%
Total	30	100%

Mean	1.00
Std. Error of Mean	0.000
Std. Deviation	0.000

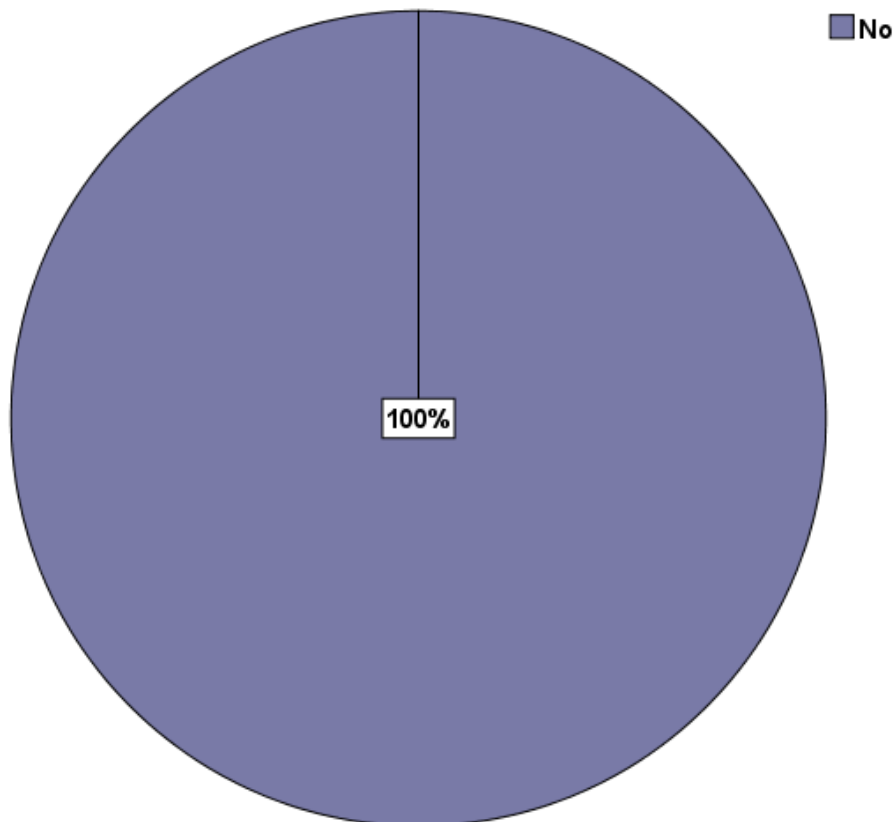


Figure (3.5) Distribution of study group according to other hereditary disease among study group

Table (3.6) Distribution of study group according to AST

AST	Frequency	Percent
10 - 20	12	40.0%
21 - 30	14	46.7%
31 -40	4	13.3%
Total	30	100%

