

## Urinary glycosaminoglycan levels in healthy adolescents between 17 – 19 years age

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### Abstract

**Introduction:** Any laboratory results become medically useful only when appropriate comparative data from healthy population is available. In absence laboratory results may not provide valuable information. Age group associated urinary excretion of Glycosaminoglycans(GAGs) are one such parameter where available comparative data is not sufficient. Hence in the present study urinary GAG excretion in a healthy adolescent of 17 to 19 years age group was performed.

**Aims & Objectives:** To determine urinary excretion levels of GAGs among healthy adolescent of 17 to 19 years age group.

**Materials and Method:** Random urine samples from 118 healthy, engineer students between 17-19yrs age were estimated for excretion of GAGs by Dimethyl-methylene blue method.

**Results and Conclusion:** Urinary GAGs level among adolescent male was in the range of 8.4 to 12.4 mg dl and in females it was 8.2 to 12 mg dl. The results of our study, suggest 8.2 to 12.4 mg dl as normal reference range for Urinary Glycosaminoglycans excretion in healthy, adolescent 17 to 19 years age group by DMMB dye method.

**Keywords:** Urinary Glycosaminoglycans, Dimethyl methylene blue, Reference value, Urine.

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### Introduction

Any laboratory results on patient's samples become medically useful only when appropriate comparative data from healthy population is available. Reference data available in books are mainly from European population and available in literature are performed on a specific ethnic group or particular geographic location or under certain experimental conditions. The same results may not hold good for population from diverse ethnic group, living under different habitat and having varied genetic makeup; meaning reference data is likely to differ among age, sex, ethnic group, genetic makeup and many other demographic variables. Also many times ranges set by kit manufacturer or standard books are adopted for interpretation of clinical condition which may not be ideal for all. Thus in absence of proper reference data reported laboratory results may not provide valuable information on the clinical condition of individual. Glycosaminoglycans(GAGs) are one such parameter where available comparative data is not sufficient to match with different age groups, gender classification, ethnic and geographical location. Normal GAGs excretion reported in infants (up to one year of age) is 18 to 40 mg/mmol of creatinine and as age advances it decreases to 3 to 8 mg/mmol of creatinine by the end of 10 years and then it remains constant thereafter.<sup>(1)</sup> As urinary GAGs excretion is emerging as early marker for renal damage<sup>(2)</sup> and importance is noted in various disease like diabetes, cardiovascular disease, Urolithiasis, systemic lupus erythematosus, hypertension.<sup>(3,4)</sup> As there is scarcity report on age wise excretion of GAGs the present study was taken up to have comparative reference value for urinary GAG

excretion in a healthy adolescent of 17 to 19 years age group.

### Aim and Objectives

To determine urinary excretion levels of GAGs among healthy adolescent of 17 to 19 years age group.

### Materials and Method

The present study was non interventional, cross sectional, analytical study performed on group of healthy 118 engineering student from 17 to 19yrs of age group by convenient sampling method at department of biochemistry of a medical college in North West Karnataka (India). All the students who visited SDM College of Medical science hospital for their routine health checkup were selected as participants. Informed consent was obtained from each individual after explaining the importance of study. The study was approved by Institutional Ethical Committee.

**Urine Sample Collection:** Random, mid stream urine sample was collected in sterile container from each participant for estimation of urinary GAGs levels. Urine sample was analyzed within 3-4hours on the same day without centrifugation. Sample containing abnormal constituents were rejected for analysis.

**Analytical Method:** Urine Glycosaminoglycans was estimated by Dimethyl methylene blue (DMMB) method.<sup>(5)</sup> DMMB was obtained from Aldrich. Chondroitin 4 sulphate purchased from Himedia Company was used for standard. Estimation was based on principle where DMMB a thiazine chromotrope binding to sulphated GAGs at pH 3 producing purple colour and the absorbance is measured at 525nm.

**Standard Control:** Chondroitin sulphate (50mg%) prepared as standard was used to prepare standard curve with concentration ranging from 2.5 to 15 mg%. Results were analyzed by SPSS ver.22, Students t test was used to compare the data. Results were expressed as Range and mean  $\pm$  SD.

## Results

Among 118 healthy, adolescent engineering students participated in present study 64 (55%) were male and 54 (45%) were female (Table 1). Mean age of male 17.9 yrs and female was 18.2yr whereas overall mean age 18.05yrs.