Mean	1.73
Std. Error of Mean	0.126
Std. Deviation	0.691

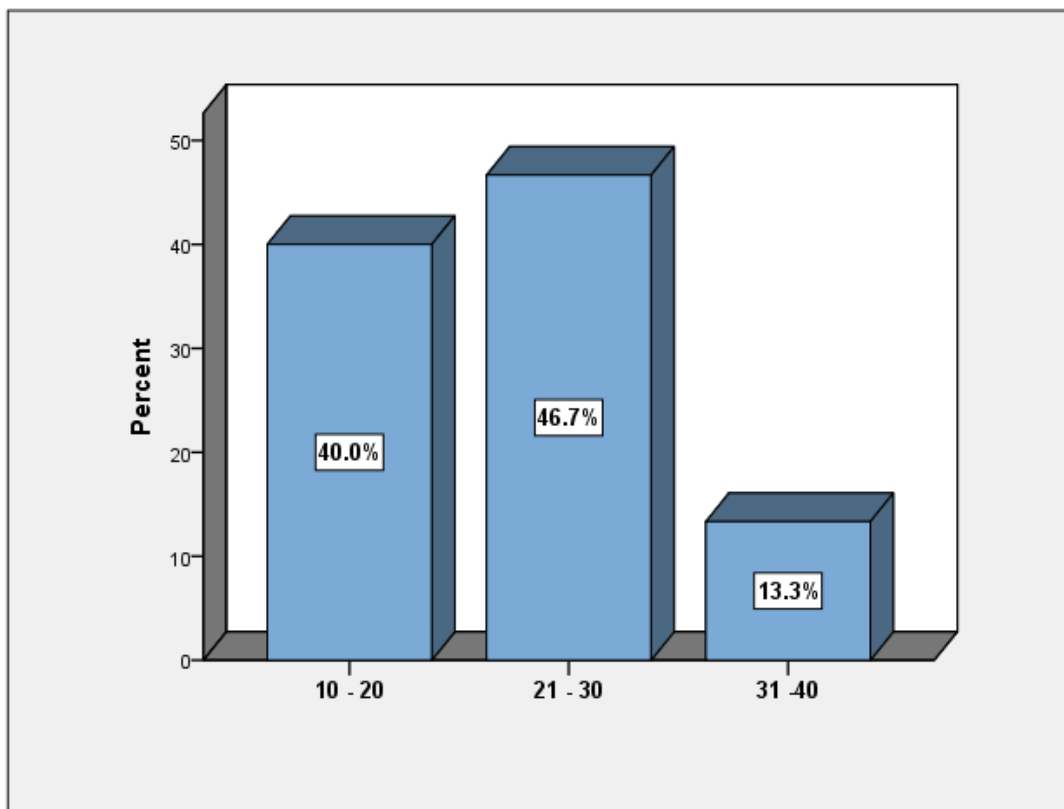


Figure (3.6) Distribution of study group according to AST

Table (3.7) Distribution of study group according to ALT

ALT	Frequency	Percent
< 10	10	33.3%
10 - 20	16	53.3%
21 - 30	2	6.7%
31 -40	2	6.7%
Total	30	100%

Mean	1.87
Std. Error of Mean	1.50
Std. Deviation	0.819

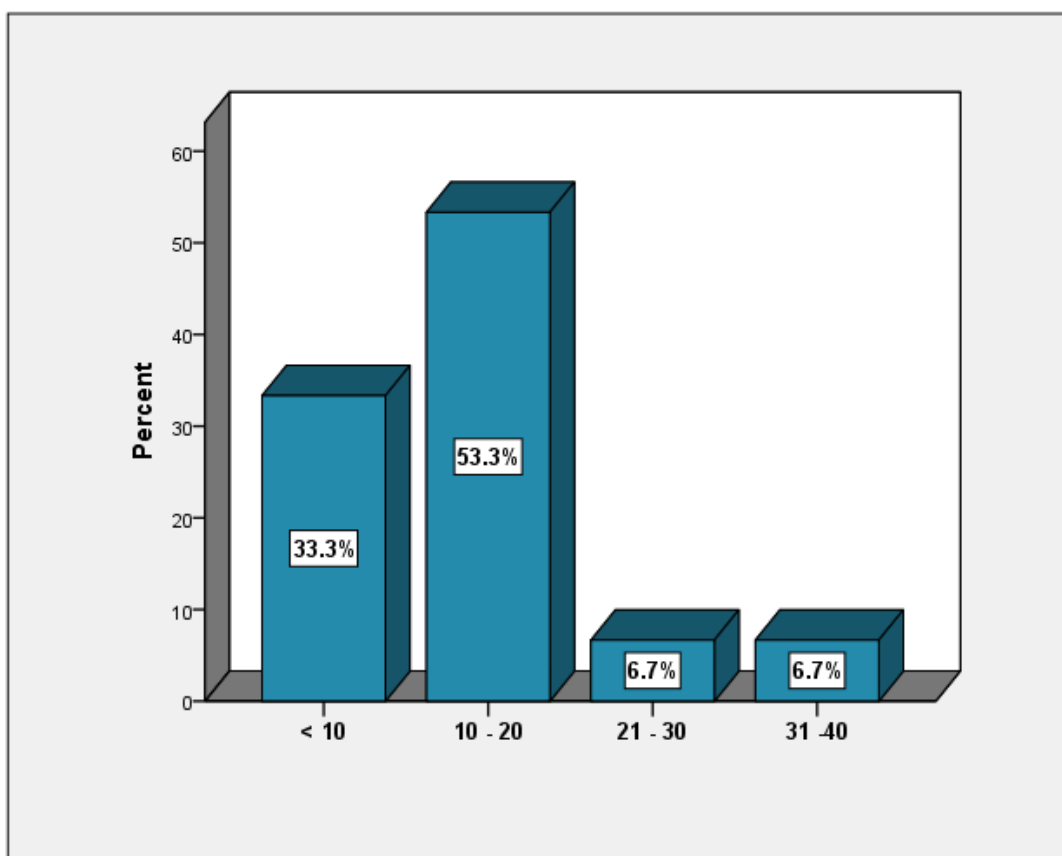


Figure (3.7) Distribution of study group according to ALT

Cross tabulation between AST and age group

		Age				Total
		25 - 35 years	36 - 45 years	46 - 55 years	> 55 years	
AST	10 - 20	1	4	4	3	12
	21 - 30	5	4	2	3	14
	31 -40	1	1	2	0	4
Total		7	9	8	6	30