Urinary GAGs level among adolescent male was in the range of 8.4 to 12.4 mg/dl and in females it was 8.2 to 12 mg/dl. The observed GAGs levels in 17 to 19 years age group ranged between 2.0 to 20.0 mg/dl (refer Table 2 and 3).

**Table 1: Gender distribution of study population. (n=118)**

Subjects	Number	Percentage	P value
Male	64	55%	
Female	54	45%	
Total	118	100%	NS

**Table 2: Excretion of Urinary GAGs (Values expressed in Mean  $\pm$  SD)**

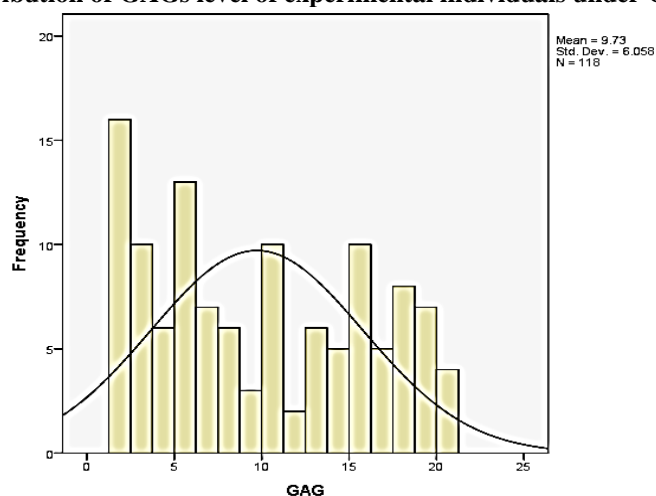
Participants	GAGs (mg/dl) Mean $\pm$ SD	Range (mg/dl)
Male	10.4 $\pm$ 2	8.4 to 12.4
Female	10.1 $\pm$ 1.9	8.2 to 12.0
Total	9.73 $\pm$ 6.05	2.0 to 20.0

**Table 3: Descriptive Statistics by SPSS Ver. 22**

	N	Range	Minimum	Maximum	Mean		Std. Deviation	Variance
	Statistic	Statistic	Statistic	Statistic	Statistic	Std. Error	Statistic	Statistic
GAG	118	19	2	20	9.73	.558	6.058	36.699
Valid N (listwise)	118							

The values obtained were evenly distributed under Gaussian curve (refer Graph A) and well within 2SD. There was no significant difference observed when GAGs levels were compared between males and females participants.

**Graph A: Distribution of GAGs level of experimental individuals under Gaussian curve**



## Discussion

Glycosaminoglycans play an important role in permeability of glomerular basement membrane. Increased loss of GAGs from GBM leads to reduction of charge selectivity of GBM which is associated with various disorders affecting GBM.<sup>(6)</sup> Estimation of GAGs in urine is one of the important independent markers to assess renal function.<sup>(7)</sup>

Studies have shown that, in healthy individuals, even when the GAGs present in serum is low; amount of excretion in urine remains high. This suggests that the important source of urinary GAGs is from kidneys<sup>(7)</sup> and excretion is influenced by associated diseases.<sup>(3,4)</sup> Hence it is required to find the normal excretion of GAGs in the adolescents and establish a normal reference range for Urinary GAGs so that, any abnormal level can be correlated to identify or diagnosed a disease state at bud stage.

GAGs can be estimation by Alcian blue method.<sup>(8)</sup> In our study we estimated GAGs by Dimethyl methylene blue method which is simple and sensitive.

In our study urinary GAGs level among adolescent between 17 to 19 years male was 8.4 to 12.4 mg/dl and females was 8.2 to 12 mg/dl. There was no significant difference ( $p= 0.408$ ) was found between males and females. This is in contrast to study done by Michelacci YM<sup>(9)</sup> but the study was performed by Alcian Blue dye method, used 24hrs urine sample and it was performed on 2 to 14 years children and 18 to 63 years aged individuals.

### Conclusion

From the results of our study, suggested normal reference range of Urinary Glycosaminoglycans excretion in healthy, adolescent 17 to 19 years age group by DMMB dye method is 8.2 to 12.4 mg/dl.

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