Symmetric Measures

		Value	Asymp. Std. Error ^a	Approx. T ^b	Approx. Sig.
Nominal by Nominal	Phi	.406			.550
	Cramer's V	.287			.550
	Contingency Coefficient	.376			.550
Interval by Interval	Pearson's R	-.211-	.152	-1.141-	.264 ^c
Ordinal by Ordinal	Spearman Correlation	-.223-	.159	-1.209-	.237 ^c
N of Valid Cases		30			

P. Value = 0.0055

Cross tabulation between AST and duration of smoking

	Duration of smoking				Total
	< 10 years	10 - 20 years	21 - 30 years	> 30 years	
10 - 20	6	2	2	2	12
AST 21 - 30	6	5	2	1	14
31 -40	2	2	0	0	4
Total	14	9	4	3	30

Symmetric Measures

		Value	Asymp. Std. Error ^a	Approx. T ^b	Approx. Sig.
Nominal by Nominal	Phi	.325			.787
	Cramer's V	.230			.787
	Contingency Coefficient	.309			.787
Interval by Interval	Pearson's R	-.152-	.160	-.812-	.424 ^c
Ordinal by Ordinal	Spearman Correlation	-.083-	.181	-.442-	.662 ^c
N of Valid Cases		30			

P. Value = 0.0078

Cross tabulation between AST and duration of number of cigarette/ day

		No of cigarette/ day				Total
		< 5 cigarettes	5 - 10 cigarettes	11 - 15 cigarettes	16 - 20 cigarettes	
AST	10 - 20	1	6	1	4	12
	21 - 30	0	10	2	2	14
	31 -40	0	4	0	0	4
Total		1	20	3	6	30

Symmetric Measures

		Value	Asymp. Std. Error ^a	Approx. T ^b	Approx. Sig.
Nominal by Nominal	Phi	.428			.481
	Cramer's V	.303			.481
	Contingency Coefficient	.394			.481
Interval by Interval	Pearson's R	-.247-	.155	-1.351-	.188 ^c
Ordinal by Ordinal	Spearman Correlation	-.213-	.178	-1.156-	.258 ^c
N of Valid Cases		30			

P. Value = 0.0048

Cross tabulation between ALT and age group

	Age				Total
	25 - 35 years	36 - 45 years	46 - 55 years	> 55 years	
< 10	3	2	3	2	10
10 - 20	4	6	3	3	16
21 - 30	0	1	0	1	2
31 - 40	0	0	2	0	2
Total	7	9	8	6	30

Symmetric Measures

		Value	Asymp. Std. Error ^a	Approx. T ^b	Approx. Sig.
Nominal by Nominal	Phi	.546			.443
	Cramer's V	.315			.443
	Contingency Coefficient	.479			.443
Interval by Interval	Pearson's R	.146	.145	.784	.440 ^c
Ordinal by Ordinal	Spearman Correlation	.112	.177	.596	.556 ^c
N of Valid Cases		30			

P. Value = 0.00443

Cross tabulation between ALT and duration of smoking

		Duration				Total
		< 10 years	10 - 20 years	21 - 30 years	> 30 years	
ALT	< 10	3	4	1	2	10
	10 - 20	10	2	3	1	16
	21 - 30	0	2	0	0	2
	31 -40	1	1	0	0	2
Total		14	9	4	3	30

Symmetric Measures

		Value	Asymp. Std. Error ^a	Approx. T ^b	Approx. Sig.
Nominal by Nominal	Phi	.591			.313
	Cramer's V	.341			.313
	Contingency Coefficient	.509			.313
Interval by Interval	Pearson's R	-.189-	.133	-1.020-	.316 ^c
Ordinal by Ordinal	Spearman Correlation	-.179-	.163	-.962-	.344 ^c
N of Valid Cases		30			

P. Value = 0.0031

Cross tabulation between ALT and duration of number of cigarette/ day

	sigday				Total
	< 5 cigarettes	5 - 10 cigarettes	11 - 15 cigarettes	16 - 20 cigarettes	
< 10	1	5	0	4	10
10 - 20	0	11	3	2	16
21 - 30	0	2	0	0	2
31 -40	0	2	0	0	2
Total	1	20	3	6	30

Symmetric Measures

		Value	Asymp. Std. Error ^a	Approx. T ^b	Approx. Sig.
Nominal by Nominal	Phi	.547			.440
	Cramer's V	.316			.440
	Contingency Coefficient	.480			.440
Interval by Interval	Pearson's R	-.251-	.144	-1.373-	.181 ^c
Ordinal by Ordinal	Spearman Correlation	-.199-	.189	-1.073-	.293 ^c
N of Valid Cases		30			

P. Value = 0.0044

Cross tabulation between AST Case and AST Control :

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)	Point Probability
Pearson Chi-Square	2.766 ^a	4	.598	.647		
Likelihood Ratio	4.098	4	.393	.578		
Fisher's Exact Test	2.986			.647		
Linear-by-Linear Association	1.265 ^b	1	.261	.335	.170	.070
N of Valid Cases	30					

a. 8 cells (88.9%) have expected count less than 5. The minimum expected count is 1.20.

b. The standardized statistic is -1.125-.

Symmetric Measures

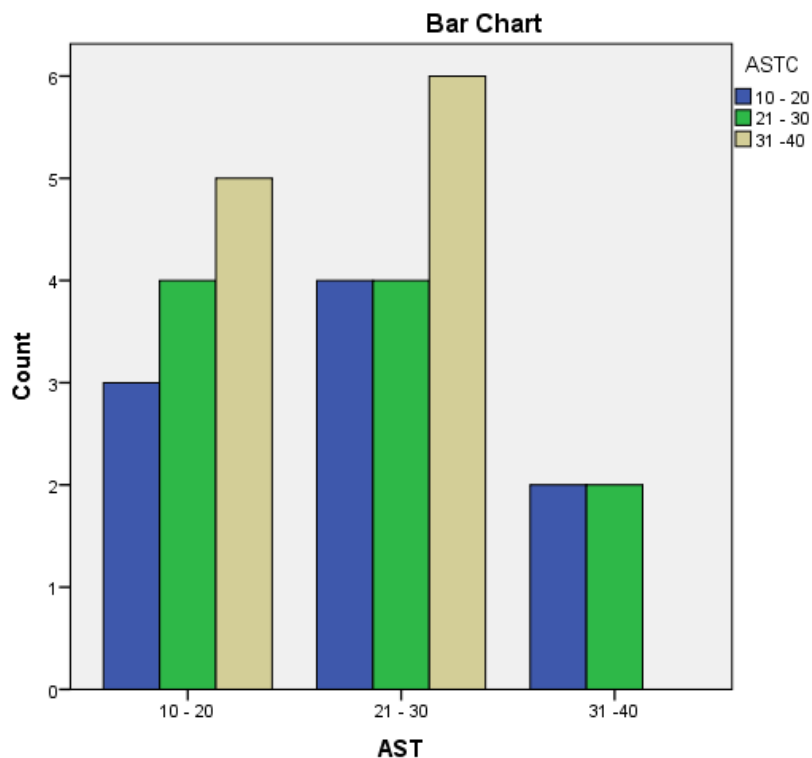
		Value	Asymp. Std. Error ^a	Approx. T ^b	Approx. Sig.	Exact Sig.
Nominal by Nominal	Phi	.304			.598	.647
	Cramer's V	.215			.598	.647
	Contingency Coefficient	.291			.598	.647
Interval by Interval	Pearson's R	-.209-	.162	-1.130-	.268 ^c	.335
Ordinal by Ordinal	Spearman Correlation	-.188-	.170	-1.012-	.320 ^c	.320
N of Valid Cases		30				

a. Not assuming the null hypothesis.

b. Using the asymptotic standard error assuming the null hypothesis.

c. Based on normal approximation.

P. Value = 0.0170



Cross tabulation between ALT Case and ALT Control :

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)	Point Probability
Pearson Chi-Square	3.928 ^a	6	.686	.668		
Likelihood Ratio	5.112	6	.529	.640		
Fisher's Exact Test	4.429			.699		
Linear-by-Linear Association	.341 ^b	1	.560	.589	.333	.112
N of Valid Cases	30					

a. 10 cells (83.3%) have expected count less than 5. The minimum expected count is .13.

b. The standardized statistic is .584.

Symmetric Measures

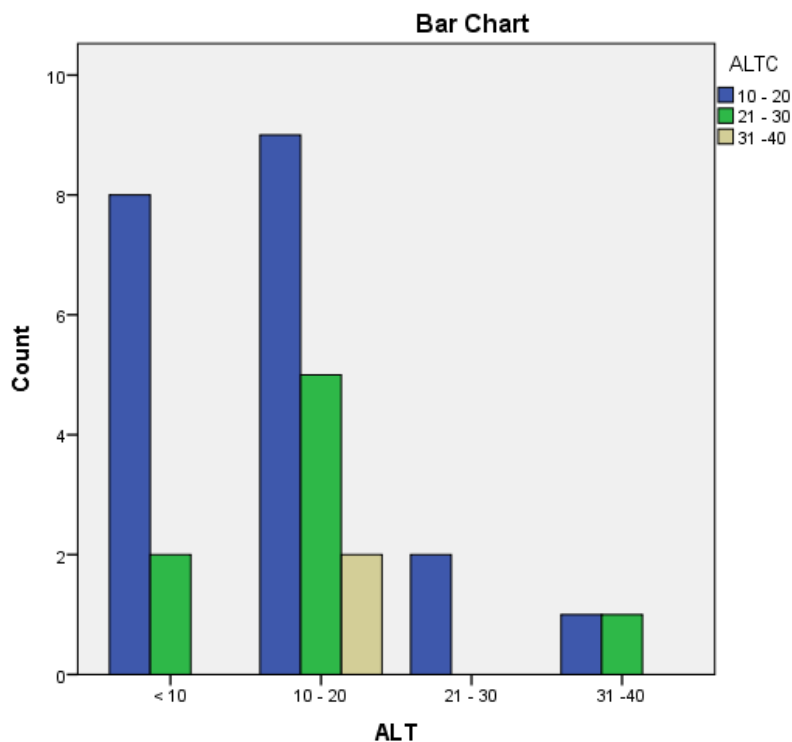
	Value	Asymp. Std. Error ^a	Approx. T ^b	Approx. Sig.	Exact Sig.
Nominal by Nominal	Phi	.362		.686	.668
	Cramer's V	.256		.686	.668
	Contingency Coefficient	.340		.686	.668
Interval by Interval	Pearson's R	.108	.139	.577	.569 ^c
Ordinal by Ordinal	Spearman Correlation	.143	.162	.766	.450 ^c
N of Valid Cases	30				

a. Not assuming the null hypothesis.

b. Using the asymptotic standard error assuming the null hypothesis.

c. Based on normal approximation.

P. Value = 0.0162



4.1 Discussion :-

Nicotine is the major component of cigarette smoke plays an important role in the development of many diseases . it causes oxidative damage to kidney , lung ,liver and heart. It is a potential oxidant , which is capable of producing free radical and reactive oxygen species.

The nicotine induce free radical to react bio membrane causing oxidative destruction of poly unsaturated fatty acid and forming cytotoxic aldehydes by lipid per-oxidation implicated in pathogenesis of number of disease.

This is across sectional study aimed to study the effect of smoking on liver enzymes activities .60 males (30 smokers and 30 nonsmokers) were enrolled in this study to study the effect of smoking on liver enzymes . After evaluation of enzymes activities by auto analyzer , the statistical analysis was done by using SPSS computer program and the results showed that all liver enzyme levels was insignificantly in smoker group when compared to non-smoker group ,not increase in activity of AST& ALT in smokers indicate tissue damage due to loss of functional integrity of cell membrane.

This result is disagree with results of study conducted in El-beidia city ,Libya done by Alsalhen to show the effect of cigarette smoking on liver function among smokers and non-smokers male , showed that the smokers had significantly higher than the non-smokers in AST , ALT and ALP activities do. Result of this study revealed that increases in AST , ALT and GGT is proportional which duration of smoking per years , also the serum ALT,AST and ALP activates GGT activities are a significant positive correlation with age , and there were no correlation between ALP with age . The serum AST , ALT , ALP and GGT activities are a significant positive correlation with duration of smoking . The serum AST , ALT , ALP activaties are significantly positive correlation with number of cigarettes and There were no correlation between GGT activaties and number of cigarettes per day⁽⁷⁾.

4.2 conclusion :-

The result of study serum AST and ALT are insignificant in smoker compare to non-smoker group . And the mean of serum AST and ALT activities are insignificant correlation with the age , insignificant correlation with duration of smoking ,and insignificant correlation with number of cigarette per day.

4.3 Recommendation :-

health program can be designed to awareness about several side effect of smoking . other study studies are suspected the increase in ALP activity in current smokers bone , needed to do further studies bone biomarker to reflect of smoking on bone . Require more sample size to get confidence result.

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**ASSEMENT OF LIVER ENZYMES AMONG SMOKER IN RIVER
NILE STATE IN 2018**

Questionnaire

Name :..... No of sample ().

Age :.....

Duration of smoking / years :.....

Number of cigarettes /day :

History of other diseases :

Results :

AST activity :u/l

ALT activity :u/